CONCENTRATION OF SELECTED HEAVY METALS IN WATER AND THE CUMMULATIVE EFFECT ON SELECTED ORGANS OF OREO-CHROMIS NILOTICUS AND CLARIAS GARIEPINUS FROM LAKE VICTORIA, KENYA

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DECLARATION

Declaration by the student

This Thesis is my original work and has not been presented for a degree in any other University. No part of this thesis may be reproduced without the prior written permission of the author and/ or University of Eldoret.

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DEDICATION

To my mum Joyce Tabarno who together with my late dad Kipkosgei Arap Chebochok, spent part of their life in seeing their daughter through many hurdles in life including education. I also dedicate this work to my husband Evans K.Tarus for his financial support and my sons; Elvis Kipkirui, Eliab Kipleting, Eli Kiprop and Eliazer Kimutai for their moral support and encouragement during the study period.

ABSTRACT

The presence of heavy metals in aquatic ecosystems has been of great concern because of their toxicity to man when their concentrations are more than the permissible levels. These metals enter the environment through different ways such as industrial activities. The objectives of the present study were; to determine the concentrations of selected heavy metals (Cu, Cd and Zn) in water samples collected from three different study sites at the shores of lake Victoria, Kenya, to determine the concentrations of the selected heavy metals in selected organs (muscles, liver, kidney, gills and intestines) of two fish species (O. niloticus and C. gariepinus) from the shores of Lake Victoria, and to determine whether there are significant differences in the levels of heavy metals among the selected organs of fish and the water from the Lake. Water samples were collected from the three sites (Kisat sewage discharge point, Molasses factory discharge point and Coca-Cola factory discharge point) at approximately 0.2 m below the water surface using half-litre plastic (PVC) bottles. Five replicate samples were taken from each site. Immediately after sampling the samples were acidified with few drops of concentrated nitric acid. After, the samples were stored in a cool box and transported to the laboratory for further processing. Fish samples were collected from the three sites using an electro-fisher, washed with deionized water, sorted by species, packed in polythene bags and transported to the laboratory in a cool box for further processing. The tissues of the organs were analyzed quantitatively using Atomic Absorption Spectrophotometer. Samples of fish organs were homogenized and diluted with distilled water before analysis. Single classification and three-way factorial ANOVA were used for statistical analyses. Statistical significance was declared at $\alpha = 0.05$. The findings of the present study revealed that the concentrations of heavy metals in water were not significantly different (P≤0.05) among sites. Cadmium ranged from 0.029- 0.045 ug/L, Copper was 0.036-0.042 ug/L while zinc ranged between 0.039-0.113 ug/L. However, the concentrations of heavy metals were significantly higher in fish organs than in water. Concentrations of heavy metals differed significantly (P≤0.05) between species. The concentrations of heavy metals differed significantly among metal types and also among fish organs. In conclusion, all the tested organs of the two fish species caught in Lake Victoria were contaminated with heavy metals. The two fish species demonstrated different capacities to accumulate heavy metals. In addition different organs of fish demonstrated different capacities to accumulate heavy metals. There was high prevalence for metal accumulation in different organs. Consumption of fish from industrial discharge points should be discouraged as it might contain high concentrations of heavy metals.

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LIST OF ABBREVIATIONS

- AAS : Atomic Absorption Spectrophotometer
- APHA : American Public Health Association.
- DHA : Docosahexenoic acid
- EU : European Union
- EPA : Eicosapentenoic acid
- FAO : Food and Agriculture Organization
- Cd : Cadmium
- Cu : Copper
- Fe : Iron
- Mn : Manganese
- pH : Potential Hydrogen
- Pd : Lead
- Zn : Zinc

KMFRI: Kenya Marine and Fisheries Research Institute

USFDA: United States Food and Drug Administration.

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CHAPTER ONE

INTRODUCTION

1.1 Background Information

Fish and other aquatic organisms give a significant contribution to the total human protein supply, and the interest for fish and shellfish is consistently expanding (Bahnaswy and Dheina, 2009). One of the incredible difficulties within years is giving nourishment to the World's growing population. Not only is the need for human protein consumption expanding with the population's growth, additionally quality demands rise as more people can afford protein-rich diets.

The contamination of the aquatic environment with substantial heavy metals has become global problem in past few years, in light of the fact that they are indestructible and the majority of them negatively affect organisms (MacFarlane and Burchett, 2000). Among ecological contaminations, metals are of major concern, because of their potential toxic impact and capacity to bioaccumulate in aquatic biological systems (Censi *et al.*, 2006). Heavy metal levels in aquatic environments are normally monitored by estimating their concentrations in water, sediments and biota (Camusso *et al.*, 1995), which for the most part exist in low levels in water and achieve extensive concentrations in sediments and biota (Namminga and Wilhm, 1976).

Essential and non-essential heavy have fundamental role in ecotoxicology, since they are profoundly persistent and all can possibly be harmful to living organisms (Storelli *et al.*, 2005). Ecological studies on levels of heavy metals in waterways, lakes, fish and sediments (Özmen *et al.*, 2004; Begüm *et al.*, 2005; Fernandes *et al.*, 2008; Pote

et al., 2008; Praveena *et al.*, 2008 and Öztürk *et al.*, 2009) have been of major focus especially during the past few years. Sediments are significant sinks for various contaminats like pesticides and Heavy metals and furthermore assume a role in the remobilization of contaminants in aquatic systems under ideal conditions and in interactions among water and sediments. Fish samples are considered as one of major pointers in freshwater frameworks for the estimation of metal contamination levels (Rashed, 2001).

Heavy metals, for example, Copper, Iron, Chromium and Nickel are essential metals since they assume a significant role in biological environments, whereas Cadmium and Lead are non-essential metals, as they are toxic, even in trace amounts (Fernandes *et al.*, 2008). For the normal metabolism of the fish to carry on, the essential metals must be up taken from water, food or sediment (Canli and Atli, 2003). These essential metals can likewise result in harmful impacts when the metal admission is excessively increased (Tüzen, 2003 and Karthikeyan *et al.*, 2007).

Contaminants enter the fish via five fundamental avenues: through food or non-food particles, gills, oral consumption of water and skin. On absorption, the contaminant is conveyed in circulation system to either a sink point or to the liver for transportation or storage. Contaminants transformed in the liver might be stored there or discharged in bile or moved to other excretory organs, for example, gills, skin or kidneys for excretion or stored in fat which is an additional hepatic tissue (Heath, 1991; Nussey *et al.*, 2000, Bolormaa *et al.*, 2006; Yoon *et al.*, 2008). Studies done in fishes have demonstrated that heavy metals may have harzadous impacts, modifying physiological processes and biochemical parameters both in tissues and blood (Hilmy *et al.*, 1987; Olojo *et al.*, 2005). Adeyeye *et al.*, (1996) demonstrated that the levels of metals was a function of fish species and aggregate more in some fish tissues than others.

Since the harmful effects of metals have been established, concentrations of heavy metals in the tissues of aquatic organisms often checked to guarantee that the levels don't comprise the health of the consumers. Increased human impacts on the environment through heavy metal contamination have however resulted to exhaustion of fish resources and significant decrease in the nutritive qualities (Srivastava and Srivastava, 2008).

1.2 Statement of the problem

There is increased demand for quality foods in several parts of the world. The determination of toxic elements in food has triggered evaluations on their harmful effects on human beings. Heavy metals released from domestic, industrial and other man made activities may contaminate the natural aquatic system extensively and may consequently have devastating effects on ecological balance of the recipient environment and diversity of aquatic organisms. The pollution of aquatic environment with heavy metals is of major concern since they are non-biodegradable and most of them have toxic effects on organisms including fish. Since fish are consumed by human beings and may accumulate large amounts of some heavy metals from the water, it is important to determine their concentration in *Clarias gariepinus* and *Oreochromis niloticus* fish in order to evaluate the possible risk of fish consumption on human health.

1.3 Justification of the study

Heavy metal pollutants are known to have deleterious effects both to human beings, animals and the environment. Aquatic environment is highly affected due to pollutants directly discharged by industrial plants and municipal sewage treatment plants while others come from polluted runoff in urban and agricultural areas and some are the result of the past contamination. The outcomes acquired from this study will provide data on background levels of metal contaminants in the water and fish types of the lake, contributing to the effective monitoring of both ecological quality and the health of the fish harboured in the lake environment. Developing regulations that protect human and aquatic ecosystem health depends on determining the concentrations of potentially-toxic elements in organisms, their effect on the organisms, and the amounts that could possibly be transferred along food chains to higher trophic levels. Presence of elevated quantity of the heavy metals in fish may signal the need to effect corrective measures to restrict the quantity of heavy metal entry into the lake from their source; therefore this study was of importance in establishing whether metal accumulation in animal tissues is different from what is found in water samples. Previous studies have found out that there is higher heavy metal accumulation in fish than in surrounding environment (Olaifa *et al.*, 2004; Rauf *et al.*, 2009).

1.4 Objectives of the study

1.4.1 General Objectives

To determine the concentration of selected heavy metals in water from different sites and selected organs of two fish species from Lake Victoria, Kenya.

1.4.2 Specific Objectives

- To determine the concentration of selected heavy metals (Cu, Cd and Zn) in water samples collected from different sites (Kisat sewage discharge point, Molasses factory discharge point and Coca-Cola factory discharge point) at the shores of Lake Victoria, Kenya
- To determine the concentration of the selected heavy metals in selected organs (muscles, liver, kidney, gills and intestines) of two fish species (*O. niloticus* and *C. gariepinus*) from the shores of lake Victoria.
- 3. To determine whether there are significant differences in the levels of heavy metals among the selected organs of fish and the water from the lake

Hypotheses

1. H_0 There is no net accumulation of levels of heavy metals in fish.

 H_1 There is net accumulation of levels of heavy metals in fish.

- H₀ There is no difference in concentration of heavy metals in various organs.
 H₁ There is difference in concentration of heavy metals in various organs.
- 3. H_0 There are no differences in accumulation of heavy metals among species.

H₁ There are differences in accumulation of heavy metals among species.

CHAPTER TWO

LITERATURE REVIEW

Water is one of the most significant natural resources. The quality of water is of imperative concern for the human since it is legitimately connected with human welfare (Kumar, 2004). A significant ecological concern resulting from industrial and urban waste disposal brought about by human activities is the pollution of soil and water. Controlled and uncontrolled waste disposal, accidental and industrial process spillage, mining and purifying of metalliferous minerals, sewage sludge application to farming soils are responsible for transferring of contaminants into non-polluted sites as residue or leachate and contribute towards contamination of our biological systems (Ghosh and Singh, 2005). A wide scope of organic and in-organic compounds that result in pollution includes; Heavy metals, flammable and putrescible substances, toxic wastes, explosives and oil based components, phenol and textile dyes. Significant constituents of inorganic contaminants are heavy metals. They result in more harm than organic contaminants (Ghosh and Singh, 2005, Gad and Saad, 2008, Jadhav *et al.*, 2010).

The contamination of the aquatic ecosystem with heavy metals has remained a global problem over the past few years, since they are indestructible and the majority of them are potentially harmful to life forms (MacFarlane and Burchett, 2000). Among environmental contaminants, metals are of significant concern; because of their potential harmful impact and capacity to bio accumulate in aquatic biological systems (Censi *et al.*, 2006). Heavy metal levels in aquatic biological systems are typically measured by estimating their concentration in water, sediments and biota (Camusso *et al.*, 1995), which for the most part exist in low levels in water and achieve extensive

levels in sediments and biota (Namminga and Wilhm, 1976). Heavy metals including both essential and non-essential have a specific role in ecotoxicology, since they are exceptionally persistent and all can possibly be poisonous to living organisms (Storelli *et al.*, 2005). Concentrations of heavy metals in rivers, lakes, fish and sediments (Özmen *et al.*, 2004; Fernandes *et al.*, 2008; Poteet *et al.*, 2008; Praveena *et al.*, 2008; Begüm *et al.*, 2009 and Öztürk *et al.*, 2009) have been a significant ecological concern particularly during the most recent decade. Sediments are significant sinks for different contaminants like pesticides and heavy metals and furthermore assume a critical role in the remobilization of contaminants in aquatic frameworks under ideal conditions and in interactions among water and sediments. Fish samples can be considered as one of the most noteworthy indicators in freshwater systems for the estimation of metal contamination level (Rashed, 2001). The commercial and edible species have been broadly researched so as to check for risks to human wellbeing (Begüm *et al.*, 2009).

Metal contamination of fresh water bodies is less visible and direct compared to other types of contaminations, however its impacts on marine systems and humans are exceptionally broad. The presence of metals varies between fish species and relies upon; age, developmental stage and other physiological factors. Fish gather significant levels of Mercury in their tissues and thus contributes a significant dietary source of this element in humans. Fish are the most common source of Mercury and Arsenic for man (Milan *et al.*, 2012). Mercury is a known human toxicant and the principal source of Mercury pollution in man are fishes. Biotransformation of Mercury and Methyl Mercury arrangement comprises a harzadous impact on human health (Emami-Khansari *et al.*, 2005). Soil microorganisms can break down organic contaminants,

while metals need immobilization or physical evacuation. Albeit numerous metals are fundamental, all metals are lethal at higher levels, in light of the fact that they cause oxidative stress by development of free radicals. Another reason why metals might be toxic is that they can replace essential metals in dyes or enzymes interupting their function. Thus, high concentrations of heavy Metals render the land unsuitable for plant growth and destroy the biodiversity (Ghosh and Singh, 2005).

2.1 Definition of heavy metals

The term heavy metals have been used to denote: Metals with atomic number 23 (that is, vanadium) onwards apart from Rubidium (Rb), Yttrium (Y), Cesium (Cs), Barium (Ba), and Francium (Fr); Metals with atomic density greater than 5; and higher metal concentrations which are poisonous to man and other living organisms when ingested from the surrounding environment. Their presence in the organism's environment is of significant public health concern due to their lethal effects to numerous living things including man (Duruibe *et al.*, 2007). Hazard to human health related with consumption of food components contaminated with heavy metals is estimated to be between 20 and 40 times higher than the ingestion of polluted water (Foran, 1990). This is due to the fact that some aquatic life forms have the ability to concentrate substantial heavy metals up to multiple 10^5 the levels present in water (Guimaraes *et al.*, 1985). The eight most regular contaminant heavy metals recorded by the Environment Protection Agency (EPA) are: Arsenic (As), Cadmium (Cd), Cromium (Cr), Copper (Cu), Mercury (Hg), Nickel (Ni), Lead (Pb), and Zinc (Zn) (Athar and Vohora, 2001).

2.2 Distribution of heavy metals in the ecosystem

Biosphere is the natural environment of living objects. It envelops the earth and contains surficial parts of the lithosphere, the lower part of the atmosphere, and the hydrosphere. A relative homeostatic environment is essential for the survival of organisms in ecosystem. This necessitates the study of the chemical composition of air, water, and soil in order to monitor any abnormal changes due to industrial progression and consequent advancement of society (Athar and Vohora, 2001).

2.3 Sources of heavy metals to aquatic system

Aquatic biological system is a definitive beneficiary of practically all toxins including heavy metals. Contamination by heavy metals in aquatic ecosystem is a growing concern and issue worldwide and as of now it has reached an alarming rate. On the other hand, metals additionally occur in limited quantities naturally and may get into aquatic ecosystem through leaching of rocks, airborne residue, forest fires and vegetation (Fernandez and Olalla, 2000). As heavy metals cannot be degraded, they are persistently deposited and incorporated in water, sediments and aquatic life forms (Linnik and Zubenko, 2000), therefore resulting in heavy metal contamination in water bodies. A number of heavy metals get into the water bodies through point sources of heavy metal pollution including, domestic wastewater effluent containing metals from metabolic wastes, industrial effluents and waste sludge substantially contributing to metal loading (Fahmy, 2000). The attributes of point sources of contamination can generally vary from well-characterized point sources (which in themselves can be single or multiple) to diffuse discharges from large numbers of small point sources (for example vehicle fumes or houses) or line sources (for instance motorway run-off) (Hang et al., 2009). Point and non-point sources likewise vary in the manner that the contaminants are discharged to the recipient water body. Besides, contaminant levels emerging from point sources may differ fundamentally with time and space reflecting both the seasonal use patterns and the overall contribution of the predetermining processes (Adamo et al., 2005).

In the past few decades, pollution of aquatic water bodies by heavy metals has evolved into a global problem. Heavy metals may enter aquatic bodies from different natural and anthropogenic (human exercises) sources, including industrial or house hold wastewater, use of pesticides and inorganic fertilizers, storm overflow, leaching from landfills, shipping and harbour activities, geological weathering of the earth hull and atmospheric deposition (Yilmaz, 2009). Contamination of the aquatic ecosystems by inorganic synthetic compounds has been viewed as a significant threat to the aquatic life forms including fishes. The agricultural seepage water containing pesticides and fertilizers and effluents of industrial activities and overflows notwithstanding sewage effluents enter the water bodies and deposit sediments with colossal amounts of inorganic anions and heavy metals (European Commission Environmental Journal, 2002). The most significant anthropogenic sources of heavy metals are industrial, oil pollution and sewage disposal (Santos et al., 2005). industrial utilization of metals, for example, metal plating, tanneries and industrial procedures using metal as catalysts, have created a lot of watery effluents that contain significant levels of heavy metals. These heavy metals include cadmium, chromium, cobalt, copper, iron, manganese, mercury, molybdenum, nickel, silver and zinc. Metal contaminated industrial effluents released into sewage treatment plants could prompt high metal pollution in the activated sludge. Water contamination has turned into a significant risk to the survival of living organisms in aquatic ecosystems (Ezeronye and Ubalua, 2004).

2.4 Roles of heavy metals on animal bodies

The biological and lethal roles of metals have been lately researched in past few years. The following metals/metalloids are viewed as essential for living organisms: Arsenic is a nonspecific growth stimulation, Cobalt is a constituent of vitamin B12,

while Chromium is a regulator of glucose and cholesterol metabolism. High levels of copper in combination with low pH are believed to be lethal to fishes. It is a constituent of oxidases which are vital for the regulation of redox reactions, respiration and ligament development.

Iron is one of the most abundant metals on the earth and is fundamental to practically all life forms in hemoglobin for respiration, cytochromes, catalysis, peroxides. Manganese is a transition metal and like the other transition metals, it happens in various states in the environment as arginase, super oxide dismutase and pyruvate carboxylase in urea synthesis, protecting mitochondria from free radical damage and citrous cycle. Molybdenum causes baldheadedness and xanthenes oxidases formation of fatty acids and uric acids respectively.

Nickel inhibits a variety of enzymes and is bound to different proteins, including metallothioneins and albumins. It is a constituent of a number of enzymes and result to indistinct development stimulation. Selenium plays a role in tissue oxidation and heart muscle problems, Tin works as a constituent of gastrin with digestive and development related roles while Vanadium acts in lipid digestion and bone mineralization. Zinc for aquatic life forms, is an essential metal however it is an environmental pollutant. The intestine is essentially the most crucial organ for zinc absorption, however little is known with respect to up take pathway for zinc in fish. As opposed to copper and iron, zinc does not form free radical particle, and in fact has cancer prevention agent properties. It is a constituent of a number of enzymes; proteases, anhydrates, super oxide dismutase with role in protein biosynthesis, energy metabolism, protection against damage by super oxide radicals and fertility (Athar and Vohora, 2001; Kaániová *et al.*, 2007).

2.5. Toxicity of selected heavy metals

2.5.1Cadmium

Cadmium is a natural component in the world's surface. It is normally found with other elements as minerals. All soils and rocks, including coal and mineral fertilizer, contain a few traces of cadmium in them. In industry and consumer items, it is utilized for batteries (Ni-Cd batteries of cell phones), paints, metal coatings and plastics. It is additionally a constituent in numerous different items, for example, alloys. Cadmium enters air from mining industry and burnt coal and domestic wastes. Its particles can travel for long distances before settling in water or ground (Singh, 2005).

Cadmium is broadly distributed at low levels in the environment and is not a fundamental component for man, animals and plants (USFDA, 1993). Lethal side effects prompted by cadmium include gastrointestinal disturbance brought about by interference of zinc-dependent enzymes, for example, carboxypeptidase that cause impaired digestion, kidney failure and hypertension (Khan *et al.*, 2008). It is likewise reported that, poisoning with cadmium in pregnant mothers has been associated with premature birth and low birth weight and recently, to disorders of the endocrine as well as immunity in children (Schoeters *et al.*, 2006). Cadmium is a highly lethal metal which has no known fundamental role in the body. Cadmium poisoning add to an enormous number of health conditions, including the major killer cardiac diseases, malignancies and diabetes (Bulnes, 2008).

Cadmium dislodges zinc in numerous metallo-enzymes and numerous symptoms of cadmium poisoning can be traced to cadmium-induced zinc deficiency. Cadmium accumulates in the kidney, liver and different other organs and is considered as more lethal than either lead or mercury (Yilmaz, 2009). It is poisonous at levels one tenth that of Lead, Mercury, Aluminum or Nickel. Cadmium poisoning is increasing in frequency today because of zinc deficiency in numerous commonly consumed foods. Zinc, which is defensive against Cadmium, is becoming increasingly insufficient in the soil and consequently in foods. Food processing and consumption of refined foods further decreases zinc absorption. Exposure to Cadmium is likewise increasing because of its utilization as a coating for iron, steel and Copper (Kalay and Canli, 2000). It is also utilized in copper alloys, stabilizers in rubber and plastics, cigarette papers, fungicides and in numerous different items. Regularly these industries at that point then contaminate water, air and food with this metal (Wilson and Pyatt, 2007).

2.5.2 Copper

Copper is generally utilized for wire production and in the electrical industries. Its principle alloys are brass (with zinc) and bronze (with tin). Other examples of use are kitchenware, water conveyance frameworks, and copper fertilizers for instance fungicides and molluscicides which has direct impact on water contamination (Bradi, 2005). Copper is considered as a basic constituent of metalloenzymes of living life forms and is required in hemoglobin synthesis and in catalysis of metabolic reactions (Moore, 2003). It assumes a pivotal role in numerous organic catalyst systems that catalyze oxidation/reduction reactions. However, if present in relatively high levels in the environment, poisoning to aquatic life forms may occur. High copper levels lead to an icrease in the rate of free radical formation (Gwozdzinski, 1995) teratogenicity (Stouthart *et al.*, 1996), and chromosomal variations (Bhunya and Pati, 1987).

Most copper minerals are relatively insoluble in water and hence little is found in natural water. Copper is available in surface water and ground water due to extensive use of pesticides sprays containing copper compounds for agricultural purposes. It is an essential element in human metabolism but can cause aneamia, disorders of the bone and connective tissues and liver damage at excessive levels (Duruibe *et al.*, 2007). Copper poisoning relies on the hardness and pH of the water, and thus it is increasingly toxic in soft water and in water with low alkalinity (Taha, 2004).

2.5.3 Zinc

Zinc is utilized in numerous industries for manufacture of dry cell batteries, alloy production, for example, metal or bronze and manufacturing galvanized coating (Momtaz, 2002). The primary sources of zinc contamination in the environment are zinc fertilizers, sewage sludges, and mining (Bradi, 2005). Zinc is a basic and essential component for the life of animal and pman (Momtaz, 2002). It is present in numerous enzymes associated with significant physiological functions like protein synthesis (Leland, Luoma and Wilkes, 1978). Zinc has been reported to cause indistinguishable signs of ailments as does lead and can be erroneously diagnosed as lead poisoning. Excess levels of zinc can cause system dysfunctions, impedance of development and propagation. The clinical indications of zinc toxicosis in human beings have been indicated as vomiting, diarrhoea, hematuria, icterus (yellow bodily fluid layer), liver and kidney dysfunction, and aneamia (Duruibe *et al.*, 2007).

2.6 Bio monitoring organisms

During the previous couple of decades, numerous species have been researched to establish their potential as biomonitoring organisms, and mollusca have turned out to be an excellent choice for heavy metal monitoring. Mollusca have a depuration mechanism to diminish heavy metal poisoning in their body. This mechanism may lessen the viability of molluscs as biomonitoring organism, as the levels of heavy metal in the mollusc may not precisely mirror the concentration in the environment. In that respect, there is a need to assess the impacts of heavy metal build up and depuration in the biomonitoring organism. Studies have demonstrated that Meretrix meretrix (consumable marine bivalve mollusc) is capable of accumulating Cu, Zn, and Pb in the natural habitat and this species might be utilized as a biomonitoring organism (Abdul et al., 2009). Ruditapes decussatus and Venerupis pullastra are commercially harvested clams with a wide availability and distribution in the shallow inshore waters. They are generally contaminated with heavy metals (Allah et al., 1997). Contamination of bivalve shellfish (for example clams, mollusks, mussels and cockles) is a significant food sanitation concern, so suppliers and retailers should be certain that the items they sell are safe. Bivalves react to changes in levels of contaminants in water, and they can concentrate contaminants from the water prompting generally higher concentrations when contrasted with those present in the water. Shellfish contamination is caused, in addition to other things, by the release of chemical substances, for example, metals, pesticides and organochlorine compounds from industrial and municipal treatment processes. Polluted mollusk shellfish (clams, oysters) may result in human poisoning when they are eaten (Gabr and Gab-Alla, 2008). While a number of heavy metals are nutrients at low levels, Pb, Cd and Hg are nonessential and perceived as significant industrial hazards, causing extreme harmful effects in higher organisms upon acute or chronic exposure. These three components are highly persistent and in the bivalent form, they form stable organic and inorganic complexes in biological systems (Allah et al., 1997] in aquatic ecosystems, heavy metals in dissolved form are easily taken up by aquatic organisms where they are strongly bound with sulfhydryl

groups of proteins and accumulate in their tissues. Fish absorb dissolved or available metals and can therefore serve as a reliable indication of metal pollution in an aquatic ecosystem. Tench (*Tinca tinca*) is considered a good test organism for heavy metal contamination because of its feeding behaviour and bottom feeding habits (Shah and Altindac, 2005). Fish embryos and larvae are generally considered to be the most sensitive to environmental pollutants, thus they have been widely used as bio-indicators for water quality evaluation (Lin and Hwang, 1998).

2.7 Importance of fish

Fish constitutes an important source of animal protein to human beings and a large number of people depend on fish and fishing activities for their livelihood. The rapidly growing human populace has expanded the requirement for food supply. Since fish are quality protein sources, the requirement for fish and shellfish food items has expanded. Worldwide, people get about 25% of their animal protein from fish and shellfish (Bahnaswy *et al.*, 2009). In 2004, about 75% (105.6 million tons) of approximate world fish production was utilized for direct human consumption (FAO, 2007). It had been anticipated that fish consumption in developing nations will increase by 57 percent, from 62.7 million tons in 1997 to 98.6 million of every 2020 (Retman and Zakaria, 2010).

The actual significance of fish in human diet is not just in its high protein content, but additionally due to the two types of Omega-3 polyunsaturated unsaturated fats: eicosapentenoic acid (EPA) and docosahexenoic acid (DHA). Omega-3 (n-3) unsaturated fats are significant for typical development where they decrease cholesterol levels and the occurrence of coronary diseases, stroke, and preterm delivery (Burger and Gochfeld, 2005). Fish additionally contain vitamins and minerals which assume fundamental role in human wellbeing. Since diet is the primary course of exposure to

heavy metals, and fish represent a part of human diet, it is not astounding that contaminated fish could be a risky dietary source of certain toxic heavy metals (Bogut, 1997). There has been a developing enthusiasm for evaluating the degrees of heavy metals in food including cultivated fish. Such focus aims at guaranteeing the security of the food supply and limiting the potential harzadous impact on human health.

CHAPTER THREE

MATERIALS AND METHODS

3.1 Study site

Lake Victoria, the second largest freshwater body in the world (area 68,800 km²), is a shallow lake (maximum depth 84 m; mean depth 40 m), has an irregular shoreline of about 3,440 km in length and lies in catchment of about 184,000 km² in area. The lake lies astride the equator between latitude 2.5°S and 1.5°N, and longitude 32° and 35°E; shared by three riparian states (Kenya 6%, Tanzania 51%, and Uganda 43%, by area). Lake Victoria catchments are constituted by five countries (Kenya, Tanzania, Uganda, Burundi, and Rwanda) and drained largely by a number of rivers (Kagera, Nzoia, Gucha-Migori, Sondu Miriu, Mara, Yala, Issanga and Biharamulo) plus many small rivers and streams. River Nile is the single outlet of Lake Victoria (Raburu, 2003).

The study covered three selected sites on the Lake Victoria along the shores of Winam Gulf. These sites were chosen due to their proximity to catchments with activities that potentially contribute to pollution of the Lake. These were the points where the molasses factory and Coca-Cola factory discharge their effluents in to the Lake. Another site was at the Kisat river mouth which receives most of the domestic sewage discharge from Kisumu town. The basis of selection of cadmium metal is that it is non-essential and has extremely toxic effect on animals and humans, while copper and zinc are essential metals and their toxic effects on animals and humans begins when they are present in high levels.



Figure 3.1: Map showing the study sites (source, www.googlemap.com)

3.2 Information on sampling sites

3.2.1 Kisat sewage discharge point

The region lies between S 00° 05. 273¹, E 034° 44.000¹, altitude 3753 F. The waters were turbid and extensively covered by algal blooms. The shores were covered by *Vossia cuspidate* which provide a good habitat for rapid growth of juveniles of the selected fish species. Appendix I shows Kisat sewage discharge point.

3.2.2 Molasses factory discharge point

The region lies between S 00^{0} 05. 557, E 034^{0} 42, 001^{1} , altitude 3764 F. The waters were slightly turbid, with *Vossia cuspidata* and *Cyperus papyrus* along the shoreline. The nutrient rich overlying water represents a natural spawning and nursing zone for *Oreonchromis niloticus* and *Clarius gariepinus* fish species. Appendix 2 shows the Mollasses factory discharge point.

3.2.3 Coca-Cola factory discharge point

The region lies between S 00^{0} 05. 726¹, E 034 44.123¹ altitude 3763 F.The waters were covered by very thick algal blooms, *Vossia cuspidata* and *Cyperus papyrus* were also found along the shoreline. Appendix 3 shows the Coca-Cola discharge point.

3.2.4 Research design

A total of 60 water samples and 120 fish samples were collected from the three study sites during the study period complete Random Block Design (CRBD) was applied during the sampling.

3.3 Sampling of water

Five different water samples were each collected using half a liter sterile polyvinyl chloride (PVC) plastic water bottles from the five designated sampling points of the three study sites. This was done by dipping the bottles below the water surface to minimize contamination by surface film. The samples of the water were acidified to pH 2 with concentrated nitric acid according to APHA (1998). Water samples were then labeled using stick labels and pencil, subsequently placed in ice-box (72L, China) and transported to the laboratory for storage in a cool box (CA 5411 FFS, U.K) at 4^oC awaiting further analysis. Precautions were undertaken in order to prevent contamination of the samples during pre-treatment included using nitric acid solutions and deionized water to clean all bottles and glass wares prior to using and washing fish samples using de-ionized water prior to dissection to remove adsorbed metals on skin.

3.4 Collection of fish samples

The fish species used in the analysis were selected based on their popularity for commercial and local subsistence used in local diet. Sampling was carried out in the month of July (rainy season). This is because during rains immense volumes of urban run-off influx characteristically deliver into the water high fluxes of suspended solids, nutrients and other pollutants washed from the land and refuse dumps which have remained common features in the nearby Kisumu town. The sampling process was performed using an electro-fisher (SAMUS 700, U.S) with the help of experts from Kenya Marine and Fisheries Research Institute (KMFRI). Appendix 4 shows the collection of the fish samples using an electro-fisher (SAMUS 700). They were washed with de-ionized water then identified by species using the taxonomic key. The fish were then separated by species and location, packed in polythene plastic bags which were labeled using self-adhesive labels, then transported to the KMFRI Laboratory inside ice-box (72 L, China).

3.5 Analysis of samples

3.5.1 Determination of heavy metals in water

The nitric acid-sulphuric acid digestion was used in this study according to APHA (1998). Samples were placed in boiling tubes then digested and concentrated on a Kjendal digestion plate from 100 mL to 25 mL for 3 hours. After digestion, the samples were cooled and the 2 mL of 30% H_2O_2 added to each sample to oxidize any resistant organic matter (Ochumba, 2005). After cooling to 25^{0} C, the digested samples were filtered through 0.45 µm nucleipore filter paper over a vacuum pump and placed in 125 mL plastic bottles and stored in a refrigerator at 4°C. The determination of the concentrations of the metals in samples was done using atomic absorption spectrophotometer (AAS, Model Spectra AA 10/20, Chinas) at the laboratory.

3.5.2 Dissection and drying of fish samples

Dissection was carried out at KMFRI Laboratory whereby the fish samples were descaled and washed with deionized water then dissected using surgical blade, scalpel and stainless steel dissecting pair of scissors. The fish samples were handled using surgical gloves as shown in appendices 6 and 7. Muscles, gills, liver, intestines and kidney organs were extracted during dissection, grouped by species and location in which fish were collected, indicated using indelible pen, packed in polythene plastic bags, kept in ice –box (72L made in china) and transported to the laboratory in University of Eldoret where they were kept in a freezer (CA 5411 FFS, U.K) at 4°C until further analysis. The muscle, gills, liver, intestines and kidney organs were dried separately in an oven (250 RFS, U.S) at a temperature of 70° C for 48 hrs in order to achieve constant weight by removing water.

3.5.3 Digestion of fish samples

The dried fish organs were ground in a mortar into powder, 0.3 g of the ground fish organs were weighed and transferred into labeled, dry and clean boiling tubes, and then 25 mL of concentrated nitric acid was added into each boiling tube. The boiling tubes with samples were then placed in Kjeldahl digestion plate and refluxed under a fume chamber at 360^oC for 2 hours. Samples were then allowed to cool and 10 mL of concentrated hydrochloric acid was added into each boiling tube. The digested sample was then filtered through Whatman filter paper No. 41 into volumetric flask and made up to 50mL mark using deionized water. The digests were poured into labeled plastic bottles then kept in a freezer (2^oC) to await further analysis of the heavy metal concentrations.

3.5.4 Determination of heavy metal concentrations in fish samples

The digests were removed from the freezer, mixed well then analyzed for Cu, Zn and Cd, using the atomic absorption spectrophotometer (VarianSpectr AA-200, Japan). The obtained results were then recorded as mg/L, wet weight.

3.6 Data analysis

Three-way analysis of variance (ANOVA) was used to indicate significant differences in metal levels among sites, species and organs and one-way (ANOVA) was used to compare metals between species in single organ (significant values, $p \le 0.05$). All data were checked, beforehand, for the homogeneity of variances and normality; the data which were not normally distributed or not homogeneous were transformed.

CHAPTER FOUR

RESULTS

4.1 Concentration of metal among species, organ and metal type

The results for the concentrations among species, metals and organs at 95% Confidence intervals are shown in Table 4.1. Concentrations varied significantly among metals and organs at P value \leq .001. Cadmium, copper and zinc had means of 0.255446 mg/L, 0.269647 mg/L and 0.410148 mg/L respectively, while kidney, muscle, gills, liver and intestines had means of 0.313157 mg/L, 0.37434 mg/L, 0.27263 mg/L, 0.278614 mg/L and 0.319994 mg/L respectively. Concentration levels among species were not significantly different.

Table 4.1: Means	for concentrations	among species.	metals and orga	ans at 95% con	fidence intervals
I ubic fill filleund	ior concentrations	unions species,	metals and or St		machiec miter vans

Mean (mg/L)					SED	P- Value
	C. gariepi-					
O. niloticus	nus					
0.307019	0.316475				0.007233	0.3751
Cadmium	copper	Zinc				
0.255446	0.269647	0.410148			0.008858	<.001
Kidney	Muscle	Gills	Liver	Intestines		
0.313157	0.374340	0.272630	0.278614	0.319994	0.011436	<.001
	0.307019 Cadmium 0.255446 Kidney	C. gariepi- O. niloticus nus 0.307019 0.316475 Cadmium copper 0.255446 0.269647 Kidney Muscle	C. gariepi- O. niloticus nus 0.307019 0.316475 Cadmium copper Zinc 0.255446 0.269647 0.410148 Kidney Muscle Gills	C. gariepi- O. niloticus nus 0.307019 0.316475 Cadmium copper Zinc 0.255446 0.269647 0.410148 Kidney Muscle Gills Liver	C. gariepi- O. niloticus nus 0.307019 0.316475 Cadmium copper Zinc 0.255446 0.269647 0.410148 Kidney Muscle Gills Liver Intestines	C. gariepi- O. niloticus nus 0.307019 0.316475 Cadmium copper Zinc 0.255446 0.269647 0.410148 Kidney Muscle Gills Liver Intestines

4.2 Concentration of metal in the different species by organ and metal type

The results for the means of concentrations in species by metal and organ type at 95% confidence intervals are presented in Table 4.2. The concentration in species depends on metal and type of organ at P-Value \leq .001. Cadmium and copper concentrations were high in *C. gariepinus* with means of 0.289772 mg/L and 0.27763 mg/L while it was low in *O. nilotocus* with means of 0.22112 mg/L and 0.261664 mg/L respectively. Zinc concentration was high in *O. nilotocus* with mean of 0.382024 mg/L

Concentration of metals were high in muscles, gills and intestines with means of 0.39052 *mg*/L 0.311927 mg/L and 0.378395 mg/L in *C. gariepinus* while it was low with means of 0.35816 mg/L 0.233333 mg/L and 0.261593 mg/L in *O. nilotocus* respectively. Metal concentrations in kidney and liver of *O. nilotocus* was high with means of 0.372507 mg/L and 0.3095 mg/L but was low in *C. gariepinus* with means of 0.253807 mg/L and 0.247729 mg/L respectively.

	Mean (mg/L)			
Level	O. niloticus	C. gariepinus	SED	P- value
Metal				
Cadmium	0.22112	0.289772	0.012528	<.001
Copper	0.261664	0.27763	0.012528	<.001
Zinc	0.438272	0.382024	0.012528	<.001
Organ				
Kidney	0.372507	0.253807	0.016173	<.001
Muscle	0.35816	0.39052	0.016173	<.001
Gills	0.233333	0.311927	0.016173	<.001
Liver	0.3095	0.247729	0.016173	<.001
Intestines	0.261593	0.378395	0.016173	<.001

 Table 4.2: Means of concentrations in species by metal and organ at 95% confidence intervals

4.3 Concentration of metal type by species and organ type

The results for the means of concentrations of metal by species and organ type at 95% confidence intervals are presented in Table 4.3. The concentration of metals depends on species and the type of organ (P-Value \leq .001). *O. niloticus* had means of 0.14122 mg/L, 0.209332 mg/L, and 0.19742 mg/L while *C. gariepinus* had means of 0.240364 mg/L 0.268488 mg/L and 0.360508 mg/L for Cadmium, Copper and Zinc respectively.

Means of metal concentrations was 0.2002 mg/L, 0.23352 mg/L and 0.50575 mg/L in kidney, 0.2384 mg/L, 0.24813 mg/L and 0.63649 mg/L in muscle, 0.4221 mg/L 0.24766 mg/L and 0.14803 mg/L in gills, 0.19383 mg/L, 0.383983 mg/L and 0.25803 mg/L in liver and 0.2227 mg/L 0.234943 mg/L and 0.50234 mg/L in intestines for cadmium, copper and zinc respectively.

Mean mg/L)						
Level		Cadmium	Copper	Zinc	SED	p- value
Species						
	O. nilotocus	0.14122	0.209332	0.19742	0.005497	<.001
	C. gariepinus	0.240364	0.268488	0.360508	0.005497	<.001
Organ						
	Kidney	0.2002	0.23352	0.50575	0.019808	<.001
	Muscle	0.2384	0.24813	0.63649	0.019808	<.001
	Gills	0.4221	0.24766	0.14813	0.019808	<.001
	Liver	0.19383	0.383983	0.25803	0.019808	<.001
	Intestines	0.2227	0.234943	0.50234	0.019808	<.001

 Table 4.3: Means of concentrations of metal by species and organ type with 95% confidence intervals

Figure 4.1a and 4.1b represent the results for species, organ interaction and species metal interaction respectively. There was significant species, metal and organ interaction at 95.0 percent LSD (P-value \leq .001). Muscle metal concentration and liver metal concentration interacted while copper concentrations and zinc concentrations interacted between *O. niloticus* and *C. gariepinus*.

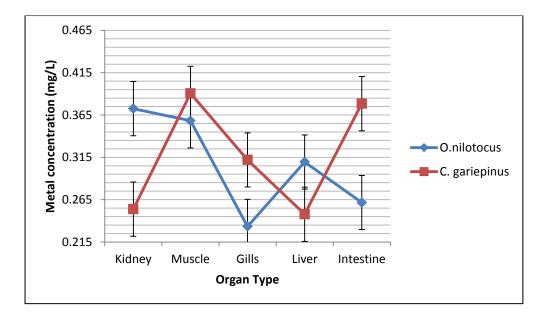


Fig 4.1a: Graph showing species organ interaction

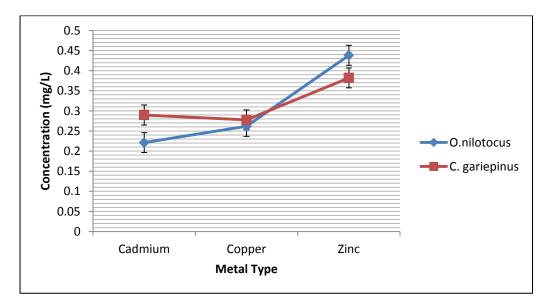


Fig 4.1b: Graph showing Species Metal interaction

4.4 Concentration of metal in organ by species and metal type

The results for the means of concentrations of metal in organ by metal and species type at 95% Confidence intervals are represented in Table 4.4. Concentration of metal depended on metal and the type of organ (P-Value \leq .001). *O. niloticus* had mean concentrations of 0.209487 mg/L, 0.146867 mg/L, 0.213313 mg/L, 0.203153 mg/L and 0.140467 mg/L while *C. gariepinus* had mean concentration of 0.27785 mg/L, 0.243187 *mg*/L, 0.264897 mg/L, 0.338653 mg/L and 0.324347 mg/L of metal in kidney, muscle, gills, liver and intestines respectively.

Kidney, muscle, gills, liver and intestines had mean concentration of 0.17683 mg/L, 0.1307 mg/L, 0.38573 mg/L, 0.1489 mg/L and 0.1118 mg/L of cadmium, 0.230575 mg/L, 0.11946 mg/L, 0.146915 mg/L, 0.43746 mg/L and 0.185 mg/L of copper and 0.3236 mg/L, 0.25978 mg/L, 0.18467 mg/L, 0.22635 mg/L and 0.40042 mg/L of zinc respectively.

Kidney	Muscle	Gills	Liver	Intestines	SED	p- value
0.209487	0.146867	0.213313	0.203153	0.140467	0.195436	<.001
0.27785	0.243187	0.264897	0.338653	0.324347	0.195436	<.001
0.17683	0.1307	0.38573	0.1489	0.1118	0.008692	<.001
0.230575	0.1946	0.146915	0.43746	0.185	0.008692	<.001
0.3236	0.25978	0.18467	0.22635	0.40042	0.008692	<.001
	0.209487 0.27785 0.17683 0.230575	0.209487 0.146867 0.27785 0.243187 0.17683 0.1307 0.230575 0.1946	0.209487 0.146867 0.213313 0.27785 0.243187 0.264897 0.17683 0.1307 0.38573 0.230575 0.1946 0.146915	0.209487 0.146867 0.213313 0.203153 0.27785 0.243187 0.264897 0.338653 0.17683 0.1307 0.38573 0.1489 0.230575 0.1946 0.146915 0.43746	0.209487 0.146867 0.213313 0.203153 0.140467 0.27785 0.243187 0.264897 0.338653 0.324347 0.17683 0.1307 0.38573 0.1489 0.1118 0.230575 0.1946 0.146915 0.43746 0.185	0.209487 0.146867 0.213313 0.203153 0.140467 0.195436 0.27785 0.243187 0.264897 0.338653 0.324347 0.195436 0.17683 0.1307 0.38573 0.1489 0.1118 0.008692 0.230575 0.1946 0.146915 0.43746 0.185 0.008692

Table 4.4: Means of concentrations of metal in organ by metal and species type with 95% confidence intervals (mg/L)

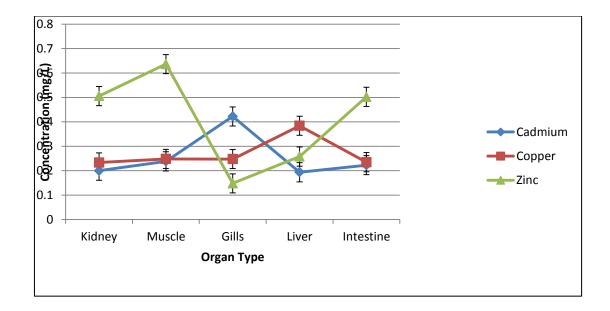


Fig 4.2: Graph showing Metal Organ interaction.

Figure 4.2 represents the results for metal organ interaction. There was significant metal organ interaction at 95.0 percent LSD (P-value≤.001). Interactions between cadmium and copper were observed in kidney, muscle and intestines while interactions between cadmium and zinc was observed in the Liver.

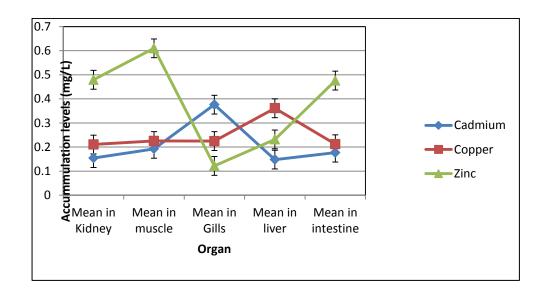


Figure 4.3: Graph of Levels of Accumulation of metals in different organs

There was significant accumulation of metals in different organs at 95.0 percent LSD (P-value≤.001). All the test organs accumulated lower levels of copper and cadmium

except the gills and liver, which had higher levels of cadmium and copper respectively. More zinc accumulated in kidney and muscle while lower levels accumulated in the gills and liver.

 Table 4.5: Means of concentrations of metal by Site with 95% confidence intervals

	Mean (mg/L)							
Level	Kisat		Mollasses comp	Coca cola comp	SED	p- value		
Metal								
	Cadmium	0.0450	0.0420	0.0290	0.0156	0.1016		
	Copper	0.0360	0.0420	0.0405	0.0156	0.1016		
	Zinc	0.0393	0.1128	0.0634	0.0156	0.1016		

The results for the means of concentrations of metal by site at 95% confidence intervals are shown in Table 4.5. Concentration of metal by site did not show any significant difference

CHAPTER FIVE

DISCUSSION

5.1 Concentration of metal among species, organ and metal type

Cadmium and copper concentrations was high in C. gariepinus with means of 0.290 mg/L and 0.278 mg/L while it was low in O. niloticus with means of 0.221 mg/L and 0.262 mg/L respectively. Zinc concentration was high in O. niloticus with mean of 0.438 mg/L while it was low in C. gariepinus with mean of 0.382 mg/L. Surface water fish in the study (C. gariepinus) have higher level of Cd compared to the demersal fish species. This could be attributed to the very fact that sediment Cd concentration of is lower owing to continuous unleash of electrons to the atmosphere through respiration processes (Kersten and Forstner, 1987), thus causing higher exposure to the surface water species. Similar results were recorded by Mwashote (2003), in a study of Indian Ocean whereby high cadmium levels were recorded at sites affected by anthropogenetic activities. The high cadmium concentration in fish was attributed to its high mobility and therefore the importance of this metal within the transportation of calcium in associate organism. Aquatic organisms also lack appropriate excretion mechanism for cadmium. The same results were recorded at Awassa and Koka Lakes in Ethiopia (Jared and Usman, 2011; Milan et al., 2012). The high cadmium concentrations pose a human health concern. There is potential influence of geochemical processes on the concentration values further because of the increase in subsistence, commercial and recreational fishing activities (Tuionen et al., 2006). Cadmium is known to have long continuance in urinary organ and liver, and is readily bound to Haemoglobin and metallothionein (Njogu et al., 2011).

Zinc was the dominant metal with the highest concentration (0.410 mg/L) in fish. Zinc concentration recorded in Lake Baringo exceeds WHO permissible limit of 10 mg/kg. The high zinc concentration levels are due to the crucial biological role in growth and metabolism in fish. Therefore, fish have active uptake and storage of Zinc (Njogu *et al.*, 2011). Ismaniza and Idaliza (2012) also recorded results similar to this study in Lake Naivasha, Kenya.

Concentration of metals was high in muscles, gills and intestines with means of 0.39052 mg/L, 0.312 mg/L and 0.378 mg/L in C. gariepinus while it was low with means of 0.358 mg/L, 0.233 mg/L and 0.262 mg/L in O. niloticus respectively. Metal concentration in Kidney and Liver of O. niloticus was high with means of 0.373 mg/L and 0.310 mg/L but was low in C. gariepinus with means of 0.254 mg/L and 0.248 mg/L respectively. From the results obtained in the study, it can be deduced that significant accumulation of heavy metals is species-related. Thus the distinct difference noticed in the levels of accumulation in specified organs of fish species is attributed to the variations in their physiological roles toward maintaining equilibrium, feeding habits, regulatory ability and behaviour of every fish. It has been reported that increased metal levels in fish tissues arise through bio-magnification at every trophic level and therefore the omnivorous bottom feeders concentrate highest metal levels (Forstner and Wittmann, 1981). C. gariepinus is a known voracious bottom feeder and would therefore have bio-accumulated high metal levels from the Lake sediment. The studies by; Ekpo et al., (2008), Edem et al., (2009) and Yilmaz, (2009) indicated that muscle is less active than the liver in accumulating heavy metals, confirming the results of the present study and it conjointly conforms with the results of previous

studies that heavy metals were more concentrated in the liver than other tissues/organs of the fish by Ali, (2007). Heavy metals in excess of the body needs of fish might represent a serious pollution supply and cause a heavy health risk (Onyia *et al.*, 2007).

5.2 Concentration of metal type by species and organ type

The Concentration of metals depends on species and the type of organ (P-Value \leq .001). The higher concentrations of cadmium and copper in the gills and liver reflects the functional roles performed by those tissues/organs. The gills of fish have the thinnest epithelium and are constantly in direct contact with all the contaminants found in water, as they act like filters, and therefore metal ions easily penetrate (Bols *et al.*, 2001).

In *C. gariepinus*, lower concentrations of the metal in muscles indicates low affinity between the element and muscles probably because muscles have low macromolecule binding potential (Karadele and Unlo, 2007). The higher concentration of Copper in liver could also be attributed to the affinity or strong coordination of metallothionein protein with the elements. The differences in metal contents in various tissues of the two species is explained by considering aspects like concentrations levels of heavy metals within the environment, ecological needs, metabolism, feeding patterns of the species, exposure time and seasonal variations (Chettopadhyay, 2002). According to the metal-organ interaction results, muscle metal concentrations and liver metal concentrations interacted while copper concentrations and zinc concentrations interacted between *O. nilotocus* and *C. gariepinus*.

5.3 Concentration of metal in organ by species and metal type

Concentration of metal depended on metal type and the organ type (P-Value<.001). Cadmium concentration values in O. niloticus tissues reflected a decreasing trend from liver, gills and muscles while in C. gariepinus the trend was found to be in the opposite direction. The distribution pattern of cadmium observed in liver, gills and muscles of fish can be related to the fact that the liver, just like in animals, functions as the organ for detoxication and storage of many types of toxins. These findings are in line with El-Nemr, (2003); Khaled, (2004); Mwashote (2003) who conjointly in their studies found that, Cd concentration in O. niloticus was highly accumulated in kidney and liver. Higher concentrations of cadmium in muscles of C. gariepinus may be related to the life style the species spending longer time at the bottom or muds. This may easily facilitate penetration metals through skin and gills. Likewise, depending on the character of the component, the absorbed toxicants can be distributed quickly to alternative tissues and organs rather than accumulating solely within the liver. The differences in metal concentration that was determined between species and distribution in different tissues and the quantity rely mostly on the concentration within the encompassing setting, feeding habit and adaptability of the species to the chemicals and mechanisms developed by the species to eject the toxicants.

Copper is an essential micronutrient needed by organisms for normal metabolic processes (WHO, 2004). Although copper is an essential element in human development of bones and growth, at high doses, copper causes vomiting, diarrhoea, nausea and headache while chronic copper toxicity results in gastrointestinal bleeding, haematuria, intravascular haemolysis and acute renal failure (Lalar et al., 2008). Hellawell(1986) classified copper as a very toxic metal in high concentration. Although copper is naturally occurring within the ecosystem in trace amounts, the concentration level in fish samples from Lake Victoria does not exceed WHO guideline values. The results are in agreement with the findings of the study carried out in Egypt Northern Delta Lake on metals in *Oreochromis niloticus* (Lalar *et al.*, 2008). Lalah *et al* (2008) also recorded similar results in Lake Victoria, Kenya that are in agreement with the current study. Interactions between cadmium and copper were observed in kidney, muscle and intestines while interactions between cadmium and zinc was observed in the Liver.

There was significant accumulation of metals in different organs at 95.0 percent LSD (P-value \leq .001). All the test organs accumulated lower levels of copper and zinc except the gills and liver, which had higher levels of cadmium and copper respectively. More zinc accumulated in kidney and muscle whereas lower levels accumulated in the gills and liver. Physical-chemical factors and biological processes occurring permanently in aquatic environments could influence the availability of different metals in water. Heavy metals have high uptake rates with high water temperature as a result of higher metallic rate, increasing the rate of metal uptake and binding (Olaifa *et al.*, 2004). Water pH also affects heavy metal accumulation with high accumulation in acidic than alkaline water. Low pH increases free metal ion concentration in water and consequently increasing bioavailability of these metals (Al-Kahtani, 2009).

5.4 Heavy metals in fish samples collected from Lake Victoria

Heavy metal accumulation was found to be high in fish than in their surrounding water environment, signifying bioaccumulation effect in higher trophic levels on food chain which is in agreement with the findings by Olaifa *et al.*, (2004) in a lake and fish farm in Ibadan, Nigeria. These findings are also in line with those of Rauf *et al.*, (2009) which also releaved higher heavy metal accumulation in fish than in surrounding water. Heavy metals enter the fish through gills, skin, oral in food and water. In the fish body, the metal is transported through the blood stream and either stored, transformed or eliminated in the liver, kidney or the gills (WHO, 2004). Fish are capable of regulating heavy metal uptake up to a certain level beyond which bioaccumulation occurs (Al-Kahtani, 2009). Bioaccumulation of any metal in fish is a factor of absorption, ingestion and excretion, heavy metal concentration and bioavailability as well as physical factors like temperature.

CHAPTER SIX

CONCLUSION AND RECOMMENDATIONS

6.1 Conclusion

Water samples collected from the three sampling sites had presence of heavy metals but the levels did not exceed the maximum permissible levels recommended by WHO.

All the tested organs of fish species caught in Lake Victoria were contaminated with heavy metals. Different organs of fish demonstrated different capacities to accumulate heavy metals. There was high prevalence for metal accumulation in different organs. According to WHO the permissible limits in fish tissues for Cu, Cd and Zn are 1.0 mg/L, 1.0mg/L and 5.0 mg/L respectively.

The heavy metal concentrations of the two fish species were found to be significantly different among organs and the water from the lake.

The concentrations of Cu, Cd, and Zn in samples of fish organs in this study were found to be lower than these maximum permitted levels. Heavy metal accumulation was found to be high in fish than in their surrounding water environment, signifying bioaccumulation effect in upper trophic levels along food chain.

The concentrations of heavy metals in the fish and water column were detected in low concentration, however the potential for metal toxicity danger may become more se-

vere in future depending upon the extent of industrial and domestic water influx into the lake due to human activities in the adjacent areas.

Stringent monitoring of the discharges into the Lake containing heavy metals is recommended. Fishing activities should be avoided among discharges; consequently consumption of fish from discharge points should be discouraged as it might contain high concentrations of heavy metals.

6.2 Recommendations

A research on accumulation of heavy metals in different trophic levels in Lake Victoria, Kenya is recommended. This will lead to more information on the possible extent of contamination with heavy metals across different animal species and guide on comprehensive corrective and preventive measures.

A study also on the seasonal variation of heavy metals in the water and sediments of Lake Victoria, Kenya needs to be done. This is based on the accumulation of heavy metals in water during different seasons due to volume of run-off water and effect of water cycling which varies with seasons.

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APPENDICES



Appendix I: Kisat sewage discharge point (5m from the shore of Lake Victoria)



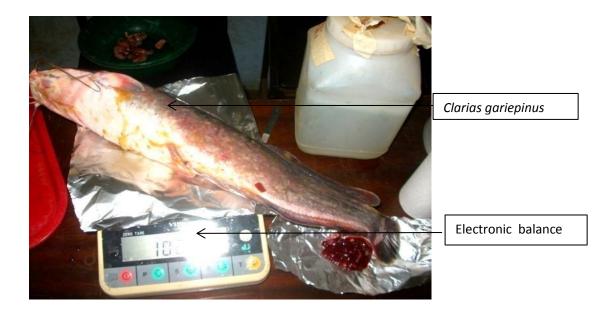
Appendix II: Mollasses factory discharge point (65m from the shore of Lake Victoria)



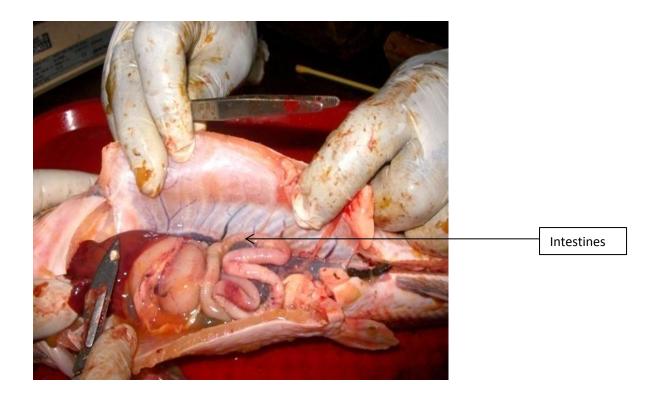
Appendix III: Coca Cola factory discharge point (50m from the shore of Lake Victoria)



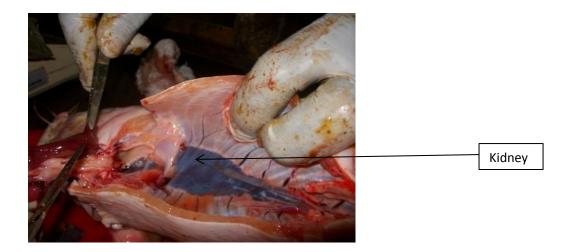
Appendix IV: The researcher collecting fish samples using an electro-fisher assisted by the KMFRI technician and a fisherman.



Appendix V: weighing of the *Clarius gariepinus* samples using an electronic balance (Shimadzu).



Appendix VI: Dissection showing the intestines of *C. gariepinus* among other organs



Appendix VII: Dissection showing the kidney of *O. niloticus*.



Appendix VIII Boiling tubes with samples in Kjeldhal digestion plate.



Appendix IX. Researcher preparing samples for digestion in the Laboratory.

Country	Metal type concentration (mg/L)					Reference
Country	Mercury	Lead	Cadmium	Copper	Zinc	Reference
Australia	<1.0	<1-7.0	<0.2-1.8	2.0	25	ANZECC, 2000
Kenya	5.0	10	10	15	50	EMCR, 2006
New Zealand	<1.0	<1-7	<0.2-1.8	4	12.5	ANZECC, 2000
Philipines	2.0	50	10	-	-	DAO, 1990
United States	1.4	65	4.3	6.2	25	EPA

Appendix X. Table of Maximum lead Metal concentrations in Fresh water fish

Metal		EU standards
	WHO standards	
Cadmium (Cd)	0.003 mg/l	0.005 mg/l
Copper (Cu)	2 mg/l	2.0 mg/l
Zinc (Zn)	3 mg/l	Not mentioned

Appendix XI. Table of comparison of acceptable levels of metals in drinking water