TOXICOLOGICAL EFFECT OF HEAVY METALS TO HUMAN HEALTH FROM CONSUMPTION OF FISHES COLLECTED FROM THE MARINE COASTAL WATER OF SABRATHA, LIBYA

Fathi Alhashmi Bashir, Faculty of Sciences, Sabratha University
Hashm Fathi Ali, Faculty of Agricultural and Veterenary Scinces, Al-Zawia University

ABSTRACT:

This study estimted the concentration of heavy metals (Cu, Zn, Cd and Pb) in three commercially important marine fishes, namely, *Thunnus thynnus*, *Sarpa salpa* and *Trachurus trachurus*. The fish samples were collected from Sabratha which are located on the western coastal waters of Libya. The organs of fish, namely, gills, liver, and muscle were carefully separated through the dissection from fish for the determination of heavy metal using Absorption Spectrophotometer. The findings of the study showed that the muscle tissues of the three species presented Zn levels far under the permissible level presented by WHO but higher than the permissible level presented by FAO. Moreover, levels of Cu in the muscles of *T. thynnus* (22.87±0.15) and *T. trachurus* (16.87±0.09) were higher than the permissible levels presented by WHO and FAO. Furthermore, the the levels of Pb in the muscles of *T. thynnus*, *S. salpa* and *T. trachurus* (1.66±0.01), (4.77±0.01) and (1.30±0.01) µg/g dry weight, respectively were higher than the permissible levels reported by WHO and FAO. The values in the muscles exceeded the standard guideline values and hence would pose health hazard to consumers.

Keywords: heavy metals, marine fish, Sabratha Coastal water

1. INTRODUCTION:

Aquatic ecosystem has various sources of pollution, resulting from human activities such as industrial processes, amplified urbanization, and waste discharge (Aladaileh et al., 2020). Coastal water has become a major concern, because of its values for socioeconomic development and human health. With the growth of human populations and commercial industries, marine water has received large amounts of pollution from a variety of sources such as recreation, fish culture, and the assimilation and transport of pollution effluents (Abida et al., 2009).

Heavy metals are significant environmental pollutants, and their toxicity is a problem of increasing significance for ecological, evolutionary, nutritional and environmental reasons. Contamination of aquatic ecosystem by toxic heavy metals has been a major environmental problem since long (Kelle et al., 2015). Some of the past episodes of heavy metal contamination in the aquatic environment have increased the awareness about their toxicity. The accumulation of toxic metals to hazardous levels in aquatic biota has become a problem of increasing concern (Kelle et al., 2015).

Pollution of heavy metals in aquatic ecosystem is growing at an alarming rate and has become an important worldwide problem. Processes such as weathering of rocks, human-induced emissions from mining, and other mining-related processes are ultimately likely to elevate heavy metal concentrations in water bodies, leading to increased pollution (Kelle et al., 2015). Among the different metals analyzed, Cd and Pb are non essential and classified as chemical hazards and maximum residual levels have been prescribed for humans (Demirezen and Uruc, 2006; Mehana et al., 2020).

Cadmium affects adversely a number of organs and tissues such as kidney, heart (Houston, 2007), lung (fibrosis), skeletal system, testes, placenta, brain and the central nervous system (CNS). Whereas, chronic Pb toxicity in humans often develop dullness, irritability, poor attention span, epigastric, constipation, vomiting, convulsions, coma and death (Bellinger et al., 1992; Gwalteney-Brant, 2002). Moreover, some of the key enzymes which help in the synthesis of heme, such as Adenosine triphosphate, carbonic anhydrase, cytochrome oxidase are inhibited by Pb (Jarup, 2003).

Essential metals, such as Cu and Zn are required in trace amounts for smooth function of different biological systems and essential for enzymatic activity and many biological processes (Padrilah et al., 2018). Gwalteney-Brant, (2005) have claim that deficiency of Zn will cause loss of appetite, growth retardation, skin changes, and immunological abnormalities. On the other hand, the nutritional benefits of fish are mainly due to the content of high-quality protein and high content of the two kinds of omega-3 polyunsaturated fatty acids. Omega-3 fatty acids (EPA) have probed to have protective effects in preventing coronary heart disease, reducing arrhythmias and thrombosis lowering plasma triglyceride levels and reducing blood clotting tendency (Domingo et al., 2007).

Fish has been recognized as an important food source for the human body which provides essential fatty acids like Omega 3, proteins, vitamins, and minerals (Enkeleda et al., 2013). Fishes are a good biondicator organisms to investigate heavy metal distribution in the aquatic environment, Fish are a useful bioindicator for the determination of metal pollution in aquatic ecosystems (Ahmad and Shuhaimi-Othman 2010). The main purpose of biomonitoring heavy metal concentrations in biota especially fish has been to determine the toxicological threat posed to organisms and also for health risks to humans from the ingestion of edible species (Ahmad and Shuhaimi-Othman 2010).

Marine coastal water around Sabratha located on the western coast of Libya is selected as study area due to the economic and strategic factors. It is a potential fish breeding ground (fishing activities), and various anthropogenic activities like draining of sewerage, dumping of hospital and industrialization, urbanization, agriculture activity. The main objectives of this research were to study the content of heavy metals in Mediterranean Sea fish in Sabratha, Libya and to match their concentrations within muscles, livers and gills of selected fish species from the west coastal water of Sabratha were examined.

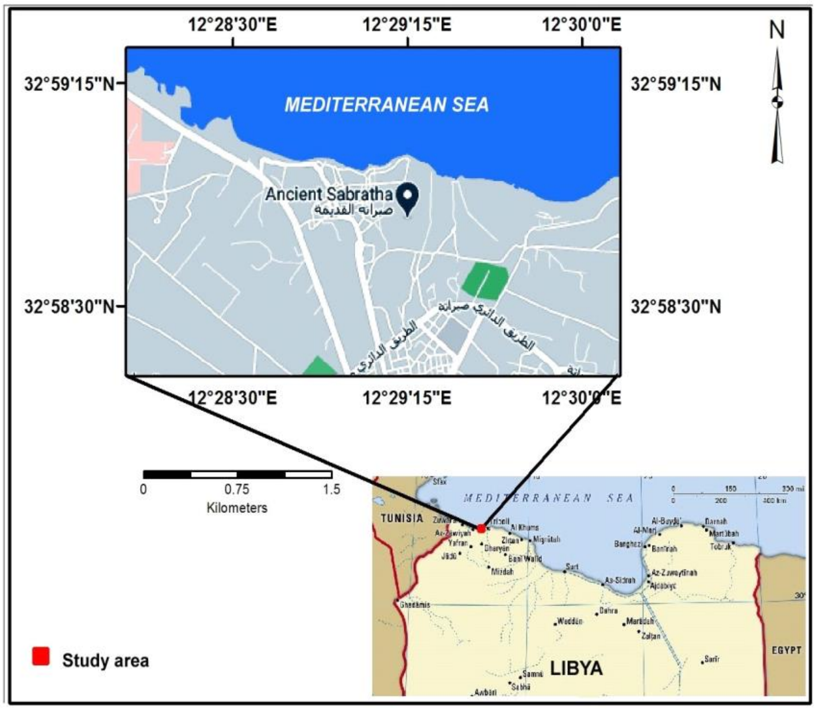
2. METHODOLOGY:

2.1. Sample preparation

Fresh fish samples were collected during September 2018 from local fishermen from Sabratha in western coast of Libya (figure 1). Three selected fish species examined were *Thunnus thynnus*, *Sarpa salpa* and *Trachurus trachurus*. These are commercially important and nutritious species in Libya. Findings of these studies are consistently important to indicate the safety of local fish products in terms of the heavy metals concentration. The samples were collected, and then brought to laboratory for the metal analysis. A total length (cm) and weight (g) of the samples were measured before dissection. Fish were gutted, washed with tap water and muscle, liver and gills were removed and dried in an oven at 70C˚-90C˚until constant weight was obtained.

2.2. Digestion procedures:

The homogenized samples (muscle, liver and gills) were digested with HNO_3 (65%) and H_2O_2 (30%). The concentration of Cu, Zn, Cd and Pb were measured using the Absorption Spectrophotometer and the results were expressed in micrograms of metal per dry weight gram of fish (µg/g).



2.3. Statistical analysis

One way analysis of variance (ANOVA) and Duncan's test were performed to assess whether heavy metal concentrations varied significantly between organs and species. For which P value less than 0.05 was considered as suggestive of statistical significance. SPSS for windows, version 16.0 was used to perform all the above-mentioned tests.

3. RESULTS

The mean metal levels (µg/g dry weight) in the muscles, livers and gills of the examined species, *T. thynnus*, *S. salpa* and *T. trachurus* from Sabratha were summarized in table (1).

Table 1. Metal levels in different organs of fish samples collected from the coastal waters of Sabratha and metal permissible limits by different authorities

Fish species	N	Organ	Metal levels ± SD (µg g⁻¹ dry wt.)			
			Zn	Cu	Cd	Pb
T. thynnus	10	Muscle	131.03±4.2	22.87±0.15	0.12±0.01	1.66±0.01
		Liver	217.37±8.4	52.96±0.8	0.32±0.05	2.07±0.05
		Gills	373.6±9.1	40.03±0.07	0.29±0.01	2.06±0.05
S. Salpa	10	Muscle	133.6±0.4	5.63±0.3	0.15±0.01	4.77±0.01
		Liver	187.67±0.71	40.72±0.4	0.31±0.01	7.64±0.04
		Gills	259.5±2.01	74.05±0.7	0.26±0.2	3.11±0.01
T. trachurus	10	Muscle	106.2±0.2	16.87±0.09	0.06±0.01	1.30±0.01
		Liver	199.4±2.1	76.8±2.4	0.63±0.15	1.69±0.03
		Gills	388.5±9.4	32.04±0.5	0.25±0.03	10.59±0.04
WHO[1]			150	10	0.2	0.2
FAO[2]			30–100	10	0.2	0.5–0.6

[1] WHO (1989),[2] FAO (1992)

Generally, the highest concentrations of Cu, Zn, Cd and Pb were found in the gills of the examined fish species. Differences in concentrations among different tissues were statistically significant for all assessed heavy metals ($p < 0.05$). The concentrations of the studied metals descending in the following order Zn > Cu > Pb > Cd in the three species. Zinc exhibited the highest concentrations in all the examined organs of the fish species, followed by Cu. On the other hand, the levels of Cd were generally the lowest. According to the results (Table 1), the highest concentrations of Zn were observed in the gills of *T. trachurus* was 388.5±9.4 µg/g and its lowest levels were recorded in the livers of the same fish was 106.2±0.2 µg/g. Moreover, the Zn concentrations in muscles of the different examined species was in the following order *S. salpa* > *T. thynnus* > *T. trachurus* . The highest mean concentrations of Zn in the muscle tissues of *S. salpa* was 133.6±0.4 µg/g and the lowest levels of Zn in muscle were determine in *T. trachurus* was 106.2±0.2 µg/g. furthermore, the lowest mean Zn levels were in the gills of *S. Salpa* were 259.5±2.01 µg/g, on the contrary, the highest mean Zn concentration was in the gills of *T. trachurus* were 388.5±9.4 µg/g. The maximum Cu levels among the examined fish species were 76.8±2.4 µg/g, 52.96±0.8 µg/g and

40.72±0.4 µg/g in livers of *T. trachurus*, *T. thynnus* and *S. salpa* respectively. Whereas its minimum levels were 5.63±0.3 µg/g, 16.87±0.09 µg/g and 22.87±0.15 µg/g in muscles of *S. salpa*, *T. trachurus* and *T. thynnus* respectively. According to our findings, the highest mean Cd concentrations were 0.63±0.15 µg/g, 0.32±0.05 µg/g and 0.31±0.01 µg/g in livers of *T. trachurus*, *T. thynnus* and *S. salpa*. On the contrary, its lowest mean levels were 0.06±0.01µg/g in muscles of *T. trachurus*, 0.12±0.01µg/g in muscles of *T. thynnus* and 0.15±0.01 µg/g in muscles of *S. salpa.* The maximum Pb levels among the examined fish species were 10.59±0.04 µg/g in gills of *T. trachurus*, 7.64±0.04 µg/g and 2.07±0.05 in livers of *S. salpa* and *T. thynnus* respectively. Whereas its minimum levels were 1.30±0.01µg/g in muscles of *T. trachurus* and 1.66±0.01µg/g in the muscles of *T. thynnus* and 1.69±0.01µg/g in the livers of *T. trachuru.*

4. DISCUSSION

The concentration of metals in muscles reflect the concentrations of metals in the waters, where the fish species lives, whereas the concentrations in liver represent storage of metals. Increased metal concentrations in liver may represent storage of sequestered products in this organ. Moreover, the concentration level of metals in gills represents the level of metals in water, where they dwell (Uysal et al. 2009). Muscles, livers and gills were chosen as target organs for assessing metal accumulation. The levels of heavy metals found in muscle, liver and gills of *T. thynnus*, *S. salpa* and *T. trachurus* are shown in Table (1) together with their means and standard deviation. This study examined the concentrations of Cu, Zn, Cd and Pb in *T. thynnus*, *S. salpa* and *T. trachurus* from Sabratha which are located on the western coastal waters of Libya. Fish are able to closely regulate internal Cu concentrations.

4.1. Copper

The concentrations of Cu levels in muscle tissues were 133.6±0.4 µg/g for *S. salpa*, 131.03±0.4 µg/g for *T. thynnus* and 106.2±0.1 µg/g for *T. trachurus* , respectively. Ackacha et al. (2010) studied the heavy metal concentrations in muscles some of fish species from Tripoli coast of Libya. Ackacha et al. (2010) results showed higher metal concentrations (204.3-430 µg/g) compared with our study. Agusa et al. (2005) studied the heavy metal concentrations in different fish species from different coastal sites of Malaysia in 2005. The values of Cu in fish samples obtained in the present study were also higher than those reported by Agusa et al. 2005 (2.53–3.41µg/g and 1.83–2.54) for fishes sampled from two stations Mersing and Port Dickson, Eastern and Western costal water of Peninsular Malaysia respectively. On the other hand, the Cu concentrations (0.26-0.82) in fish muscles collected from Tuzla lagoon, Turkey in investigated conducted by Dural et al. (2006) were lower than our results in this study. Copper levels in the liver in the present study were higher than the literature (0.35-12.03).

4.2. Znic

Muscle tissues of the three species presented Zn levels under the permissible level presented by WHO but higher than the permissible level presented by FAO. Mean Zn concentrations in the muscles were 133.6±0.4 µg/g for *S. salpa*, 131.03±0.4 µg/g for *T. thynnus* and 106.2±0.1 µg/g for *T. trachurus,* respectively. However, Zn levels in liver of *T. thynnus* were 217.37±8.4 µg/g dry wt. Moreover, mean Zn concentrations in gills of *T. trachurus* were 388.5±0.4 µg/g dry wt. A lot of studies indicated that different concentrations of heavy metals in different fish species might be a result of different ecological needs, metabolism, and feeding pattern (Yilmaz, 2003). Moreover, metal accumulation may depend on the seasonal variation (Deram et al. 2006). Levels of the essential metals in the fish samples were higher than those of the non-essential metals. Among the four metals under study, Zn showed the highest level of accumulation. From the literature studies, a study was conducted in Northeast Mediterranean by Canli & Atli (2003) was showed lower levels for Zn for muscles (16.48-37.39) and liver (68.99-110) than the cueernt study. Anther study was conducted in El-Khoms coast of Libya by Matwally & Fouad (2008), results was showed Zn concentrations in muscles (5.56-27.77 µg/g) and its concentration in liver which low so far than our study.

4.3. Cadmium

The Cd concentrations are generally low in all the samples. The highest Cd levels in muscles of *S. salpa* was (0.15±0.01 µg/g) in this research were less than the values reported by WHO and FAO. And The highest Cd levels in liver was (0.63±0.15µg/g) in *T. trachurus*, whereas the highest Cd concentration in gills was in *T. thynnus* (0.29±0.1µg/g). Ackacha et al. (2010) results showed higher metal concentrations in muscles of some fish species (0.07-1.21µg/g) compared with our study. Anther study was conducted in El-Khoms coast of Libya by Matwally & Fouad (2008) was showed similar Cd concentrations in muscles (0.04-0.17 µg/g) with our study but lower concentration in liver (0.17-0.28 µg/g) than our study (0.31-0.63 µg/g). One more study was conducted in El-Mex, Egypt Coast by Abdallah (2008) showed higher Cd concentrations in muscles of some marine fish (0.162.93µg/g) compared with our study.

4.4. Lead

Muscle tissues of the three species presented Pb levels higher than the permissible level presented by WHO and FAO. The findings of this study showed that, The highest Pb concentration was in muscles of *S. salpa* (4.77±0.01µg/g), whereas the lowest Pb concentration was in muscles of *T. trachurus* (1.30±0.1µg/g). The highest Pb concentration was in liver of *S. salpa* (7.64±0.04µg/g), whereas the lowest Pb concentration was in muscles of *T. trachurus* (1.69±0.03µg/g). The highest Pb concentration was in gills of *T. trachurus* (10.59±0.04µg/g), whereas the lowest Pb concentration was in muscles of *T. thynnus* (2.06±0.05µg/g). Agusa et al. (2005) found results for Pb in fishes caught from Terengganu on the eastern cost of Malaysia lower than our results in muscle (0.028-0.063) and (0.33-0.41) in liver. Anther study was conducted in El-Khoms coast of Libya by Matwally & Fouad (2008) was showed lower Pb concentrations in muscles (0.01-0.04µg/g) and liver (1.32-1.94 µg/g) than our study.

Cadmium and Pb have higher tendencies to bioaccumulate in the fish liver tissues which involves in the detoxification process. The sequence of metal contents in various organs of fish identified was: gills > liver > muscle. A pattern of significantly higher heavy metal accumulation in gills than in the muscle was observed in many studies (Dural et al. 2006; Storelli et al. 2006). In the present study, liver had significantly higher heavy metal concentrations than the gills and muscle. The difference in accumulation potential between Muscle, liver, and gills can be explained by the activity of metallothioneins, proteins that are present in liver but not in the muscle, which have the ability to bind certain heavy metals and thus allow the tissue to accumulate them at a high degree (Iwegbue, 2008). Meanwhile fish liver acts as major site for homeostasis (Reynders et al 2006). Gills are an important route of metal uptake and thus gills metal levels can be used to assess metal exposure (Catsiki and Strogyloudi, 1999). On the other hand, lowest concentrations of metals among all the tissues were found in muscle. Muscles analyses are used to investigate the direct transference of heavy metals into human bodies, as the muscles are the main edible part of the fishes and major target of metal storage. Therefore, studying the level of metal accumulation constitutes a tool for environmental assessment and for determining public health risk (Reinfelder et al., 1998).

5. CONCLUSION

The bioaccumulation was quite evident for Pb among the examined fish species. The descending order of metal levels obtained for liver, muscle and gills content was Zn>Cu>Pb>Cd. Zinc concentrations were the highest in all organs of all examined species from the study area. The highest metal concentrations were found in the liver and gills, while the muscle tended to accumulate less metal. Moreover, The findings showed that the Muscle tissues of the three species presented Zn levels under the permissible level presented by WHO but higher than the permissible level presented by FAO. Moreover, levels of Cu in the muscles of *T. thynnus* and *T. trachurus* were higher than the permissible level presented by WHO and FAO. Furthermore, the the levels of Pb in the muscles of *T. thynnus, S. salpa* and *T. trachurus* were were higher than the permissible level reported by WHO and FAO. These results can also be used to understand the chemical quality of fish and to evaluate the possible risk associated with their consumption. The heavy metal concentrations in the majority of the samples analyzed were higher than the prescribed limits set by various authorities.

6. ACKNOWLEDGEMENTS

I am grateful to staffs of the laboratory of sciences faculty who helped me to achieve my analysis.

7. REFERENCES

Abdallah, M. A. M. (2008). Trace Element Levels in Some Commercially Valuable Fish Species from Coastal Waters of Mediterranean Sea, Egypt. *Journal of Marine Systems* 73(1–2): 114-122.

Abida, B., HariKrishna S. and Irfanulla K. (2009), Analysis of heavy metals in water and Sediments samples of Madivala Lakes of Bangalore, Karnataka International Journal of ChemTech Research, 1(2): 245-249.

Ackacha, M. A., Khalifa, K. M., Hamil, A. M., Al-Houni, A. Q. A. (2010). Determination of Heavy Metals in Fish Species of the Mediterranean Sea (Libyan coastline) Using Atomic Absorption Spectrometry. *International Journal of Chem. Tech. Ressearch* 108(2): 794-800.

Agusa, T., Kunito, T., Yasunaga, G., Iwata, H., Subramanian, A., Ismail, A. & Tanabe, S. (2005). Concentrations of Trace Elements in Marine Fish and Its Risk Assessment in Malaysia. *Marine Pollution Bulletin* 51(8–12): 896-911.

Ahmad, A. K. & Shuhaimi-Othman, M. 2010. Heavy Metal Concentrations in Sediments and Fishes from Lake Chini, Pahang, Malaysia. *Journal of Biological Sciences* 10(2): 93-100.

Aladaileh, S. H., Khafaga, A. F., El-Hack, M. E. A., Al-Gabri, N. A., Abukhalil, M. H., Alfwuaires, M. A. (2020). Spirulina platensis ameliorates the sub chronic toxicities of lead in rabbits via anti-oxidative, anti-inflammatory, and immune stimulatory properties. *Sci.Total Environ*.701:134879.

Bellinger, D. C., Stiles, K. M. and Needelman, H. L., (1992). Low-level lead exposure, intelligence and cademic achievement: a long-term follow-up study. Pediatrics 90, 855–861.

Canli, M. and G. Atli, (2003) “The relationships between heavy metal (Cd, Cr, Cu, Fe, Pb, Zn) levels and the size of six Mediterranean fish species,” *Environmental Pollution*, vol. 121, no. 1, pp. 129–136.

Castro-González, M.I. and Méndez-Armenta, M., (2008). Heavy metals: implications associated to fish consumption. *Environmental Toxicology and Pharmacology* 26: 263–271

Catsiki A. V. and Strogyloudi E. (1999). Survey of metal levels in common fish species from Greek waters. *Sci Total Environ* 237(238):387–400

Demirezen, D., and Uruc, K. (2006). Comparative study of trace elements in certain fish, meat and meat products. *Meat Sci*. 74, 255–260.

Deram A, Denayer FO, Petit D and Van Haluwyn C. (2006). Seasonal variations of cadmium and zinc in Arrhenatherum elatius, a perennial grass species from highly contaminated soils. *Environ Pollut* 140:62–70.

Domingo, J. L., Bocio, A., Falcó, G. & Llobet, J. M. (2007). Benefits and Risks of Fish Consumption: Part I. A Quantitative Analysis of the Intake of Omega-3 Fatty Acids and Chemical Contaminants. *Toxicology* 230(2–3): 219-226.

Dural, M., Goksu, M. Z. L., Ozak, A.A., Derici, B. (2006). Bioaccumulation of some heavy metals in different tissues of Dicentrarchus labrax L, 1758, *Sparus aurata* L, 1758, and *Mugil cephalus* L, 1758 from the Camlik Lagoon of the eastern coast of Mediterranean (Turkey), *Environ. Monit. Assess*. 118: 65–74.

Enkeleda, Luljeta D, Jetmira A, Muhamet Z, Imer H, Dariana B, Fatgzim L.C. (2013). Heavy metals in fish for public consumption an consumer protection, Natura Montenegrina, Podogorica. 9(3): 843-851.

FAO.(1983). Compilation of legal limits for hazardous substances in fish and fishery products. FAO Fish Circ, 464, 5.

Farkas, A. and S. J. Specziar,(2003). "Age and size specific patterns of Heavy metals in the organs of fresh water fish *Abramisbrama*L. populating a low-contaminated site," *Water Research*, vol. 37, pp. 959–964.

Gwalteney-Brant, S.M., (2002). Heavy metals. In: Haschek, W. M., Rosseaux, C. G., Wallig, A.M. (Eds.), Handbook of Toxicologic Pathology. Academic Press, New York, pp. 701–732.

Hogstr C., and Haux C, (2001). Binding and detoxification of heavy metals in lower vertebrates with reference to metallothionein. *Compd. Biochem. Physiol*. C 100, 137.

Houston, M. C., (2007). The role of mercury and cadmium heavy metals in vascular disease, hypertension, coronary heart disease, and myocardial infraction. *Altern. Ther. Health Med*. 13, S128–S133.

Iwegbue, C. M. A., (2008). "Heavy metal composition of livers and kidneys of cattle from southern Nigeria," *VeterinarskiArhiv*, vol. 78, no. 5, pp. 401–410.

Jarup, L., (2003). Hazards of heavy metal contamination. British Medical Bulletin 68: 167-182.

Keke, U. N., Arimoro, F. O., Ayanwale, A. V., and Aliyu, S. M. (2015). Physicochemical parameters and heavy metals content of surface water in downstream Kaduna River, Zungeru, Nigerstate, Nigeria. *Appl. Sci. Res. J*. 3,46–57.

Kelle, H. I., Ngbede E. O., Oguezi V. U. and Ibekwe F. C. (2015). Determination of heavy metals in fish (Clarias gariapinus) Orgas from Asaba Major Markets, Delta State, Nigeria. *American Chemical Science Journal*. 5(2): 135-147.

Mehana, E. E., Khafaga, A. F., Elblehi, S. S., Abd El-Hack, M. E., Naiel, M. A., Bin-Jumah, M. (2020). Biomonitoring of heavy metal pollution using acanthocephalans parasite in ecosystem :an updated overview. Animals, 10:811.

Metwally, M. A. A. and Fouad, I. M. (2008). Biochemical changes induced by heavy metal pollution jn marine fishes at Khomse Coast, Libya. *Global veterinaria* 2 (6), 308-311.

Padrilah, S. N., Shukor, M. Y. A., Yasid, N. A., Ahmad, S. A., Sabullah, M. K., and Shamaan, N. A. (2018). Toxicity effects of fish histopathology on copper accumulation. *Pertanika, J. Trop. Agric. Sci*., 519–540.

Reinfelder, J. R, Fisher N. S, Luoma S. N, Nichols J. W and Wang W. X., (1998). Trace element trophic transfer in aquatic organisms: A critique of the kinetic model approach. *Science of the Total Environment*, 219, 117– 135.

Reynders, H, K. van Campenhout, L. Bervoets, W. M. De Coen, and R. Blust, (2006). "Dynamics of cadmium accumulation and effects in common carp (Cyprinuscarpio) during simultaneous exposure to water and food (Tubifextubifex)," *Environmental Toxicology and Chemistry*, vol. 25, no. 6, pp. 1558–1567.

Rietzler A.C., Fonseca A. L. and Lopes G.P. (2001). Heavy metals in tributaries of Pampulha reservoir. Minas. Gerais. *Braz.J. Biol*.**61**, 363.

Storelli, M. M., Barone, G., Storelli, A. & Marcotrigiano, G. O. (2006). Trace metals in tissues of mugilids (*Mugil auratus*, *Mugil capito*, and *Mugil labrosus*) from the Mediterranean Sea. *Bulletin of Environmental Contamination and Toxicology* 77(1): 43-50.

Tarrio J., Jaffor M. and Ashraf M., (1991). Levels of selected heavy metals in commercial fish from five fresh water lake Pakistan. *Toxicology and Environmental Chemistry*.**33**, 133.

Uysal, K., Emre, Y. & Köse, E. (2008). The Determination of Heavy Metal Accumulation Ratios in Muscle, Skin and Gills of Some Migratory Fish Species by Inductively Coupled Plasma-Optical Emission Spectrometry (ICP-OES) in Beymelek Lagoon (Antalya/Turkey). *Microchemical Journal* 90(1): 67-70.

Yilmaz A. B. (2003). Levels of heavy metals (Fe, Cu, Ni, Cr, Pb, and Zn) in tissue of Mugil cephalus and Trachurus mediterraneus from Iskenderun Bay, Turkey. *Environ Res* 92:277–281.