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'Burdens' of selected heavy metals in common fish species from specific Kenyan freshwaters

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Abstract
Heavy metals are a group of chemical contaminants available in the environment in different abundancies. Food is the major vehicle for entry into the human system. Fish may contribute a major source of heavy metals in the food chain. Concentration of heavy metals in commercially important species of fish from fish markets in and around the fresh water bodies area were evaluated using a flame atomic absorption spectrometer. A total of twelve fish species were sampled from all regions as follows: two from Lake Turkana, six from Lake Victoria and four from river Tana were analysed. The concentration ranges of Zn, Ni, Cu, Pb, and Cd in the samples were 0.1703 – 0.6672, 0.2661 – 0.5073, 0.0104 – 0.419, 0.1831 – 0.327 and 0.0058 – 0.0404 mg/kg, respectively. This study showed that metals were present in the samples at different levels but below the maximum residual levels from EU and USFDA and hence fish from these areas are safe for human consumption.

Keywords: Freshwater; Heavy metals; Fish species

1. Introduction
The contamination of freshwaters with a wide range of pollutants has become a matter of great concern over the last few decades. Heavy metals are natural trace components of the aquatic environment, but their levels is increasing due to domestic, industrial, mining and agricultural activities [1, 2]. Discharge of heavy metals into river or any aquatic environment can change both aquatic species diversity and ecosystems, due to their toxicity and accumulative behaviour [3]. Aquatic organisms such as fish and shell fish accumulate metals to concentrations many times higher than present in water or sediment [4, 5]. They can take up metals concentrated at different levels in their different body organs [6]. Certain environmental conditions such as salinity, pH and water hardness can play an important role in heavy metals accumulation in the living organisms up to toxic concentrations and cause ecological damage [7]. Thus, heavy metals acquired through the food chain as a result of pollution are potential chemical hazards threatening consumers. At low levels, some heavy metals such as copper, cobalt, zinc, iron and manganese are essential for enzymatic activity and many biological processes. Other metals, such as cadmium, mercury, and lead have no known essential role in living organisms, and are toxic at even low concentrations. The essential metals are also toxic at high concentrations [8]. Studies carried out on fish have shown that heavy metals may have toxic effects including altering physiological activities and biochemical parameters both in tissue and in blood of fish [9, 11]. The consequence of heavy metal pollution can be hazardous to man through his food. Therefore, it is important to monitor heavy metal in aquatic environments (water, sediment and biota).
In most parts of Kenya, water resources are scarce and insufficient to meet the growing demands of a rapidly increasing population. As a consequence, the water resources situation is now precarious and of great concern to the Government. All water bodies are looked upon as a source of exploitation for urban, agricultural and industrial uses. Many water bodies are affected by increasing salinity, pollution and eutrophication due to intensive agricultural practices.

2. Methodology
2.1 Fish collection
Sampling was done between January and September 2009 from 3 different freshwater bodies [Lake Turkana (3°31'7.75"N, 35°56'27.59"E), Lake Victoria - Dunga beach (0° 8'42.13"S,

Lake Victoria - Dunga beach (0° 8'42.13"S, 34°44'12.12"E) and river Tana–Garissa town (0°27'29.28"S, 39°37'59.40"E)]. Common fish species found in each region were purchased directly from fishermen, common names were noted and fishes were stored in a cold box at -4 °C to prevent autolytic post-mortem changes after rigor mortis during transportation to the laboratory where they were placed in clean well-labelled polyethylene bags before preserving them in a refrigerator. Although a variety of freshwater fish species inhabits these freshwater bodies, the common ones; a total of twelve fish species were sampled from all regions as follows: two from Lake Turkana (namely Tilapia and Mudfish), six from Lake Victoria (namely Tilapia, Nile perch, Mudfish, Catfish, Labeo and Bonny fish) and four from river Tana (namely Tilapia, Choka, Labeo and Bonny fish) were sampled in the present work. The fish samples were put in a cool box filled with ice and transported to JKUAT laboratory.

2.2 Materials and Methods

The study was conducted at the Jomo Kenyatta University of Agriculture and Technology's Chemistry (JKUAT) Research Laboratory. Sample preparations and analysis were carried out using standard methods of analysis as described in APHA [12] Helrich [13] and [14]. All reagents used were analytical grade reagents. Glassware was soaked in acid overnight before washing them. Distilled and de-ionized water was used throughout the analysis.

2.3 Digestion

5 g of the wet/dry weight of fish muscles from each fish were weighed in a 250 ml conical flask and 20 ml of a mixture of nitric/perchloric/sulphuric acid in the ratio of 3.1.1 respectively was added [15]. Each conical flask was heated at 150 °C until all brown fumes ended down in fume chamber. The digested sample was cooled before adding small amount of distilled water. The sample was filtered into a 100ml volumetric flask using a filter paper (Whatman No. 42). The solution was topped to the mark using distilled water. The resulting solution was then transferred into a clean-labelled polythene bottle then stored in a refrigerator. Each sample was done in triplicate. Method blank was prepared in similar method.

2.4 Analyses of heavy metals through Flame Atomic Absorption Spectrometer (FAAS)

Heavy metals were determined for the digested both wet and dry fish species from lake Victoria, lake Turkana and river Tana and their concentration levels calculated using calibration curves shown below. Cadmium (Cd), copper (Cu), lead (Pb), zinc (Zn) and nickel (Ni) heavy metals in the water samples and various fish species were analysed by use of an atomic absorption spectrophotometer (AAS – Buck Scientific VGP 210 Model). A series of standards were prepared for instrumental calibration by serial dilution of a working solution (100 ppm) prepared from analytical grade 1000 ppm stock solutions from BDH Poole. A standard and blank sample was run after every seven samples to check instrumental drift. The concentrations of the metals were assayed in triplicates and the accuracy of the instrument checked by triplication of samples. The percentage recoveries were determined for all metals and were found to be above 86%. Different stock solutions were prepared

2.5 Data Analysis

The concentrations of heavy metals in various matrices are presented as arithmetic mean with standard deviation [mean ± standard deviation). The results are presented in tables and bar graphs. The data was analysed using arithmetic mean, standard deviation, range, F-test, t-test and one-way ANOVA, all statistical tests were done at P = 0.05 [16].

3. Results & Discussion

Table 1: Physical parameters of different fish species analysed

Fish Species			Physical Parameters			
Scientific name	**Common name**		**Length**	**Width**	**Height**	**Weight**
Labeo gregorii	*Choka*	*Tana*	30.1±1.6	4.7±0.5	7.7±0.5	251.5±44.7
Oreochromis spilurus	*Tilapia*		16.5±0.7	2.9±0.2	6.2±0.2	78.7±6.5
Schilbe intermedius	*Butterfish*		19.4±1.4	2.5±0.3	4.8±0.3	62.2±15.9
Labeo cylindricus	*Labeo*		16.9±0.1	3±0.1	3.8±0.2	46.3±1.9
Protopterus aethiopicus	*Mudfish*	*Victoria*	53.8±1.1	9.7±0.1	7.5±0.2	943.7±43.3
Oreochromis variabilis	*Tilapia*		30.8±0.6	4.8±0.3	12.3±0.8	636.5±51.3
Clarias gariepinus	*Catfish*		53.9±1.1	9.8±0.1	7.6±0.2	953.7±41.7
Chiloglanis somereni	*Labeo*		16.9±0.3	3.4±0.2	4.3±0.6	55.8±12.2
Schilbe intermedius	*Butter Fish*		19.6±1.8	3.2±0.5	4.9±0.4	64.5±24.8
Lates niloticus	*Nile Perch*		36.1±0.6	4.4±0.5	10.2±0.2	495.6±9.0
Protopterus aethiopicus	*Mudfish*	*Turkana*	73.7±11.2	13.9±2.4	7. 5±0.5	1207.7±59.9
Oreochromis niloticus	*Tilapia*		24.4±1.7	3.2±0.4	9.2±0.7	202.4±8.9

Numbers represent means ± standard errors of three replicates

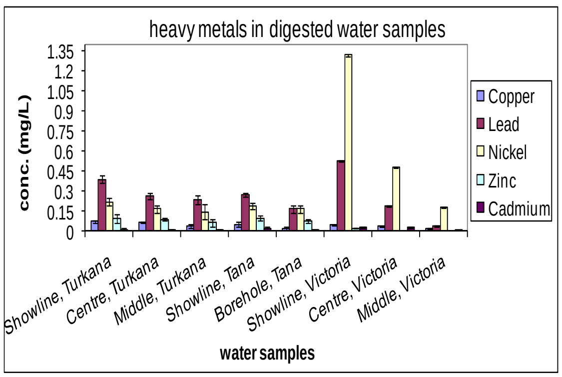
3.1 Heavy Metals

3.1.1 Concentration of Zinc

For the fish samples, zinc concentration ranged between 0.1703 – 0.6672 mg/Kg and a mean of 0.4112 ± 0.1140 mg/Kg. The highest zinc concentration was found in mudfish-Turkana [0.667 mg/Kg] > tilapia-Turkana [0.647 mg/Kg] > butterfish-Victoria [0.593 mg/Kg] > butterfish-Tana [0.586 mg/Kg]. The high zinc content in mudfish (Turkana) could be attributed to its large size due to bioaccumulation of zinc with time and its feeding habits because it feeds on mud (sometimes) which may have deposited zinc in the water. The lowest zinc was recorded in choka (Tana) with 0.1703 ± 0.0176 mg/Kg which could be attributed to the nature of water body which is flowing as opposed to a water reservoir (lake).

The zinc concentration in the samples compares well with the earlier report in grey mullet from the Tigris River [17] Zinc is an essential trace element for both animals and humans. The recommended daily allowance is 10 mg/day in growing children and 15 mg/day for adults [18]. A deficiency of zinc is marked by retarded growth, loss of taste and hypogonadism, leading to decreased fertility. Zinc toxicity is rare but, at concentrations in water up to 40 mg/kg, may induce toxicity, characterized by symptoms of irritability, muscular stiffness and pain, loss of appetite, and nausea [18]. Zinc appears to have a protective effect against the toxicities of both cadmium [19] and lead [20].

For the water samples, Zinc is in the range of 0.073 – 0.1197 mg/L and a mean of 0.0849 ± 0.0107 mg/L. The highest zinc concentration was found at sites in show line –Victoria [0.180 mg/L] > show line-Tana [0.111 mg/L] > show line-Turkana [0.102 mg/L]. The high zinc in show lines indicates inflows from human activities along the water bodies. The mean zinc content in the fresh water bodies is below the maximum permissible limits for irrigation water of 2 mg/L and below maximum allowable limits for sources of domestic water of 1.5 mg/L [21]. The zinc content in show line (Victoria) is higher than maximum allowable limits for effluent discharge into the environment. The concentrations of zinc fall within the ranges of freshwater ecosystems [22] and also below the maximum allowable levels in drinking water [23–25]. Results of the present study are lower than those reported by Kisamo [26] in the Lake Victoria Basin.



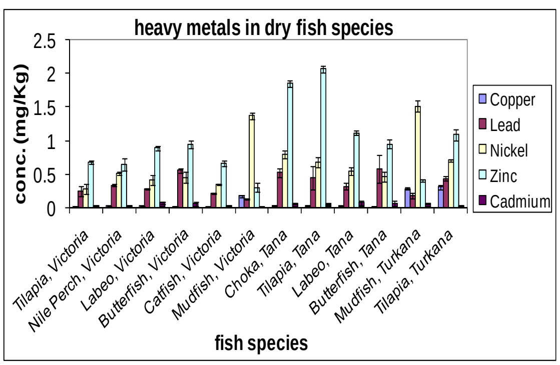
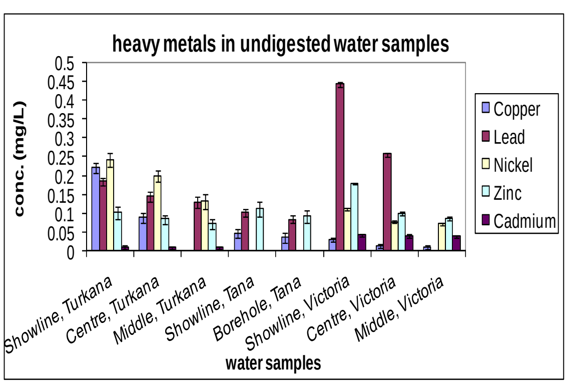


Fig 1: Concentration of heavy metals in both we and dry fish species



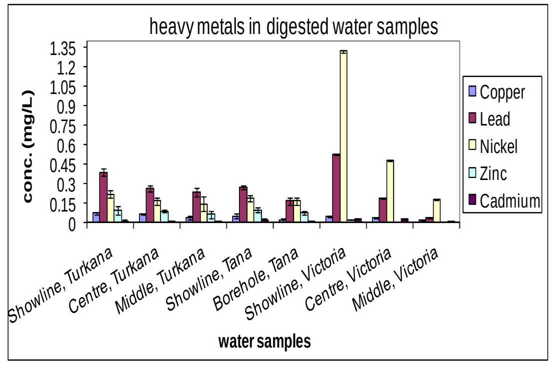


Fig 2: Concentration of heavy metals in both undigested and digested water samples

3.1.2 Concentration of Nickel

For the fish samples, nickel is in the range of 0.2661 – 0.5073 mg/Kg and a mean of 0.3603 ± 0.0827 mg/Kg. The highest nickel concentration was found in tilapia-Turkana [0.507 mg/Kg] > mudfish-Turkana [0.463 mg/Kg] > Nile perch-Victoria [0.444 mg/Kg] > tilapia-Tana [0.380 mg/Kg] > catfish-Victoria [0.368 mg/Kg]. The high nickel content in tilapia (Turkana) could be attributed to location of the lake in the rift valley and nickel is naturally abundant in the earth's crust. The lowest nickel was recorded in Labeo (Victoria) with a mean of 0.2661 ± 0.0372 mg/Kg which could be attributed to its small size compared with other fish species and are normally found in the interior of the lake where pollution is minimal due to dilution effect and bio-extraction by water hyacinth found in abundance in the lake.

The MRL for nickel is 70–80 mg/kg [27], and the samples analysed showed concentrations only up to 0.5073 mg/kg. The major source of nickel for humans is food and uptake from natural sources, as well as food processing [28-30]. The normal range of oral intake of nickel for humans is 300–600 µg/day. An increased incidence of cancer of the lung and nasal cavity has been reported in workers in nickel smelters [28-30].

Water samples had nickel concentration in the range of 0.0714 - 0.2432 mg/L and a mean of 0.1336 ± 0.047 mg/L. The nickel concentrations from various sites show a wide variation; the highest nickel concentrations were found at sites at show line-Turkana [0.243 mg/L] > centre-Turkana [0.199 mg/L] > middle-Turkana [0.133 mg/L] > show line-Victoria [0.113 mg/L].

Lake Turkana had the highest nickel concentration; this could be due to its location in the rift valley exposing most metals found in the earth's crust and show line (Turkana) had the highest among the three Turkana samples due to various man-made activities beside the lake including washing among others. Middle (Victoria) shows the lowest nickel concentration of 0.0714 ± 0.0033 mg/L, this could be due to uptake of nickel by water hyacinth which is thriving at the site. The nickel content in the water is lower than the maximum allowable concentration for nickel in effluent discharge of 0.3 mg/L [21]. Nickel was not detected in water from show line (Tana) and borehole (Tana).

The high nickel concentration agrees with the relatively high contents in the earth's crust and is thought to come from geochemical processes [22, 31]. Results of the present study are higher than those reported by Oching' *et al.* [32], [33] and Kubo [34] indicating increased input of nickel in the catchment. Nickel concentrations in water were within the concentration range reported by Ochieng' *et al.* [32] and fall within the ranges of freshwater ecosystems [22] and also below the maximum allowable in drinking water [23, 25, 35, 36].

3.1.3 Concentration of Copper

For the fish samples, copper is in the range of 0.0104 – 0.419 mg/Kg and a mean of 0.0981 ± 0.0087 mg/Kg. The highest copper concentration was found in tilapia-Turkana [0.419 mg/Kg] > mudfish-Turkana [0.318 mg/Kg] > mudfish-Victoria [0.294 mg/Kg] > Nile perch-Victoria [0.024 mg/Kg]. This high copper content in tilapia (Turkana) could be attributed to location of the lake in the rift valley. The lowest copper was recorded in Labeo (Victoria) with 0.0104 ± 0.0077 mg/Kg which could be attributed to its small size compared with other fish species and are normally found in the interior of the lake where pollution is minimal due to dilution effect and bio-extraction by water hyacinth in the lake.

But the concentrations in the samples were much below the toxic limit of 30 mg/kg [37]. Copper is an essential part of several enzymes and it is necessary for the synthesis of haemoglobin. The richest sources of copper are shellfish, especially oysters and crustaceans [38]. No deficiencies of copper in adults have been reported but, in infants, anaemia and hypoproteinaemia are reported [38].

For the water samples, the copper concentration was in the range of 0.0107 – 0.2217 mg/L and a mean 0.0773 ± 0.0136 mg/L. The highest copper content is found at the show line (Turkana) indicating significant inflows of copper from the numerous activities along the show line of the lake. Middle (Victoria) despite having several canals show low copper content in the water, this could be due to the uptake of the soluble copper by the thriving water hyacinth and other aquatic plants growing in this region. Copper was not detected in water from middle (Turkana).

For each fresh water bodies there is a decreasing trend of concentration from the show line to the middle which could be as a result of dilution effect or removal of copper through adsorption onto particulate matter. Results of the present study are lower than those reported by Ochieng' *et al.* [32], but compare well with those reported by [33] and [26]. The copper content in the samples are within the limits of freshwater ecosystems [22] and are below the maximum acceptable limits in drinking water except for show line [Turkana] which was slightly above [23, 25, 35, 36].

3.1.4 Concentration of Cadmium

For the fish samples, Cadmium is in the range of 0.0058 – 0.0404 mg/Kg and a mean of 0.0193 ± 0.0054 mg/Kg. The highest Cadmium concentration was found in tilapia-Victoria [0.040 mg/Kg] > choka-Tana [0.029 mg/Kg] > butterfish-Victoria [0.026 mg/Kg] = butterfish-Tana [0.026 mg/Kg]. The high Cadmium content in tilapia (Victoria) could be attributed to location of the lake near an industrial town with many industrial waste and sewage effluent. The lowest Cadmium was recorded in mudfish (Turkana) with 0.0058 ± 0.0015 mg/Kg which could be attributed lack of industrial pollution in the area and minimal agricultural activities within the region.

For all the samples the cadmium content was much below the legal limit of 1 mg/kg meat [39]. All the samples were found to contain Cd below the EC limit of 0.1 mg/kg and none was above the FAO limit [37, 40]. Humans are exposed to cadmium through food and the average daily intake for adults has been estimated to be approximately 50 mg [19]. The threshold for acute cadmium toxicity would appear to be a total ingestion of 3–15 mg. Severe toxic symptoms are reported to occur with ingestions of 10–326 mg. Fatal ingestions of cadmium, producing shock and acute renal failure, occur from ingestions exceeding 350 mg [41].

For the water samples, the cadmium concentration was in the range of 0.0091 – 0.0416 mg/L with a mean of 0.0096 ± 0.0014 mg/L. This shows wide variation between samples from different sites, the highest cadmium concentration is found at show line-Victoria [0.042 mg/L] > centre-Victoria [0.040 mg/L] >middle-Victoria [0.039 mg/L]. Cadmium was not detected in any sample collected from Tana River. Show line (Victoria) had the highest concentration of 0.0416 ± 0.0042 mg/L this can be attributed to concentration of the metal by the blue green algae in the Lake. Cadmium concentrations in the water agree with those reported by [33] and Kubo [34]. The concentrations fall within the range of freshwater ecosystems [22] and are below the maximum acceptable levels in drinking water [23, 25, 35, 36]. Results of the present study are in agreement with those reported by Kisamo [26] at the Lake Victoria basin.

3.1.5 Concentration of Lead

For the fish samples, lead is in the range of 0.1831 – 0.327 mg/Kg and a mean of 0.247 ± 0.045 mg/Kg. The highest lead concentration was found in Nile perch-Victoria [0.327 mg/Kg] > choka-Tana [0.292 mg/Kg] > catfish-Victoria [0.287 mg/Kg] > tilapia-Victoria [0.238 mg/Kg]. High concentration of lead in Nile perch (Victoria) could be attributed to location of the lake near an industrial town and full of other anthropogenic activities with many industrial process wastes and sewage effluent discharging to the lake. The lowest lead concentration was recorded in mudfish (Turkana) with 0.1831 ± 0.0569 mg/Kg which could be attributed lack of industrial pollution in the area and minimal agricultural activities within the region.

The highest concentration was detected in one of the twelve samples of Nile perch (Victoria) (0.327 mg/kg). However, all the samples contained lead below 0.4 mg/kg (EC, 2001) as well as below 0.5 mg/kg [37, 40]. Lead causes renal failure and liver damage in humans [42, 43].

For the water samples, lead concentration was in the range of 0.084 – 0.4429 mg/L with a mean of 0.1682 ± 0.0264 mg/L. This shows a wide variation between samples from different sampling sites. The highest lead concentration was recorded at show line-Victoria [0.443 mg/L] > Centre-Victoria [0.257 mg/L] > show line-Turkana [0.185 mg/L] > Centre-Turkana [0.145 mg/L]. The high concentration at show line (Victoria) could be resulting from inflows from surface run-off upstream and numerous human activities going on beside the lake; the high lead concentrations at the show line indicate inflows from sewage. Lead was not detected in the water collected from the middle (Victoria).

Lead concentrations in water from the lake agree with those reported by [33] and Kubo [34] but are lower than those reported by Kisamo [26]. The concentrations fall within the range of freshwater ecosystems [22] and are above the maximum acceptable levels in drinking water [23, 25, 35, 36].

The beneficial health effects of fish are based on several studies from the last 25 years, most of which have linked fish consumption to several health benefits [44-46]. However, some studies have reported contradictory results, since there exists an increased levels of heavy metals that can affect the health of consumers however, fish consumption is recommend due to all its nutritional benefits not only by n−3 fatty acids [47, 48]. However, one potential risk of dietary fish intake is its content of heavy metals, recent evidence suggests that high heavy metal content in fish may diminish the cardio protective effect of fish intake [49]. In humans, cadmium, lead and/or arsenic, have also been associated to serious health effects on adults and children and one source of exposition is the intake of fish with high content of either of these metals. This has been demonstrated by several authors; Burger and Gochfeld [50]; Andreji *et al*. [51]; Falco *et al*. [52]; Has-Schon *et al*. [53] determined the levels of these metals in different fish species which may be out of permissible limits. Provisional tolerable weekly intake (PTWI) for adults or children increased due to fish accumulation of substantial amounts of metals in the fish tissue and thus, risk is also dependent on the different metals residing in different fish. The American Heart Association suggests eating fish at least two times per week in order to reach the daily intake of omega-3 fatty acids recommended for healthy adults with no history of heart disease [46]. In particular fatty fish such as anchovies, bluefish, carp, catfish, halibut, herring, lake trout, mackerel, pompano, salmon, striped sea bass, tuna [albacore), and whitefish, or an equivalent of 0.3–0.5 g/d e PUFAs are also recommended.

The intake of fish should be regulated; information regarding the species of fish consumed and its possible levels of content of heavy metals can be of immense benefit to diminish the hazard to public health [48]. The potential harm from other metals suggests people eat smaller quantities of fish known to accumulate mercury. However they should also eat a diversity of fish in order to avoid consuming unhealthy quantities of heavy metals. Therefore, the public should bear in mind that standards have a margin of safety [49].

4. Conclusion

This research has provided background information on the concentrations of five metals in freshwater fish species collected in fresh waters. However, given the relatively poor understanding of freshwater food webs and metabolic processes of freshwater fish, it will be advisable to maintain chemical monitoring of commercial fish species in these fresh water body regions.

The heavy metal concentrations in the majority of the samples analysed were well within the prescribed limits set by various authorities, except in a few cases. Therefore, the fish and water from this region, in general, are safe for human consumption. The concentrations of heavy metals analysed in muscle tissue are all well within EU limits for human consumption. The concentrations of cadmium, copper and zinc in fish liver are higher than in muscle tissue.

The data confirm the suitability of several species of freshwater fish as foodstuffs, assessments of dietary intake of trace metals in areas where fish form a significant component of the diet should take this into account in both typical diets and diets of critical groups.

Health institutions, public and private organizations must have a continuous communication about risk-benefit of fish consumption, this confirms the interest to analyse of bases on which a public policy is elaborated, as well as, the responsibility for regulating the quality and improve the balance between benefit and risk of the fish human consumption

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