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Toxic Metal (Pb, Hg and As) Contamination of Muscle, Gill and Liver Tissues of *Otolithes rubber*, *Pampus argenteus*, *Parastromateus niger, Scomberomorus commerson* and *Onchorynchus mykiss*

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Abstract: The Pb, Hg and As content of the muscle, gill and liver of marin organisms *Otolithes rubber*, *Pampus argenteus*, *Parastromateus niger, Scomberomorus commerson*, *Onchorynchus mykiss* were measured. Analysis of lead and arsenic was carried out by anodic stripping voltammetry method. Hg analysis was done spectrophotometrically. It was found that mercury concentration reaches to maximum in the gill of *Parastromateus niger* (0.47 µg/g) followed by the liver of *Otolithes rubber* (0.001 µg/g), lead content reaches to maximum in the gill of *Parastromateus niger* (0.11 µg/g), followed by the liver of *Scomberomorus commerson*, (0.005 µg/g), arsenic concentration is highest in the liver of *Otolithes rubber* (0.26 µg/g), followed by the muscle of *Pampus argenteus* (0.007 µg/g). The results of this study indicate that the concentration of Pb, Hg and As in the different tissues of the studied marine organisms were significantly lower than the permissible levels for these toxic metals.

Key words: Toxic metal analysis · Marine organism · Otolithes rubber · Pampus argenteus · Parastromateus niger · Scomberomorus commerson · Onchorynchus mykiss.

INTRODUCTION

The incorporation of toxic metal ions into the biological systems include significant health hazard [1, 2]. Toxic metals such as Pb, Hg and As, are biological contaminant of special concern due to a wide distribution in the environment and likely adverse effects for human health. The accumulation of toxic metals can have middle-term and long term health risks and strict periodic surveillance of these contaminants is therefore advisable [3, 4]. Fish tissues can contain toxic metals from their presence in the water [5]. Toxic metal pollution of the sea water is less visible and direct than other types of marine pollution but its effects on marine ecosystems and humans are intense and very extensive. The toxic effects of heavy metals, particularly arsenic, mercury and lead, have been broadly studied [1-5]. The accumulations of metals vary greatly between both the fish species and/or tissues. Generally fish translocate larger quantities of metals in gill to their liver than to their muscle [6]. At this time, the analysis of heavy metals in the different fish species received much attention [7-20]. The aim of the present study is monitoring the concentration of toxic metals such as Pb, Hg and As in different tissues of some marine organisms such as Otolithes rubber, Pampus argenteus, Parastromateus niger, Scomberomorus commerson, Onchorynchus mykiss. Arsenic and lead are analyzed by anodic stripping voltammetry and Hg is analyzed by spectrophotoimetric methods [21].

MATERIALS AND METHODS

Chemical and Reagents: All chemical reagents were of analytical reagent grade, purchased from Merck (Germany). All solutions were prepared with doubly distilled water. Stock standard solution of Pb(II), Hg(II) and As(II) (1000 mg/L) were prepared by dissolving the appropriate amount of metal salts/oxid dissolving in doubly distilled water and diluting to 1000 ml in the volumetric flask.

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Apparatus: All absorbance measurements were carried out on a Scinco's PDA UV-Vis. Spectrophotometer (photodiode array) equipped with 1.0-cm quartz cells. All voltammetric measurements were carried out using a polarographic processor, model 746 VA (Metrohm), in combination with a polarographic stand model, 747 VA (Metrohm). The electrode stand consist of a hanging mercury drop electrode (HMDE) as working electrode, a double junction Ag/AgCl (3M KCl, saturated AgCl and 3M KCl in the bridge) as reference electrode and platinum wire, with considerably larger surface area than that of HMDE, as auxiliary electrode. All potentials quoted are relative to Ag/AgCl reference electrode. Stirring was carried out by a large Teflon road with 2000 rpm speed. A 780 pH Meter (Metrohm), equipped with a combined Ag/AgCl glass electrode was used for pH measurements. Eppendorf reference variable micropipettes were used to pipette microlitre volume of solutions. All glassware was soaked over night in 10% (v/v) nitric acid, followed by washing with 10% (v/v) hydrochloric acid and rinsed with double distilled water and dried before using.

Sample Preparation: Although voltammetric techniques are inherently precise and accurate, the results obtained using these techniques may be invalidated due to contamination caused by poor sample handling and preparation. Therefore, stringent conditions should be routinely used for trace analysis. For example, all reagents, standard solutions, etc., should be ultra-pure and all glassware needs to be scrupulously cleaned. Similarly, stringent conditions should be used for sampling and the pretreatment of samples; these two stages should be simplified as much as possible to minimize the potential for sample loss and contamination. Different complex matrices of the analytical sample require prior mineralization for most analytical methods and this step is critical in the whole analytical procedure for the determination of metal concentration. Sample digestion techniques, such as microwave and conventional acid digestion method for heavy metal determination have been used widely for the dissolution of target elemental analytes. These digestions techniques, however, require the use of concentrated mineral acids and high temperatures [22]. In this study fish samples were cleaned with doubly distilled water and then dissected. Two grams of muscle, liver or gill tissue of the fishes was removed and weighed for the analysis of each metal. For estimation of heavy metal content 2 g of each tissue was taken in a 100-ml Borosil beaker. To this, 2 ml of HNO_3 and 1 ml of $HClO_4$ was added and kept for digestion on a hot plate at 100°C till complete digestion was achieved (Complete digestion involves removal of organic matter by reacting with acids.). It was ensured that the residue obtained after digestion was free from organic matter which acts as impurities in metal analysis [20, 23, 24]. Residue was reconstituted using 1 M of 10 ml Hydrochloric acid (HCl) for further analysis.

Sample Analysis: To analysis of the concentration of Pb(II) and As(II) in different tissues (muscle, gill and liver) of Otolithes rubber, Pampus argenteus, Parastromateus niger, Scomberomorus commerson and Onchorynchus mykiss by anodic stripping voltammetry, in the electrochemical cell 5 mL of each sample solution and 1 mL acetate-acetic acid buffer solution were transferred and diluted to 10 mL by doubly distilled water. The solution was deaereated by passing pure nitrogen for 5 min. The deposition potential was applied to a fresh mercury drop while the solution was stirred. After the deposition step and further 10 s (equilibrium time) the voltammograms were recorded. Different concentration of the standard metal ions was added to the cell, while keeping the deposition time constant. The solution was stirred and purged with nitrogen for 1 min. after each spike. The concentration of Pb(II) and As(II) in the electrolytic cell were calculated in the sample solutions by using standard addition method. The concentration of Hg(II) in the marin organism tissues are analyzed by spectrophotometric method [21].

RESULTS AND DISCUSSIONS

Anodic stripping voltammetry method is applied to the determination of Pb and As in the different tissues (muscle, gill and liver) of marine organisms Otolithes rubber, Pampus argenteus, Parastromateus niger, Scomberomorus commerson, Onchorynchus mykiss. The analysis of Hg was done by spectrophotometric method. The results of toxic metal analysis of muscle, gill and liver of Otolithes rubber is presented in Table 1. toxic metal analysis of this marine organism show that Pb level of the liver tissue is lower than other tissues. Also, As concentration in the muscle tissue is lower than other tissues. As shown in Table 1, Pb, Hg and As content of different tissues of this marine organism is below the permissible level reported by others [11, 13]. The results of toxic metal analysis of Pampus argenteus is presented in Table 2. as shown in Table 2, this marine organism

Table 1: Determination of Pb(II), Hg(II) and As(II) in muscle, gill and liver of *Otolithes ruber*

Metal ion	Muscle (µg/g)	Gill (µg/g)	Liver (µg/g)
Hg (II)	0.030	0.040	-
Pb (II)	0.075	0.075	0.026
As (II)	0.011	0.210	0.260

Table 2: Determination of Pb(II), Hg(II) and As(II) in muscle, gill and liver of Pampus argenteus

Metal ion	Muscle (µg/g)	Gill (µg/g)	Liver (µg/g)
Hg (II)	0.259	0.122	0.070
Pb (II)	0.015	0.031	0.022
As (II)	0.007	0.022	0.063

Table 3: Determination of Pb(II), Hg(II) and As(II) in muscle, gill and liver of *Parastromateus niger*

Metal ion	Muscle (µg/g)	Gill (µg/g)	Liver (µg/g)
Hg (II)	0.040	0.470	0.410
Pb (II)	0.090	0.110	0.100
As (II)	0.015	0.010	0.014

Table 4: Determination of Pb(II), Hg(II) and As(II) in muscle, gill and liver of *Scomberomorus commerson*

Metal ion	Muscle (µg/g)	Gill (µg/g)	Liver (µg/g)
Hg (II)	0.001	0.037	0.060
Pb (II)	0.007	0.082	0.005
As (II)	0.040	0.016	0.026

Table 5: Determination of Pb(II), Hg(II) and As(II) in muscle, gill and liver of *Onchorynchus mykiss*

Metal ion	Muscle (µg/g)	Gill (µg/g)	Liver (µg/g)
Hg (II)	0.004	0.012	0.034
Pb (II)	0.011	0.051	0.018
As (II)	0.008	0.160	0.022

translocate larger quantity of Hg in the muscle to their gill than to their liver. However the results of Pb, Hg and As content of different tissues of this marine organism is below the permissible level [11, 13]. Toxic metal analysis of Parastromateus niger is presented in Table 3. according to the results presented in Table 3, the distribution of toxic metals in the different tissues of this organism is some. However, the only different come from the concentration of Hg in the muscle. As shown in Table 3, Pb, Hg and As content of different tissues of this marine organism is below the permissible level The results of toxic metal analysis of muscle, gill and liver tissues of Scomberomorus commerson are presented in Table 4. toxic metal analysis of gill, muscle and liver tissues of Onchorynchus mykiss are presented in Table 5. The results of marine organism toxic metal content show that mercury concentration reaches to maximum in the gill of *Parastromateus niger* (0.47 µg/g) followed by the liver of *Otolithes rubber* (0.001 µg/g). Lead content reaches to maximum in the gill of *Parastromateus niger* (0.11 µg/g), followed by the liver of *Scomberomorus commerson*, (0.005 µg/g). Arsenic concentration is highest in the liver of *Otolithes rubber* (0.26 µg/g), followed by the muscle of *Pampus argenteus* (0.007 µg/g). Toxic metal contamination in muscle tissue of fish is of particular interest because of the potential risk to human who consume fish. The results of this study show that the concentration of arsenic, lead and mercury of marine organisms Otolithes rubber, Pampus argenteus, Parastromateus niger, Scomberomorus commerson, Onchorynchus mykiss are 0.007-0.04, 0.007-0.09 and 0.001 to 0.259, respectively. These results are in the concentration range of other reports [11, 13]. The results of this study indicate that marine organisms of this study have concentrations were lover than the permissible levels for these toxic metals [25]. Their contribution to the body burden is negligible and the fish species seem to be safe for human consumption.

CONCLUSION

In this paper, toxic metal (lead, mercury and arsenic) contamination of different tissues (muscle, gill and liver) of Otolithes rubber, Pampus argenteus, Parastromateus niger, Scomberomorus commerson, Onchorynchus mykiss are analyzed. The results of toxic metal analysis indicate that toxic metal content of the analyzed marine organisms are well below the permissible levels for these toxic metals.

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