International Journal of Research in Agriculture and Forestry
Volume 5, Issue 1, 2018, PP 8-14
ISSN 2394-5907 (Print) & ISSN 2394-5915 (Online)

Metal Contaminants in Largemouth Bass (Micropterus salmoides, Lacépède, 1802) from Different Origins

António Moitinho Rodrigues[1,2], Paulo Antunes[3], Luísa Paulo[3], Maria Eduarda Pereira[4], Luís Pinto-de-Andrade[1,2,3]

[1]*School of Agriculture - Polytechnic Institute of Castelo Branco, Qt.ª Sr.ª Mércules, Castelo Branco, Portugal.*
[2]*CERNAS-IPCB - Supported by National Funds through FCT - Foundation for Science and Technology, Portugal.*
[3]*CATAA – Zona Industrial de Castelo Branco, Rua A, Castelo Branco, Portugal.*
[4]*CESAM/Department of Chemistry – University of Aveiro, Campus Universitário de Santiago, Aveiro, Portugal.*

***Corresponding Author:** *António Moitinho Rodrigues, School of Agriculture - Polytechnic Institute of Castelo Branco, Qt.ª Sr.ª Mércules, Castelo Branco, Portugal. amrodrig@ipcb.pt*

ABSTRACT

The aim of this work was to evaluate the presence of some metals (Cd, Cr, Cu, Fe, Hg, Mn, Pb, Zn) on largemouth bass (Micropterus salmoides) liver and muscle tissue collected in two different areas: section of Tagus River that makes border between Portugal and Spain and in three small irrigation reservoirs located in Tagus River basin. Individuals were weighed and measured. Age were determined by examining fish scales, and sex determined by gonads observation. Samples of dorsolateral muscle, tail muscle and liver were collected to evaluate metal contaminants. We concluded that metallic concentrations of largemouth bass muscle tissues were below the maximum permissible for a safety utilization of theses fishes in human nutrition.

Keywords: *Heavy Metals, Cadmium, Lead, Mercury, Chromium, Muscle Tissue, Tagus River, Micropterus salmoides, Largemouth Bass.*

INTRODUCTION

Largemouth bass (*Micropterus salmoides*, Lacépède, 1802) is a freshwater fish originating from the United States of America (USA). In the end of the XIX Century it was introduced in Portugal - Azores Islands. In main land it was introduced in 1952 (Sanches and Rodrigues, 2011).

Like in the USA, largemouth bass is one of the most popular freshwater sport's fish in Portugal and it is very important in regional cuisine. It is much appreciated due to their exquisite flavour.

All eaten largemouth bass in Portugal are collected in large dams (Tagus and Guadiana rivers basins) and small irrigation dams (Rodrigues and Sanches, 2012). However, there's a lack of information about metal contaminants of largemouth bass collected in Tagus River.

Heavy metals are considered the most important form of pollution in the aquatic environment because of their toxicity and because metal contents in aquatic environment have been rose by increased activities in industrial, domestic, and agricultural systems (Tuzen, 2009; Kumar *et al.*, 2011; Mousavi *et al.*, 2012). According Mousavi *et al.* (2012) the first outbreak of poisoning caused by consumption of fish contaminated with heavy metals in humans was observed in Japan - Minamata Bay in 1953, during which more than 43 local residents died and more than 700 suffered permanent disabilities after consumption of contaminated fish by industrial sewage of a factory.

For decades, Tagus River received environmental pollutants from non-point and point sources that included intensive agriculture, industrial entities, municipalities, mining activities and a Spanish nuclear power plant. This nuclear power plant is in a radius around to 150 km far from the local were largemouth bass were collected.

Heavy metals can enter from contaminated aquatic systems into fish body by different routes and accumulate in organisms' tissues (Tuzen, 2009; Mahamadi and Chapeyama, 2013). Some of the metals found in fish might be essential as they play important roles in biological systems of the fish as well as in the human being. The common heavy metals that are found in fish are cadmium (Cd), chromium (Cr), copper (Cu), iron (Fe), mercury (Hg), manganese (Mn), lead (Pb) and zinc (Zn). Cu, Fe, Mn and Zn are essential metals while Cd, Cr, Hg and Pb are toxic metals (Kumar *et al.*, 2011; Bat *et al.*, 2017). However essential metals can have toxic and adverse effects at high concentrations (Tuzen, 2009). Metal accumulation in fish tissues has been recognized to be mainly found in liver and gills. In muscle tissue such accumulation is not so severe (Jia *et al.*, 2017). Since fish are highly consumed by human beings and may accumulate large amounts of some metals from the water and sediments, it is important to determine the concentration of heavy metals in commercial fish in order to evaluate the possible risk of fish consumption for human health (Ekpo *et al.*, 2008; Lopes, 2009; Tabinda *et al.*, 2010; Khoshnoud *et al.*, 2011; Kumar *et al.*, 2011).

Although some studies have been done to determine largemouth bass heavy metals from small irrigation dams (Belo *et al.*, 2007), prior to this study the status of contaminants in largemouth bass collected from the section of Tagus River that makes border between Portugal and Spain was unknown. Obviously, it is very important to know if those fishes are in safety conditions to be consumed by human beings.

The aim of this work was to evaluate some metals (Cd, Cr, Cu, Fe, Hg, Mn, Pb, and Zn) present on liver and muscle tissue of largemouth bass collected in Tagus River (TR) (N=9), compared with fish collected in three small irrigation reservoirs (IR) (N=11), located in Tagus River basin.

MATERIAL AND METHODS

Present research reflects a concern presented to the authors by some authorities regarding the heavy metal contamination of a popular freshwater sport's fish very important in Portuguese regional cuisine especially in the countryside. The Tagus River is the longest river of the Iberian Peninsula with several hydroelectric dams, lentic systems where local population can fish largemouth bass.

After fishing, fishes were stored at -20ºC. In sampling operations, they were slowly unfrozen, weighted, measured and aged by fish scale examination and sex determined by gonads observation. Samples for dorsolateral muscle and tail muscle were collected from the right side of the fish.

Total mercury was determined in freeze-dried samples by atomic absorption spectrometry (AAS) with thermal decomposition and gold amalgamation using an Advanced Mercury Analyser (LECO model AMA-254), as described by Costley *et al.* (2000). The analysis is performed directly on the sample, without digestion or specific pre-treatment, avoiding mercury losses or contamination as well as matrix interferences. At least three replicate measurements were carried out for each sample (acceptable relative standard deviation among replicates: <10%). Analytical quality control was performed by using Certified Reference Material TORT 2 - Lobster Hepatopancreas. About 200 mg of dry liver sample and 1 g of freeze dried muscle were digested with a mixture of HNO_3 (10% v/v) and H_2O_2 (30% v/v) at 100ºC in a block digestion system (Digiprep MS) until complete digestion (12h). The final residue was diluted to 50 mL with HNO_3 (10% v/v) and filtered. The other heavy metals were carried out using an Inductively Coupled Plasma-Optical Emission Spectrometry (ICP-OES- Activa M, Horiba Jobin Yvon), operating at 1000 W plasma power, 15 L min^{-1} plasma gas flow, 0.02 L min^{-1} nebulizer air flow and 1.0 bar air pressure. The analytical wavelengths (nm) were set of the following: Cd (228.802), Cr (205.571), Cu (327.395), Fe (259.940), Mn (257.611), Pb (283.305) and Zn (213.857) (ISO, 2007). All percentages of recovery for total mercury were within the range of 80–120% (N=19). For other metals, all standard solutions used were prepared by diluting 1000 mg L^{-1} single-element standard solutions (Prolabo, Titrinorm) in HNO_3 0.1% (v/v). Quality control samples were from a multi-elemental standard solution of 100 mg L^{-1} containing all analysed elements from SCP science was used. HNO_3 65% (Merck) and H_2O_2 30% (Prolabo) were of analytical grade and high purity Milli-Q water was used for dilutions.

Table 1 shows the characterization of the largemouth bass according TR or IR provenience. The largemouth bass TR average weight was 435.14 g (±109.15), average length 278.33 mm (±23.28), average K condition factor 1.98 (±0.09) and average age 3.11 years (±0,78) and these figures were similar (p>0.05) to largemouth bass IR average weight 410.84 g (±137.71), average length 278.36 mm (±31.13), average K condition factor 1.86 (±0.17) and average age 3.18 years (±0,60). Also liver weight was similar in largemouth bass TR (4.74 g ±1.29) and IR (4.25 g ±1.51) (p>0.05).

Statistical analysis was carried out with Paired-Samples T Test. For each metallic contaminant was determined mean and standard deviation (±sd). Differences in mean values were accepted as being statistically significant if p≤0.05.

Using linear regression analysis (Pearson correlation) we try to found the relationship between each metallic contaminant and largemouth bass weight, length and K condition factor.

Results

The mean of Cd, Cr, Cu, Fe, Hg, Mn, Pb, and Zn concentrations in the muscle of largemouth bass TR and IR are shown in Table 2.

In this work Cd, Cr, and Pb figures in all samples of largemouth bass muscle were lower than the apparatus limit of quantification (LOQ) (0.05, 0.03 and 0.2 $mg.kg^{-1}$ wet weight respectively) (Tables 2 and 3). The LOQ is the minimum concentration that can be quantified at a specified level of precision or accuracy (or both).

Table1. *TR (N=9) and IR (N=11) Largemouth Bass Average Weight, Length, K Condition Factor and Age and Liver Weight.*

	Weigh (g)	Length (mm)	K condition factor	Age (year)	Liver weight (g)
Largemouth bass TR	435.14±109.15	278.33±23.28	1.98±0.09	3.11±0.78	4.74±1.29
Largemouth bass IR	410.84±137.71	278.36±31.13	1.86±0.17	3.18±0.60	4.25±1.51
Total	421.78^{ns}±123.09	278.35^{ns}±27.17	1.91^{ns}±0.15	3.15^{ns}±0.67	4.47^{ns}±1.41

Note: *TR – largemouth bass collected in Tagus River; IR - largemouth bass collected in three small irrigation reservoirs located in Tagus River basin; p – significant values; ns - the difference is not significant for p>0.05.*

Table2. *Metal Concentration ($mg.kg^{-1}$ ww) on Muscle of Largemouth Bass TR (N=9) and IR (N=11).*

Elements	TR ($mg.kg^{-1}$ ww)	IR ($mg.kg^{-1}$ ww)	Total ($mg.kg^{-1}$ ww)	Max.($mg.kg^{-1}$ ww)	Max. tolerable ($mg.kg^{-1}$ ww)
Cd	<LOQ(0.05)	<LOQ(0.05)	-	-	0.05 **(a)**
Cr	<LOQ(0.03)	<LOQ(0.03)	-	-	1 **(d)**
Cu	0.159±0.016	0.148±0.017	0.153^{ns}±0.018	0.181 (**TR**)	50 **(b, c)**
Fe	1.450±0.278	1.803±0.438	1.644^{*}±0.408	2.528 (**IR**)	50 **(e)**
Hg	0.078±0.018	0.223±0.173	0.158^{*}±0.146	0.546 (**IR**)	0.5 **(a)**
Mn	0.038±0.036	0.048±0.033	0.043^{ns}±0.034	0.096 (**IR**)	4 **(f)**
Pb	<LOQ(0.2)	<LOQ(0.2)	-	-	0.2 **(a)**
Zn	3.760±0.263	4.171±0.490	3.986^{*}±0.447	5.323 (**IR**)	100 **(g)**

Note: *ww – wet weight; LOQ – limit of quantification; ns - the difference is not significant for p>0.05; * - p≤0.05; (a) EU, 2008; (b) Leung et al., 2014; (c) Jia et al., 2017; (d) US EPA, 1980; (e) Elkareem, 2014; (f) Tuzen, 2009; (g) Mahamadi and Chapeyama, 2013.*

Table3. *Metal Concentration ($mg.kg^{-1}$ ww) on Liver of Largemouth Bass TR (N=9) and IR (N=11).*

Elements	Liver TR ($mg.kg^{-1}$ww)	Liver IR ($mg.kg^{-1}$ww)	p – Value
Cd	<LOQ(0.05)	<LOQ(0.05)	-
Cr	<LOQ(0.03)	<LOQ(0.03)	-
Cu	2.05±0.60	5.44±6.06	0.114
Fe	74.26±23.10	280.98±214.07	0.010
Hg	0.03±0.006	0.11±0.01	0.050
Mn	1.61±0.18	1.50±0.53	0.567
Pb	<LOQ(0.2)	<LOQ(0.2)	-
Zn	17.38±1.51	18.91±5.09	0.399

Note: *p – Significant values; ww – wet weight.*

Discussion

The mean values of Cd in both TR and IR fish were lower than EU (2008) and FAO (1983) limits (0.2 $mg.kg^{-1}$ ww) and lower than the figures reported by Belo *et al.* (2007) and Leung *et al.* (2014) (*Micropterus salmoides*) and Yousaf *et al.* (2012) (*Wallago attu*).

Cr content in both largemouth bass muscle tissues TR and IR were lower than US EPA (1989) limit (1 $mg.kg^{-1}$ ww) and lower than the figures reported by Leung et al. (2014) (*Micropterus salmoides*), Tabinda *et al.* (2010) (*Labeo rohita*), Al Farraj *et al*. (2011) (*Sepia* pharaonis), Mousavi et al. (2012) (*Oncorhynchus mykiss*) and Elkareem *et al.* (2014) (*Clarias lazera*). There's no difference between Cu concentration in largemouth bass TR (0.159 $mg.kg^{-1}$ ww ±0.016) and IR (0.148 $mg.kg^{-1}$ ww ±0.017).

However, the non-significant Cu higher value of TR largemouth bass could be related with the type of Spanish agriculture along Tagus River and the use of Cu for the vineyards treatment. In both cases TR and IR largemouth bass Cu were much lower than Leung *et al.* (2014) and Jia *et al.* (2017) limit (50 $mg.kg^{-1}$ ww) and FAO (1983) limit (10 $mg.kg^{-1}$ ww) and lower than the results of Belo *et al*. (2007) (*Micropterus salmoides*), Fernandes *et al*. (2008) (*Liza* saliens), Khoshnoud et al. (2011) (*Scomberomorus commerson* and *Otolithes ruber*) and Yousaf *et al*. (2012) (*Wallago attu*) but higher than Tabinda *et al.* (2010) (*Labeo rohita*) and Jia *et al.* (2017) (*Carassius auratus, Pelteobagrus fulvidraco* and *Squaliobarbus curriculus*).

Although the Fe concentration in both TR and IR fish muscles were much lower than Elkareem *et al*. (2014) limit (146 $mg.kg^{-1}$ ww) and lower than results reported by Belo *et al.* (2007) (*Micropterus salmoides*) and Yousaf *et al.* (2012) (*Wallago attu*), there were slightly differences ($p \leq 0.05$) between Fe concentration in IR (1.803 $mg.kg^{-1}$ ww ±0.438) fish muscles and in TR (1.450 $mg.kg^{-1}$ ww ±0.278). This difference could be related with the schist soil around the three small irrigation dams (IR).

We did not find any significant difference between Mn in TR (0.038 $mg.kg^{-1}$ ww ±0.036) and IR largemouth bass muscle tissues (0.048 $mg.kg^{-1}$ww ±0.033) and the values of Mn in both fish TR and IR were lower than the daily upper intake limit referred by Tuzen (2009) (11 $mg.day^{-1}$) and much lower than values reported by Belo *et al.* (2007) and Leung *et al.* (2014) for *Micropterus salmoides*.

Pb contents in both TR and IR fish were lower than FAO (1983) limit (1.5 $mg.kg^{-1}$ ww) and EU (2008) limit (0.2 $mg.kg^{-1}$ ww) and lower than results reported by Belo et al. (2007), (*Micropterus salmoides*), Fernandes *et al*. (2008) (*Liza saliens*) and Yousaf *et al*. (2012) (*Wallago attu*).

Zn concentration was greater in IR fish muscles (4.171 $mg.kg^{-1}$ ww ±0.490) than in TR fish muscle (3.760 $mg.kg^{-1}$ ww ±0.263) ($p \leq 0.05$). Both TR and IR fish muscles were found to contain Zn concentration much below limits which are 100 $mg.kg^{-1}$ ww (Mahamadi and Chapeyama, 2013) or 150 $mg.kg^{-1}$ ww (FAO, 1983). Also our figures are lower than reported by Belo *et al.* (2007) and Mahamadi and Chapeyama (2013) (*Micropterus salmoides*), Fernandes *et al.* (2008) (*Liza saliens*) and Yousaf *et al.* (2012) (*Wallago attu*) but higher than Khoshnoud *et al*. (2011) (*Micropterus salmoides*).

Finally, the average values of Hg in both fish TR and IR were below EU (2008) limit (0.5 $mg.kg^{-1}$ ww) and FAO (1983) limit (0.14 $mg.kg^{-1}$ ww). However, our results were higher than results reported by Ekpo *et al.* (2008) and Khoshnoud et al. (2011) (Metacembelus iconnbergii, *Clarias lazera, Citharinus citharus, Tilapia zillii*) but lower than total Hg concentration in largemouth bass muscle tissue (ranged from 0.12 to 0.98 $mg.kg^{-1}$ ww) reported by Gehringer *et al*. (2013). There's a difference ($p \leq 0.05$) between Hg concentration in TR fish muscles (0.078 $mg.kg^{-1}$ ww ±0.018) and IR (0.223 $mg.kg^{-1}$ ww ±0.173). We find one IR largemouth bass with Hg value slightly higher (0.546 $mg.kg^{-1}$ ww) than the acceptable value (0.5 $mg.kg^{-1}$ ww) (EU, 2008). It was a largemouth bass with 587.3 g weight, 324 mm length and 4 years old. This fish represents 9.1% of IR largemouth bass and 5.0% of all largemouth bass collected. However, 0.546 $mg.kg^{-1}$ ww it is a lower value than the accepted for other fishes (1 $mg.kg^{-1}$ ww) which includes European eel (*Anguilla anguilla*), pike (*Esox lucius*) and other carnivorous fish (EU, 2008). An apex predator like largemouth bass has the potential, due to its trophic position, to bioaccumulate high levels of Hg (Tabinda *et al.*, 2010; Kumar *et al.*, 2011). Measured Hg tissue levels in largemouth bass can range from very low (<0.1 $mg.kg^{-1}$ ww) to extremely high (>10 $mg.kg^{-1}$ ww) (Rumbold and Fink 2006). According ADPH (2010) of the sixty-six mercury advisories in the Alabama coastal plain (USA), forty-nine were issued for, or included, largemouth bass. In our study 100% of TR largemouth bass and 36.4% of IR largemouth bass are Hg concentration lower than 0.1 $mg.kg^{-1}$ ww.

In our study we found a positive correlation between Hg largemouth bass muscle concentration and length ($r=0.690$; $p \leq 0.01$), weight ($r=0.619$; $p \leq 0.01$) and fish age ($r=0.554$; $p \leq 0.05$) indicating that largemouth bass are accumulating greater amount of Hg as biggest and older they are. Like Gehringer *et al*. (2013), in this study total Hg concentration in muscle tissue was not related to K condition factor. However considering only largemouth bass from IR we found a very high positive correlation between Hg largemouth bass muscle concentration and length ($r=0.942$; $p \leq 0.01$), weight ($r=0.930$; $p \leq 0.01$) and fish age ($r=0.873$; $p \leq 0.01$). Small irrigation reservoirs are closed systems with no hydrodynamic capacity to recycle the diffuse sources of contamination.

According to our results, the concern may be greater with consumption of largemouth bass caught on small irrigation reservoirs. For this reason, we estimate the equation Hg ($mg.kg^{-1}$ ww) = -1,241 + 0.005 x Length (mm) ($r2=0.902$; $p \leq 0.01$). With this equation the fisherman can estimate the Hg concentration in largemouth bass muscle tissue after measuring the fish.

The metal concentrations on liver of largemouth bass TR and IR are shown in Table 3 and were higher than metal concentration on muscle tissue for all analyzed heavy metals, with exception of Hg. This lower level of Hg on liver relative to other tissues may be attributed to the high coordination behavior of MT polypeptides with respect to either cognate or noncognate metal ions as reported by Palacios *et al.* (2014). In addition, liver is the principal organ responsible for the detoxification, transportation, and storage of toxic substances and it is an active site of pathological effects induced by contamination. Higher levels of Hg in muscle than in liver are characteristic of some species, in particular on non-contaminated environments (Havelkova *et al.*, 2008). In the samples analyzed, average levels of metal in liver are below limits, except for Fe. In general, fish liver is not consumed or it is consumed in small amounts, therefore liver is not referred in legislation. In all cases it would be safe to consume a considerable amount of fish liver.

CONCLUSIONS

Only Fe, Hg and Zn showed significant differences in largemouth bass muscle tissue from TR and IR. Largemouth bass collected in small irrigation reservoirs presented higher values than largemouth bass from Tagus River despite there were no direct contamination in irrigation reservoirs. Therefore, this indicates that Tagus River is not accumulating metals in the studied section, and can recycle the inputs from contaminants that it receives.

We concluded that metallic concentrations of largemouth bass muscle tissues collected in the section of Tagus River that makes border between Portugal and Spain and collected in small irrigation reservoirs located in Tagus River basin were below the maximum permissible for a safety utilization of theses fishes as human food. Nevertheless, a good program of heavy metals control, over those and other fishes should be applied with a certain frequency to prevent future eventual contamination of fishes and the natural consequences to human health.

REFERENCES

[1] ADPH (Alabama Department of Public Health), 2010. Alabama Fish Consumption Advisories. Available from: https://www.google.pt/?gfe_rd=cr&ei=xGraU6bSEfDe8geEzoHgDQ&gwsrd=ssl#q=Alabama+Department+of+Public+Health+largemouth+bass [accessed 01 August 2014].

[2] Al Farraj, S., A.H. El-Gendy, H. Alyahya and M. El-Hedeny, 2011. Heavy Metals Accumulation in the mantle of the common cuttlefish Sepia pharaonis from the Arabian gulf. Australian Journal of Basic and Applied Sciences, 5(6): 897-905.

[3] Bat, L., E. Arici and D. Urkmez, 2017. Heavy metal levels in the Black Sea sprat (Sprattus sprattus). International Journal of Research in Agriculture and Forestry, 4(6): 1-8.

[4] Belo, A.P., V.O. Castro and A.M. Rodrigues, 2007. Determination of some metal-ions in the bodies of black-bass (Micropterus salmoides) and tench (Tinca tinca), and from water reservoirs close to border of Portugal/Spain. International Journal of Agriculture and Biology, 9(3): 408-411.

[5] Costley, C., K. Mossop, J. Dean, L. Garden, J. Marshall and J. Carroll, 2000. Determination of mercury in environmental and biological samples using pyrolysis atomic absorption spectrometry with gold amalgamation. Analytica Chimica Acta, 405(1-2): 179-183.

[6] Ekpo, K.E., I.O. Asia, K.O. Amayo and D.A. Jegede, 2008. Determination of lead, cadmium and mercury in surrounding water and organs of some species of fish from Ikpoba river in Benin city, Nigeria. International Journal of Physical Sciences, 3(11): 289-292.

[7] Elkareem, M.M.A.A., A.M.H. Karrar, and AA.K.S. Ali, 2014. Relationship of biometric size-weight, nutritive value, and metal concentrations in Clarias lazera (Cuvier and

Valenciennes) reared in treated wastewater. Jordan Journal of Biological Sciences, 7(3): 217-225.

[8] EU, 2008. Commission regulation (EC) No 629/2008 of 2 July 2008 amending Regulation (EC) No 1881/2006 setting maximum levels for certain contaminants in foodstuffs. Official Journal of the European Union, L 173, 3.7.2008.

[9] FAO, 1983. Compilation of legal Limits for Hazardous Substance in Fish and Fishery Products. Food and Agricultural Organization Fishery circular 466: 5-100.

[10] Fernandes, C., A. Fontaínhas-Fernandes, D. Cabral and M.A. Salgado, 2008. Heavy metals in water, sediment and tissues of Liza saliens from Esmoriz-Paramos lagoon, Portugal. Environmental Monitoring Assessment, 136: 267–275.

[11] Gehringer, D.B., M.E. Finkelstein, K.H. Coale, M. Stephenson and J.B. Geller, 2013. Assessing mercury exposure and biomarkers in largemouth bass (Micropterus salmoides) from a contaminated river system in California. Archives of Environmental Contamination and Toxicology, 64(3): 484-493.

[12] Havelkova, M., L. Dusek, D. Nemethova, G. Poleszczuk and Z. Svobodova, 2008. Comparison of mercury distribution between liver and muscle – a biomonitoring of fish from lightly and heavily contaminated localities. Sensors, 8: 4095-4109.

[13] ISO, 2007. Water quality — Determination of selected elements by inductively coupled plasma optical emission spectrometry (ICP-OES). International Standard ISO 11885: 5-14.

[14] Jia, Y., L. Wang, Z. Qu, C. Wang and Z. Yang, 2017. Effects on heavy metal accumulation in freshwater fishes: species, tissues, and sizes. Environmental Science and Pollution Research, 24: 9379-9386.

[15] Khoshnoud, M.J., K. Mobini, K. Javidnia, P. Hosseinkhezri and K.A Jamshid, 2011. Heavy metals (Zn, Cu, Pb, Cd and Hg) contents and fatty acids ratios in two fish species (Scomberomorus commerson and Otolithes ruber) of the Persian gulf. Iranian Journal of Pharmaceutical Sciences, 7(3): 191-196.

[16] Kumar, B., D.P. Mukherjee, S. Kumar, M. Mishra, D. Prakash, S.K. Singh, and C.S. Sharma, 2011. Bioaccumulation of heavy metals in muscle tissue of fishes from selected aquaculture ponds in east Kolkata wetlands. Annals of Biology Research, 2(5): 125-134.

[17] Leung, H.M., A.O.W. Leung, H.S. Wang, K.K. Ma, Y. Liang, K.C. Ho, K.C. Cheung, F. Tohidi, and K.K.L. Yung, 2014. Assessment of heavy metals/metalloid (As, Pb, Cd, Ni, Zn, Cr, Cu, Mn) concentrations in edible fish species tissue in the Pearl river delta (PRD), China. Marine Pollution Bulletin, 78: 235-245.

[18] Lopes, A.M.R.M., 2009. Avaliação da contaminação em metais pesados no pescado: análise da situação do pescado comercializado em Portugal e dos alertas emitidos pelo sistema RASFF (Rapid Alert System for Food and Feed). Dissertação apresentada para obtenção do grau de Mestre em Tecnologia e Segurança Alimentar, FCT –UNL, Lisboa.

[19] Mahamadi, C., and R. Chapeyama, 2013. Determinations of cadmium, lead, nickel, and zinc in Micropterus salmoides and Oreochromis niloticus by flame atomic absorption spectrometry after cloud point extraction. Toxicological & Environmental Chemistry, 95(1): 163-171.

[20] Mousavi, S.L., A. Moghimi and M. Nasimi, 2012. Investigation of pollution level of some heavy metals including cadmium, lead, chromium, and nickel in the flesh of farmed rainbow trout in Sepidan city of Fars province. Journal of American Science, 8(7): 140-143.

[21] Palacios, O.; Pérez-Rafael, S.; Pagani, A.; Dallinger, R.; Atrian, S.; Capdevila, M. 2014. Cognate and noncognate metal ion coordination in metal-specific metallothioneins: The Helix pomatia system as a model. Journal of Biology Inorganic Chemistry, 19, 923–935.

[22] Rodrigues, A.M. and J.C. Sanches, 2012. A produção comercial de achigãs (Micropterus salmoides). Agroforum, 28: 45-53.

[23] Rumbold, D. G., and L.E. Fink, 2006. Extreme spatial variability and unprecedented methyl mercury concentrations within a constructed wetland. Environmental Monitoring and Assessment, 112: 115-135.

[24] Sanches, J.C. and A.M. Rodrigues, 2011. O achigã (Micropterus salmoides), uma espécie com interesse para a pesca desportiva. Agroforum, 26: 17-22.

[25] Tabinda, A.B., Husain, M. Hussain, I. Ahmed, and A. Yasar, 2010. Accumulation of toxic and essential trace metals in fish and prawns from Keti Bunder Thatta district, Sindh, Pakistan Journal of Zoology, 42(5): 631-638.

[26] Tuzen, M., 2009. Toxic and essential trace elements contents in fish species from the Black Sea, Turkey. Food and Chemical Toxicoloy, 47: 1785-1790.

[27] US EPA, 1980. Ambient water quality criteria for chromium. Environmental Criteria and Assessment Office, Cincinnati, OH. EPA 440/5-80-035. NTIS PB 81-117467. Available from National Technical Information Service, Springfield, VA.

[28] Yousaf, M., A. Salam, M. Naeem and M.Y. Khokhar, 2012. Effect of body size on elemental concentration in wild Wallago Attu (Bloch and Schneider) from southern Punjab, Pakistan. African Journal of Biotechnology, 11(7): 1764-1767.

Citation: *R. António Moitinho, A. Paulo, P. Luísa, P. Maria Eduarda and A. Luís Pinto-de, "Metal Contaminants in Largemouth Bass (Micropterus salmoides, Lacépède, 1802) from Different Origins", International Journal of Research in Agriculture and Forestry, vol. 5, no. 1, pp. 8-14, 2018.*