SK Mavuti
Department of Animal Sciences, School of Agricultural Sciences and Agribusiness, Pwani University, Kilifi, Kenya

JM Mbaria
Department of Public Health, Pharmacology and Toxicology, Faculty of Veterinary Medicine, University of Nairobi, Kangemi Nairobi, Kenya

JG Maina
Department of Animal Production, Faculty of Veterinary Medicine, University of Nairobi, Kangemi Nairobi, Kenya

PG Mbuthia
Department of Veterinary Pathology, Microbiology and Parasitology, Faculty of Veterinary Medicine, University of Nairobi, Kangemi Nairobi, Kenya

RM Waruiru
Department of Veterinary Pathology, Microbiology and Parasitology, Faculty of Veterinary Medicine, University of Nairobi, Kangemi Nairobi, Kenya

Corresponding Author:
SK Mavuti
Department of Animal Sciences, School of Agricultural Sciences and Agribusiness, Pwani University, Kilifi, Kenya

Levels of lead, mercury and cadmium in farmed *Oriochromis niloticus* and *Clarias gariepinus* in Nyeri county, Kenya

SK Mavuti, JM Mbaria, JG Maina, PG Mbuthia and RM Waruiru

DOI: https://doi.org/10.22271/fish.2021.v9.i4c.2540

Abstract
This study evaluated the presence of selected heavy metals in farmed tilapia and catfish) in Nyeri County, Kenya.
Live fish comprising of 17 catfish and 33 tilapias were purchased from 15 fish farmers with 31(62%) caught from earthen ponds while 19 (38%) were from liner ponds. The fish were dissected to obtain muscle tissue samples for analysis for levels of lead, mercury and cadmium residues using the Flame Atomic Absorption Spectrometry.
Tilapia fish accumulated higher concentrations of lead, mercury and cadmium in the muscles compared to catfish. The difference of lead and mercury ion concentration between fish species was not significant (p>0.05). Fish obtained from earthen ponds recorded a higher mean concentration of the three heavy metals compared to those from liner ponds.
Mercury concentration in fish muscles was slightly higher than European Union limits (0.1 Mg/Kg) but lower than codex alimentarius (0.5 Mg/Kg) limits.
There is need to continuously monitor levels of heavy metals especially mercury in farmed fish tissues in Nyeri County and other peri-urban areas in Kenya.

Keywords: catfish, fish muscle, ponds, tilapia

Introduction
The fish farming industry in Kenya is faced with challenges which include: inadequate knowledge on fish farm management, environmental and nutritional requirements for optimal productivity among others [1]. Continuous monitoring besides understanding of the chemical environmental conditions within the fish pond, is a desirable requirement to a successful fish pond management strategy. Water is very important to fish farming as farm land is to crop agriculture since fish perform all their body functions in water. Stress to the fish which occurs when they are subjected to poor environmental condition is greatly reduced when good management of water quality is practiced [2].

The main sources of contamination in fish pond water are pathogenic microbes, heavy metals, organic and inorganic chemicals [3]. Semi-treated effluents from agro based industries like sugar, paper, coffee, tea, dairy and fish tanneries are discharged to rivers [4] which are sources of water for fish ponds.

[5] observed that heavy metals such as mercury, cadmium, chromium, zinc, copper, manganese, cobalt, nickel, iron, vanadium and molybdenum cause heavy pollution especially in river systems, fish ponds and lakes in zones where industries release effluents into sewage and agricultural drains. Some of the metals like copper, cobalt, zinc, nickel and molybdenum are naturally present in low concentrations in soils and water.

Inadequate fish farmers' management practices influence types and quantities of chemical pollutants found in water and fish. Water fed into fish ponds contain heavy metals and agrochemicals from run-off waters, sewage, effluents and domestic waste [6]. Their presence in water and farmed fish may affect fish quality and human health.

Studies to analyze presence of heavy metals and chemicals in fish tissue samples have been done in Lake Victoria [7, 8, 9] but there is limited analysis from farmed fish.

Tetu and Nyeri Central sub-counties are in the agro-ecological zone II where farmers grow tea, coffee and other crops. Dairy farming and fish keeping are also practiced. Fish ponds in the two sub-counties are fed with water from rivers, streams and water pans. Water used to replenish these fish ponds is obtained from the same sources. Sewage from the municipality's effluents (slums) and domestic waste end up in some or all of these rivers thus polluting them. The same water may end up into the ponds.

There was need to assess the levels of pollution with heavy metals of selected fish ponds in the two sub-counties to ascertain the suitability of such ponds for fish farming.

Materials and Methods
Fish tissue samples (n=50) were collected from all purchased fish during postmortem and analyzed for presence of heavy metal residues. This was done using the Flame Atomic Absorption Spectrometry at Kenya Plant Health Inspectorate Service (KEPHIS) analytical chemistry laboratory. Contamination levels of lead, mercury and cadmium were also analyzed.

Analysis for lead and cadmium
This was done at KEPHIS using the lead and cadmium method (Graphite Furnace Atomic Absorption Spectrometry). One gram (1g) of fish tissue (muscle) was weighed into ultra clean and dry inert polymeric microwave vessels. This was followed by slow addition of 9 ml of concentrated nitric acid (under the fume chamber) into each microwave vessels. Two (2) ml of hydrogen peroxide were then added slowly into the microwave vessels. One (1) ml of de-ionized water was then added and the sample left to react for approximately 6 minutes prior to sealing the vessel in order to allow any accumulated gases to escape. The vessels were then sealed, placed on the rotor and placed in the microwave. The microwave vessels were then heated gradually to 180ºC for not less than 10 minutes and then held at that temperature for 10 minutes after which the microwave automatically begins to cool the sample until it indicates when cooling process is complete. After cooling, the samples were quantitatively placed into 100 ml volumetric flasks and topped to the mark with de-ionized water. The samples were analyzed using an instrument utilizing the Graphite Furnace Atomic Absorption Spectrometry [10].

Stock solutions for each metal were prepared by dissolving 1g of pure lead and cadmium metal strip in 1:1 nitric acid and diluted to 1 liter to give a stock solution of 1000 ppm. For each of the stock solutions working standards of 1 Mg/Kg, 5 Mg/Kg, and 10 Mg/Kg were made and calibration curves plotted. Metal concentrations were calculated using a standard curve generated using 1 Mg/Kg, 5 Mg/Kg, and 10 Mg/Kg, and the results were expressed in µg/ Kg of dry tissue weight.

Analysis for mercury
This was done at KEPHIS using the mercury method (Cold Vapour Atomic Absorption Technique). One gram of fish muscle was weighed into ultra clean and dry inert polymeric microwave vessels. This was followed by slowly adding 9 ml of concentrated nitric acid (under the fume chamber) into each microwave vessel. Two (2) ml of hydrogen peroxide were then added slowly into the microwave vessels. One

milliliter of de-ionized water was then added and the sample left to react for approximately 6 minutes prior to sealing the vessels in order to allow any accumulated gases to escape. The vessels were then sealed, placed on the rotor and placed in the microwave. The microwave vessels were heated gradually to 180 ºC for not less than 10 minutes and then held at that temperature for 10 minutes after which the microwave automatically begins to cool the sample until it indicates when cooling process is complete. After cooling, the samples were quantitatively placed into 100 ml volumetric flasks. Sixty (60) ml of potassium permanganate solution (6% v/v) were added. This was followed by addition of 6 ml of potassium persulphate (5% solution w/v). The sample was left with the caps not tightened for a minimum of 2 hours. Hydroxylamine hydrochloride solution (20% m/v) was carefully added to remove the excess potassium permanganate. De-ionized water was added to bring the sample volume to 100 mls. The resulting solution was thoroughly mixed and analyzed using an instrument utilizing the vapor generation technique [11]. Stock solution for mercury was prepared by dissolving 1g in 1:1 nitric acid and diluted to 1 liter to give a stock solution of 1000 ppm. Working standards of 1 Mg/Kg, 5 Mg/Kg, and 10 Mg/Kg were made and calibration curves plotted. Mercury concentration was calculated using a standard curve generated using 1 Mg/Kg, 5 Mg/Kg, and 10 Mg/Kg, and the results were expressed in µg/ Kg of dry tissue weight.

Data analysis
Presence of heavy metals in fish muscles was assessed using chi-square to get their variation between pond types and different fish species. Different range tests were deployed to compare means of parameters between different pond types according to [12].

Results
The heavy metal concentration detected ranged between 0.092-0.659 Mg/Kg for lead, 0.061-0.306 Mg/Kg for cadmium and 0.026-0.134 Mg/Kg for mercury, respectively. Tilapia accumulated higher concentrations of lead (0.679 Mg/Kg), mercury (0.306 Mg/Kg) and cadmium (0.134 Mg/Kg) in the muscles compared to catfish. Data obtained showed that lead had the highest mean concentration of 0.326 Mg/Kg followed by mercury (0.165 Mg/Kg) and cadmium (0.069 Mg/Kg) (Table 1).

Fish obtained from earthen ponds recorded a higher mean concentration (lead 0.346 Mg/Kg; mercury 0.171 Mg/Kg; cadmium 0.083 Mg/Kg) compared to fish from liner ponds (lead 0.302 Mg/Kg; mercury 0.158 Mg/Kg; cadmium 0.051 Mg/Kg). Cadmium ion concentration varied significantly (p=0.000) between the two fish pond types. There was no significant difference between lead (p=0.206) and mercury (p=0.516) ion concentration in the two fish pond types (Table 1). The difference of lead (p=0.120) and mercury (p=0.528) ion concentration between fish species was not significant but was significantly different for cadmium (p=0.000).

Nyeri Central Sub-county recorded a slightly higher lead and mercury concentration in fish muscles than Tetu Sub-county with an equal concentration for cadmium being observed in both sub-counties. There were no significant differences in the concentrations of lead (p=0.861), mercury (p=0.792) and cadmium (p=0.966) among the sub-counties.

Table 1: Levels of lead, cadmium and mercury concentration in fish muscles from two fish species and pond types in Tetu and Nyeri Central sub-counties

Heavy metal	Lead (Pb) Mg/Kg	Cadmium (Cd) Mg/Kg	Mercury (Hg) Mg/Kg
Fish pond type			
Earthen	0.346	0.083	0.071
Liner	0.302	0.051	0.158
Fish species			
Tilapia	0.346	0.082	0.169
Catfish	0.289	0.043	0.157
Sub-county			
Tetu	0.323	0.069	0.163
Nyeri Central	0.329	0.069	0.168
P - Value			
Fish pond type	0.206	0	0.516
Fish species	0.12	0	0.528
Sub-county	0.861	0.966	0.792

Key: Mg/Kg: Milligrams per Kilogram

Discussion

Fish muscle tissues were analyzed for the presence of three heavy metal elements namely lead, cadmium and mercury. Lead had a mean concentration of 0.326 ppm (range 0.092-0.659 ppm) followed by mercury 0.165 ppm (0.061-0.306 ppm) with cadmium having the lowest mean concentration of 0.069 ppm (range 0.026-0.134 ppm). The most likely source of these heavy metals in fish muscles was water from storm runoffs and drainage which may enter into water bodies like rivers that serve as fish pond water sources.

The highest concentration of lead (0.679 ppm), mercury (0.306 ppm) and cadmium (0.134 ppm) was observed in tilapia muscles relative to catfish which had lower concentrations of the three heavy metals. Earlier studies in fish have indicated that muscle is not an active tissue in accumulating heavy metals [13, 14, 15]). The muscle tends to accumulate fewer metals relative to the other fish organs due to the presence of mucous layer on the fish skin which forms complexes with the heavy metals and prevents their entry into fish muscle [16, 17, 18]). Tilapia are covered by scales with less mucous on their skin compared to the slimy catfish that are covered by more mucous. This probably can be attributed to the lower heavy metal concentrations in catfish than in tilapia in the present study.

The mean lead concentration in fish muscles (0.326 ppm) was lower than the maximum permissible limits stipulated by [19] of 0.5 ppm for lead but within European Union and codex alimentarius limits of 0.3 ppm. [20] reported a mean concentration of 3.78 ppm and 12.22 ppm for lead in tilapia muscles in Kiambu and Machakos counties, respectively. The higher levels of lead reported in his study favourably compared to reports by [21] that indicated mean lead concentrations of 30.7 mg/L in sediments in Tanzania. [22] reported lead levels that were higher than 0.5 ppm (26.8 ppm) in contrast to the present study. It was observed that there was no significant difference (p=0.120) in the mean lead concentrations among the fish species in this study.

The mean mercury concentration in fish muscles (0.165 ppm) was slightly higher than the maximum permissible limits stipulated by European Union of 0.1 ppm but lower than 0.5 ppm codex alimentarius limits. [23] in Croatia reported mercury concentrations ranging from 0.024 to 0.308 ppm. Hygienic limit of mercury content in fish meat in the Republic of Croatia is 0.5 ppm while most European countries have a limit that ranges from 0.05 ppm to 0.07 ppm, except from the Czech Republic where the limit is 0.1 ppm in fish meat [24].

The mean cadmium concentration in fish muscles (0.069 ppm) was lower than that observed by [22] of 1.79 ppm or [20] of 1.66ppm in Kiambu and 1.12 ppm in Machakos. The mean cadmium concentrations in this study slightly exceeded the maximum permissible limits stipulated by [19] of 0.05 ppm but were within codex alimentarius (0.3 ppm) and European Union (0.1 ppm) limits.

Fish obtained from earthen ponds recorded a higher mean heavy metal concentration (lead 0.346 ppm; mercury 0.171 ppm and cadmium 0.083 ppm) compared to fish from lined ponds (lead 0.302 ppm; mercury 0.158 ppm and cadmium 0.051 ppm). Cadmium ion concentration varied significantly (p=0.000) between the two fish pond types. However, no significant difference for lead (p=0.206) and mercury (p=0.516) was noted in the present study.

Most of the earthen ponds were constructed on a wetland with a stream entering and exiting the pond. Surface runoffs and drainage waters contaminated with lead, mercury and probably higher concentrations of cadmium most likely entered into earthen compared to liner ponds where enhanced management practices (i.e., raising pond edges and weed control) were undertaken to reduce pond pollution.

Nyeri Central Sub-county recorded a slightly higher lead and mercury concentration in fish muscles than Tetu Sub-county. Nyeri Central is an urban sub-county and effluent from the municipality may be discharged into the rivers that feed the fish ponds. This is unlike Tetu which is a rural sub-county.

Conclusion and recommendations

This study has shown that mercury concentration in fish muscles was slightly higher than European Union limits while lead concentration was within European Union and codex alimentarius limits in farmed tilapia and catfish from Nyeri County.

There is need for continuous monitoring of levels of heavy metals especially mercury in farmed fish tissues in Nyeri County and other peri-urban areas in Kenya.

References

1. Munguti JM, Kim JD, Ogello EO. An Overview of

Kenyan Aquaculture: Current Status, Challenges, and Opportunities for Future Development. Journal of Fisheries and Aquatic Sciences 2014;17(1):1-11.
2. Fisheries Maestro. Guide on Best Management Practices in Fish Farming in Nigeria Series 5 2014.
3. Sosbey MD. Managing water in the home. Accelerated health gains from improved water supply, World Health Organization sustainable development and healthy environment. World Health Organization. Geneva 2002.
4. Nzomo R. Sustainable management of African lakes-The case of Lake Victoria. A paper presented in the first living lakes African Regional Conference on 27th -30th October 2005, at Imperial Hotel, Kisumu, Kenya 2005.
5. Ida CJ. Heavy Metals in Suchindramkulam (a Lentic Water Body) of Kanyakumari District, Tamil Nadu, India. Journal of Tropical and Experimental Biology 2012;8(3 and 4):141-145.
6. Mavuti SK, Waruiru RM, Mbuthia PG, Maina JG, Mbaria JM. Evaluation of fish farmer management practices in Nyeri County, Kenya. International Journal of Fisheries and Aquatic Studies 2017;5(3):165-170
7. Onyari JM, Wandiga SO. Distribution of Cr, Pb, Cd, Zn, Fe and Mn in Lake Victoria sediments, East Africa. Bulletin Environmental Contamination Toxicology 1989;42:807-813.
8. Kasozi GN. Analysis of organochlorine pesticide residues in the aquatic ecosystem of Lake Victoria. M. Sc. Thesis, Department of Chemistry, Makerere University 2001.
9. Madadi OV, Wandiga SO, Jumba IO. The status of persistent organic pollutants in Lake Victoria catchment. In: Odada, Eric, O., Daniel, O. (Eds.), Proceedings of the 11th World Lakes Conference 2006;2:107-112.
10. United States Environmental Protection Agency 1998 EPA Method 3051A. Microwave assisted acid digestion of sediments, sludge, soils and oils. US EPA Office of Solid Waste. Washington D.C 1998.
11. United States Environmental Protection Agency. Office of Solid Waste and Emergency Response. Test Methods for Evaluating Solid Waste, SW-846, 3rd ed. Update II. Method 7471A 1994.
12. Steel RDG, Torrie D. Principles and procedures of statistics, MCGraw – Hill Book Co. New York 1980, 269p.
13. Khan AT, Weis JS, D'andrea L. Bioaccumulation of four heavy metals in two populations of Grass Shrimp, (*Palaemonotes pugio*). Bulletin of environmental contamination and toxicology 1989;42:339-343
14. Karadede H, Unlu E. Concentrations of some heavy metals in water, sediment and fish species from the Ataturk Dam Lake (Euphrates), Turkey, Chemosphere 2000;41(9):1371-1376.
15. Alam MGM, Tanaka A, Allison G, Laurenson LJB, Stagnitti F, Snow ET. A Comparison of trace element concentrations in cultured and wild carp (*Cyprinus carpio*) of Lake Kasumigaura, Japan. Ecotoxicology and Environmental Safety 2002;53:348-354.
16. Schlenk D, Benson WH. Target organ Toxicity in Marine and Freshwater Teleosts, United States of America: CRC press 2001,198-199p.
17. Altindag A, Yigit S. Assessment of heavy metal concentrations in the food web of lake Beysehir, Turkey. Chemosphere 2005,552-555.
18. Uysal K, Emre Y, Kose E. 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 2008;90:67-70.
19. Food and Agriculture Organization of the United Nations. Codex Alimentarius Commission, standard program codex committee on food additives and contaminants 24th Session, Hague 1992.
20. Omwenga IM. Assessment of Pesticide and Heavy Metal Residues in Tilapia Fish from Machakos and Kiambu Counties, Kenya A MSc Thesis Department of Public Health, Pharmacology and Toxicology University of Nairobi 2013.
21. Kishe MA, Machiwa JF. Distribution of heavy metals in sediments of Mwanza gulf of Lake Victoria, Tanzania. Environment international Journal 2003;28:619-625
22. Hounkpatin AS, Edorh A, Patrick, Y, Salifou S, Gnandi K, Koumlou L *et al*. Assessment of exposure risk and cadmium via fish consumption in the Lacusrian village of Ganvie in Benin Republic. Journal of Environmental Chemistry and Ecotoxicology 2012;4(1):1-10
23. Bogut I, Has-Schön E, Janson R, Antunović Z, Bodakoš D. Concentrations of Hg, Pb, Cd and as in meat of fish-pond carp (*Cyprinus Carpio*) 2000;17(1):1-3
24. Svobodova Z, Vikusova B, Machova J, Bastl J, Hrbkova M, Svobodnik J. Monitoring of Foreign Substances in Fishes from the Elbe River in the Čelakovice Locality, Bulletin VURH, Vodnany 1993;29:47-61.