Heavy Metals in Waste Water, Soil and Vegetables around Chunga Gardens in Lusaka and their Potential Health Risks

By

Nyirongo Kunda

A thesis submitted to the School of Public Health, University of Zambia in fulfilment of the requirements of the Degree of Master of Public Health in Environmental Health

The University of Zambia

2020

COPYRIGHT

DECLARATION

I, Kunda Nyirongo declare that this thesis entitled "Heavy Metals in Wastewater, Soil and Vegetables around Chunga Gardens in Lusaka and their Potential Health Risks" herein presented for the Degree of Master of Public Health (Environmental Health) represents my own work and has not been previously submitted either wholly or in part for other Degree at this or any other University nor is it being currently submitted for any other Degree.

CANDIDATE

Name: Kunda Nyirongo

Signed: ___________________________

Date: __________________________

SUPERVISOR

Name: Ms. Patricia Mubita

Signed: ___________________________

Date: __________________________

SUPERVISOR

Name: Ms. Chisala .D. Meki

Signed: ___________________________

Date: __________________________

CERTIFICATE OF APPROVAL

This dissertation by Kunda Nyirongo entitled “Heavy Metals in Wastewater, Soil and Vegetables around Chunga Gardens in Lusaka and their Potential Health Risks” is approved as fulfilling the requirement for the award of the Degree of Master of Public Health (Environmental Health) by the University of Zambia.

Examiner I
Name: ________________________________
Signature: _____________________________
Date: _________________________________

Examiner II
Name: ________________________________
Signature: _____________________________
Date: _________________________________

Examiner III
Name: ________________________________
Signature: _____________________________
Date: _________________________________

ABSTRACT

Dietary exposure to several heavy metals at high concentrations including Cadmium (Cd), Copper (Cu), Lead (Pb) and Zinc (Zn), have been recognized as a risk to human health. This is through the consumption of vegetables. Hence, the study aimed at determining the concentration and bioaccumulation factors of heavy metals in wastewater, soil and vegetables around Chunga gardens in Lusaka District of Zambia and their potential human health risks. A cross sectional study design was conducted, with a total of 45 samples collected, which included: 9 wastewater samples, 8 soil samples and 28 vegetable samples namely *Brassica rapa* (Chinese Cabbage), *Brassica napus* (Rape), *Brassica juncea* (Mustard Green:Mpilu) and *Cucurbita moschata* (Pumpkin Leaves) to ascertain heavy metal concentrations.

Heavy metal analysis was determined using Atomic Absorption Spectrophotometer (AAS) and Stata version 14 was used for data analysis. The study revealed that the concentrations of Cd, Cu, Pb and Zn were beyond the maximum permissible standards in wastewater, soil and vegetables based on the World Health Organisation standards. The concentrations of heavy metals found in the wastewater ranged in the order of Cu (2.59 mg/l - 0.01 mg/l) > Pb (0.66 mg/l - 0.01 mg/l) > Zn (0.60 mg/l - 0.01 mg/l) > Cd (0.03 mg/l - 0.01 mg/l).The concentrations of heavy metals found in the agricultural soil were in the sequence of Cu (133.00 mg/kg – 2.59 mg/kg) >Zn (73.90 mg/kg - 0.87 mg/kg) > Pb (42.20 mg/kg -8.20 mg/kg) > Cd(19.00 mg/kg – 0.10 mg/kg). The concentrations of heavy metals in edible parts of selected vegetables were in the order of Zn (418.26 mg/kg - 23.15 mg/kg) > Cu (416.185 mg/kg -0.94 mg/kg) > Pb (254.515 mg/kg - 0.01 mg/kg) > Cd (5.08 mg/kg- 0.01 mg/kg).The median concentrations of Cd, Cu, Pb and Zn were not different from the vegetables irrigated with wastewater and the freshwater (control group). Median Concentrations of heavy metals in wastewater, soil and vegetables were statistically different ($p<0.05$).Based on the observed bio-accumulation factors, *Brassica napus* showed more hyper-accumulation potential as compared to the other vegetable samples.

The potential non-carcinogenic health risk assessment showed that Cd, Cu, Pb and Zn are heavy metals which are likely to produce non-carcinogenic adverse health effects, while average carcinogenic risk values obtained for Pb and Cd using Cancer Slope Factors (CSF) in this study indicated a lifetime (70 years) probability of contracting cancer from the ingestion of contaminated vegetables to both adults and children. Based on the findings of the study repetitive consumption of the contaminated vegetables might pose undesirable health effects inclusive cancer for the consumers. Therefore, the study emphasizes the need for systematic monitoring of the environment contaminated with heavy metals to avoid health risks as well as an assessment of pollution source distribution is highly recommended.

Keywords: *Heavy metals, Wastewater; Irrigation; Soil; Vegetables; Concentration; Permissible limits; Health Risk; Bio-accumulation Factor:*

DEDICATION

I dedicate this work to the Almighty God who made all things possible and to all men and women of God who have given my life a meaning. I also dedicate this work to my mother Fania Shisholeka, my younger brother Simon Shisholeka and my aunt Mercy Choma Because of their unfaltering love, devotion, and motivation, I have been able to find within myself the fortitude and wherewithal to achieve my academic goals. This work is also dedicated to my late father Mukanda Edson Shisholeka, my late grandmother Esnart Ngoma and my late grandfather Fidelis Bernad Kaimbi, who contributed positively to my life and would have been delighted with my academic success.

ACKNOWLEDGEMENTS

I would like to thank all Master of Public Health Lecturers and Fellow Master of Public Health Students. Special thanks go to my Supervisors: Ms Chisala Meki and Ms Patricia Mubita for their positive contribution, for they have made me the scientific researcher I am today. Special appreciation goes to Ms Fania Shisholeka for the technical and financial help rendered towards my research project. I also acknowledge the support I received from Lusaka Water and Sewerage during the data collection. I am equally indebted to the School of Agriculture Laboratory Staff of the University of Zambia for their valuable support in analysing my wastewater, soil and vegetable samples. Above all, I give glory and honour to my God for being with me all the time.

TABLE OF CONTENTS

Contents

LIST OF TABLES

LIST OF FIGURES

ABBREVIATIONS/ACRONYMS

ATSDR	Agency for Toxic Substances and Disease Registry
CDI	Chronic daily intakes of metals
Cd	Cadmium
Cu	Copper
FAO	Food and Agriculture Organization
FDA	Food and Drugs Act
FSA	Food Safety Agency
HQ	Hazard Quotient
LCC	Lusaka City Council
ML	Maximum limit
Pb	Lead
USEPA	United States Environmental Protection Agency
WHO	World Health Organization
ZEMA Zambia	Environmental Management Agency
Zn	Zinc

OPERATIONAL DEFINITIONS OF TERMS

Around Chunga gardens – Gardens located not more than 10km from the Chunga Sewage Treatment Plant.

Carcinogenic – Substances that are not characterized by a threshold below which the body is able to cope or recover from an exposure. All repeated exposures to a carcinogenic substance add up, and the risk is never zero, at low doses, the risk is proportional to the exposure.

Chronic Daily Intake – Consist of a lifetime average daily dose taken over an assumed 70 year human lifetime.

Dose – A measure of intake of a substance, usually expressed in units of mg/kg-day (mg of contaminant per kg body weight per day).

Hazard Quotient - This is a dimensionless quantity, which entails the risk of being exposed to a toxic chemical.

Heavy Metals – These are metallic elements that have a high atomic weight and cause damage to living things even at low concentrations, by accumulating along the food chain. The heavy metals of highest public concern are arsenic, lead, cadmium, asbestos, thallium, copper and mercury.

Non-carcinogenic – Substances that are characterized by a threshold below which the body is

able to cope with or recover from an exposure. A brief or low exposure leaves no consequence until the next exposure.

Maximum level (ML) – this is the maximum concentration of contamination in food substance recommended by the World Health Organisation (WHO) to be legally permitted in that commodity.

RfD – Reference Dose, The daily exposure level which, during an entire lifetime of a human, appears to be without appreciable risk.

Control Group- Fresh water and vegetable samples watered without wastewater.

Bio-accumulation factor (BF) – This is the ratio of metal concentration in plant biomass to that in the soil.

Wastewater -This is water whose quality is compromised by anthropogenic influence, domestic liquid waste discharge, and waste from industries and/or agriculture, and encompasses a number of potential contaminants and concentrations. For this study wastewater means the treated waste water that is released from the Chunga wastewater treatment plant into the Chunga stream and maintains the wastewater characteristics for odour, colour and turbidity.

CHAPTER ONE

INTRODUCTION

1.1 BACKGROUND INFORMATION

Food safety is a major public health concern worldwide. Food consumption has been identified as the major pathway for human exposure to certain environmental contaminants, accounting for > 90% of intake compared to inhalation or dermal routes of exposure (Adelekan and Abegunde, 2011). About 30% of human cancers are caused by low exposure to initiating carcinogenic contaminants in the diet (Simutanyi, 2008). During the last decades, the increasing demand for food safety has stimulated research regarding the risk associated with consumption of foods contaminated by pesticides, heavy metals and / or toxins (Sracek et al., 2012).

Bull 2003, reported that identifying excessive accumulation of trace elements in crops is one of the most important aspects of food quality assurance. Thus international regulations such as the World Health Organisation (WHO) heavy metal standards on food quality have lowered the maximum permissible levels of toxic metals in crops due to an increased awareness of the risk these metals pose to the food chain (Nhapi et al., 2012). Regular monitoring programmes of heavy metal contents in crops have been carried out for decades in most developed countries (Kavcar et al., 2009). On the other hand, in developing countries such as Zambia, limited information is available regarding heavy metal contamination in wastewater, soil and vegetables (WHO/UNICEF, 2015).

According to Koki et al. (2015), Heavy metals are defined as “metallic chemical elements that have a relatively high density and are toxic or poisonous at low concentrations”. Koki *et al* (2015), noted that heavy metals are ubiquitous in the environment and are caused by natural and anthropological sources such as: irrigation with contaminated water, addition of fertilizers and metal-based pesticides, sewage sludge, organic manure. Koki et al., (2015), further stated that the food chain is the major pathway of heavy metals exposure to humans while wastewater as well as soil accounts for the pathway in vegetables (Khan et al., 2008).

World Health Organization reported that water quantity and quality are both subjects of concern(WHO, 2011). Reclamation of wastewater is one of the main options as a new source of water in agriculture usage, due to the scarcity of good quality irrigation water. Paulinus (2015), defined wastewater as “used water from any combination of domestic, industrial, commercial or agricultural activities and any sewer inflow or sewer infiltration". Paulinus (2015), noted that long term irrigation through wastewater results in build-up of heavy metals in soil that can restrict soil functioning resulting in toxicity to vegetables and contamination of the food chain thus affecting food quality and safety (Sardar et al., 2013; Qaiyum et al., 2011).

Khan et al. (2008) added that peri-urban farmers use wastewater for irrigation as they assume that wastewater is a perquisite in vegetable nutrition. In spite of containing essential plant nutrients such as N, P, K, Ca, Mg and Fe which are important for plant growth the yield of the vegetables is reduced due to the presence of heavy metals in the soil. This is because metabolic processes of the vegetables are disturbed by heavy metals (Singh *et al*., 2001; Hanif et al., 2006).

Despite the inherent danger of the consumption of heavy metal contaminated food crop and their potential health risks, a number of people have continued to engage in wastewater usage for irrigation (Muzungaire et al., 2015; Simutaniyi, 2008). Paulinus (2015), stated that vegetable species differ in their efficiency to absorb heavy metals, as bioaccumulation of metals by vegetables is affected by many factors. In general, variations in vegetable species, the growth stage of the vegetables and element characteristics control absorption, accumulation and translocation of metals. Furthermore, physiological adaptations also control toxic metal accumulations by sequestering metals in the roots and edible parts which may be at phytotoxic levels or originate health risks (Ojekunle et al., 2016).

Wu *et al*. (2009), reported that Human exposure to heavy metals such as Cadmium, Copper, lead and Zinc have been linked with increased blood acidity, developmental retardation, various cancers, kidney damage, emotional instability, vision disturbances and even death. The populations most affected by heavy metal toxicity are women or very young children (Morais and De Lourdes 2012). Therefore, there has been an aggregate concern, mainly in the developing countries, regarding exposures, intakes and absorption of heavy metals by human beings. Populations are increasingly demanding a cleaner environment, and reductions in the amounts of contaminants reaching people as a result of increasing human activities (Blacksmith Institute, 2006). A practical implication of this trend, in the developed countries, has been the imposition of restrictive legislations regarding environmental pollution from developed countries (US EPA, 2004).

In a nutshell, toxicity associated with exposure to heavy metals if unrecognized or inappropriately treated represents a clinically significant medical problem, having greater impact on increasing the morbidity and mortality rate (Momodu, 2010). Thus the present study assessed heavy metal concentrations in the wastewater, soil and vegetables around Chunga gardens, determined the bio-accumulation index of the vegetables and ascertained the health risk due to consumption of the vegetables. For this study the status of four heavy metals (Pb, Cd, Cu, and Zn,) in four vegetables namely: *Cucurbita moschata* (Pumpkin Leaves), *Brassica napus* (Rape), *Brassica napa* (Chinese cabbage) and *Brassica juncea* (Mustard Green:Mpilu) were determined.

1.2 PROBLEM STATEMENT

Studies conducted in Zambia regarding peri urban farming reveal that problems pertaining to heavy metal contaminated wastewater use in vegetables have not been adequately tackled (Shitumbanuma & Tembo, 2006). A study by Ojekunle et al. (2016) noted that, the major problem associated with wastewater irrigation include insufficient information on the heavy metal concentration in the wastewater, soils and vegetables (Nachiyunde & Nishijima, 2013). In addition, it is important to mention that excessive accumulation of heavy metals in wastewater may not only result in environmental contamination, but may lead to elevated heavy metal uptake by vegetables, which may affect food quality and compromise health. Regardless of the problems associated with wastewater irrigation farming, it is a source of income for a large number of the peri urban poor in Zambia (Ikenka et al., 2010; Ettler et al., 2011).

Wastewater reclamation is being used in Chunga township as it is easily accessed by the marginalized groups in the production of vegetables. Owing to the fact that the benefits include increase in vegetable yield, income generation and improved food security at household level. This has resulted in the garden owners diverting wastewater from the Chunga stream where the Chunga treatment plant discharges end effluents, by using water pump and creating trenches to direct the flow to their vegetables (Kribek & Nyambe, 2010). Hence this common operational pattern has caused severe concerns with regards the environmental sound use of municipal wastewater for irrigation for the small scale peri urban gardens. Often the condition of the wastewater discharged in to the Chunga stream is unknown or undocumented as well as its impact on health as the Chunga treatment plant doesn't take heavy metals in to treatment as the most probable is the biological treatment. Thus the status of the wastewater on heavy metals is unknown when released in the environment and with within the vicinity of the Chunga treatment plant where a number of vegetable gardens are located (Itodo et al., 2011;Khan etal.,2013; Liu et al.,2013).

It is imperative to mention that if the vegetables being sold do not meet the maximum limits (ML) of heavy metal content in them in accordance with regulations, consumers are prone to diseases and subsequently creating health problems. In 2004 Chisanga, conducted a study to assess the effects of using wastewater on vegetable growing and the associated socio-economic impacts on farmers in the Kafue Lagoon Areas in Lusaka. The study did not adequately tackle the assessment of the concentration of heavy metals in soil and vegetables.

Thus, this study aimed to assess heavy metals in wastewater, soil and vegetables around chunga gardens and their potential human health risks in Lusaka District.

1.3 STUDY JUSTIFICATION

In spite of the various laws and policies that guide food quality and wastewater standards such as the Food and Drug Act, Public Health Act in addition to the statutory regulatory organisations such as ZEMA, available literature indicates a gap in the true extent of heavy metal pollution in soil and vegetables irrigated with wastewater as well as the quality of the wastewater (Tembo, 2006). Yet such information is vital for the production of sufficient knowledge on the quality of the wastewater, soil and the vegetables. In this context, the present study was undertaken to.

Thus, the study has provided valuable insights into what needs to be accomplished in the context of heavy metal contamination in agricultural fields. In this regard therefore, the findings from the study can be used in the establishment of management practices and stringent monitoring procedures in wastewater usage, and creation of awareness of the hazards and risks associated to the use of wastewater for vegetable production. Henceforth in order to ensure food safety for the urban and peri-urban in Zambia so as to decrease the exposure of heavy metals in vegetables, policy makers and other agencies concerned with standards of wastewater such as the National Water and Sanitation Council (NWASCO) and Ministry of Health (MOH) may be tasked to implement policies that safe guard the consumers.

The findings are also useful to other stakeholders, such as Lusaka water and sewerage in improving the design and refinement of wastewater reclamation and economic valuation tools which support changes in water management and planning in water pollution context, According to the results in the study, the wastewater released in to the Chunga stream does not comply with WHO standards. The wastewater released from the treatment plant is not processed thoroughly as concentrations of heavy metals in the wastewater are above set standards.

Members of the public may also use the findings of the research to make informed decision on consumption and preparation of the vegetables as the bioaccumulation index showed that *Cucurbita moschata* and *Brassica juncea* are vegetables that do not highly accumulate heavy metals as compared to *Brassica napus* and *Brassica rapa*. *Cucurbita moschata* and *Brassica juncea* can be safely grown for consumption.

In addition to the benefits stated above, the study has added to the existing body of knowledge on heavy metal contamination in wastewater, soil and vegetables around chunga gardens as well as the potential health risk along with the positive social change in the community.

1.4 RESEARCH QUESTIONS

1. What is the concentration of selected heavy metals in the wastewater, soil and vegetables around Chunga gardens in Lusaka District and their potential health risks?

2. What are the bioaccumulation factors of soil and vegetables around Chunga gardens in Lusaka District?

1.5 OBJECTIVES

1.5.0 General Objective

To assess heavy metals in wastewater, soil and vegetables around Chunga gardens in Lusaka and their potential health risks.

1.5.1.1 Specific Objectives

1. To establish the concentrations of heavy metals in the wastewater, soils and vegetables around Chunga gardens in Lusaka.

2. To determine the bioaccumulation factors of concentration of heavy metals in soil and vegetables around Chunga gardens in Lusaka.

3. To ascertain the potential health risks in the consumption of vegetables grown using wastewater around Chunga gardens in Lusaka.

1.6 CONCEPTUAL FRAMEWORK

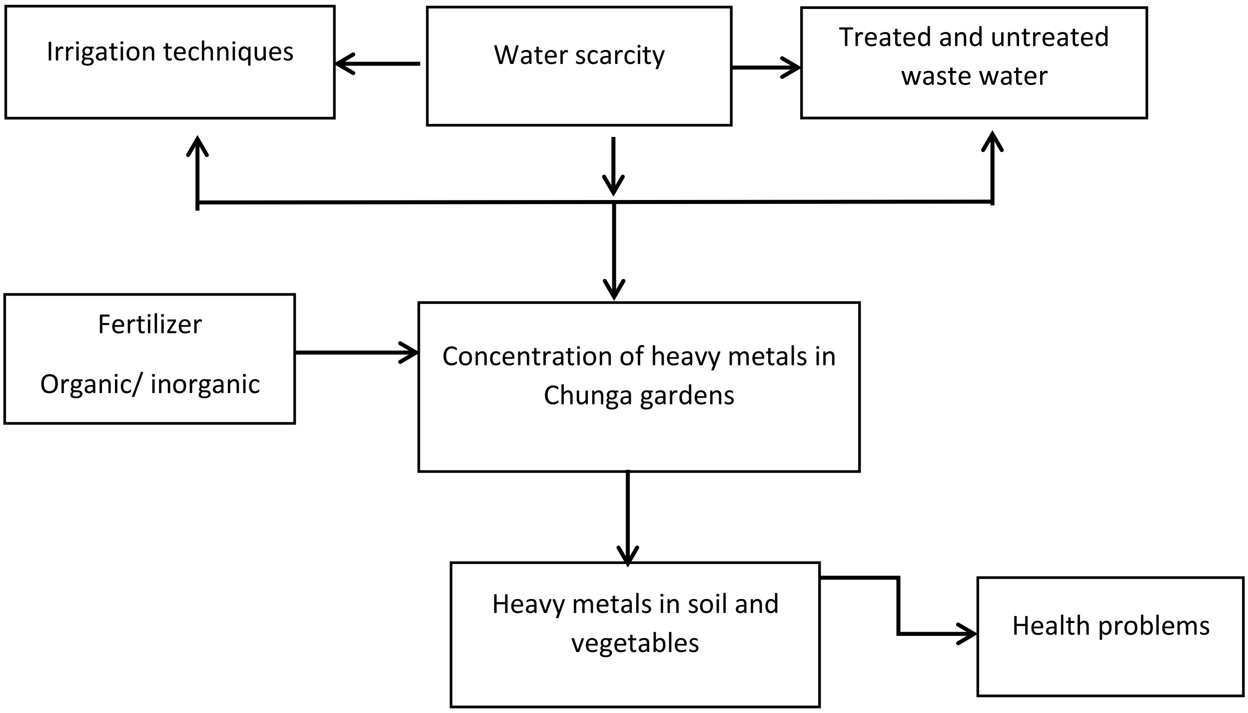


Figure 1 Depicts the Conceptual framework of heavy metal concentrations around Chunga gardens with plausible causes of high elemental concentration being attributed to variables such as wastewater running from the chunga treatment plant, with organic and inorganic fertilizer as a secondary factor, leading to high concentrations of different heavy metals in soil and vegetables. The heavy metals in the vegetables may lead to carcinogenic and non-carcinogenic human health implications. Adapted from Tembo & Cernak (2006).

CHAPTER TWO

LITERATURE REVIEW

2.1 OVERVIEW OF HEAVY METAL POLLUTION

Heavy metal contamination in waterbodies and the environment is of major concern and more prominent in medium sized cities in developing countries, primarily due to causative factors such as: irrigation with contaminated water, the addition of fertilizers, metal based pesticides and industrial emissions in the environment (Nazir et al. 2015).

According to Nazir et al. (2015), heavy metals constitute a heterogeneous group of elements which widely vary in their chemical properties and biological functions. Mora et al. (2009) reported that unlike organic pollutants, heavy metals do not decay and thus pose a different kind of challenge for remediation. Literature currently indicates that plants and microorganisms are tentatively being used to remove specific heavy metals such as mercury through bioremediation. Plants exhibit hyper accumulation for that reason they are used to remove heavy metals from soils by concentrating them in their bio matter (Farooq et al. 2012).

However in a developing country with limited laboratory equipment bioremediation is an expensive remediation method. As a result heavy metal pollution has been stated as one of the most prominent problems affecting Zambia and the effects manifest more in humans and animals. Suffice to mention that, certain heavy metals are essential to maintain the metabolism of the human body. Though at higher concentrations, they can be poisonous for instance heavy metals like copper, cadmium and lead are highly toxic in nature (Momodu & Anyakora, 2010).

A study conducted by Tembo & Cernak (2006) reveals that heavy metal pollution is a serious threat in Zambia, hence their study aimed at evaluating the spatial distribution of heavy metals in the main provinces of Zambia so as to understand the characteristics of the pollution in each province. Rivers, lake sediments and soil samples were collected from provinces constituting of water bodies and analysed for ten heavy metals (Cr, Co, Ni, Cu, Zn, As, Cd, Pb, Sr and Hg). The results indicated that heavy metal pollution in Zambia has strong regional differences. Using cluster analysis, the patterns of heavy metal pollution were divided into three major clusters: Central, Copperbelt, Southern and other areas. The result of the survey showed that, 81.29% of the samples in the three major clusters described irrigation wastewater as one of the major sources of heavy metal pollution in the environment.

2.2 WASTEWATER TREATMENT

Global assessment on wastewater treatment management systems in developing countries report that local authorities and other mandated service providers have the responsibility for the adequate management of wastewater treatment plants, however their institutional capacities to offer quality services remain questionable (Shah et al., 2012: Nhapia et al., 2012:Obodai et al., 2011). Institutional weakness to manage wastewater and sanitation services is evident where the population densities are extremely high, henceforth problems arising from wastewater cannot be avoided (Shah et al. 2012).

Tchounwou (2012) describes wastewater treatment as “a multi-stage process used to renovate wastewater before the effluent re-enters a body of water or is reused”. The definition suggests that the goal is to reduce and remove organic/inorganic matter, solids, nutrients, disease-causing organisms and other pollutants from wastewater. The interpretation by the Environmental Management Act of 2011 also points to the same understanding as tchounwou except that the legislation emphasises on discharging improved effluent quality to avoid pollution (Simutanyi, 2008). Although this is not explicitly said, it is implied to explain the reduction or removal of environmental pollutants to effluent standards permitted by the legislation. Therefore the standard and management of wastewater treatment plays a vital role in determining the amount of heavy metals released in the environment (Tembo & Cernk, 2006).

According to Simutanyi (2008), most wastewater treatment systems are based on natural processes. The treatment designs are aimed at treating organic pollutants and not inorganic.

In Zambia stabilization ponds are used in the treatment of wastewater which only treat the organic pollutants.

2.3 QUANTITY OF WASTEWATER

Charles (2004), conducted a study on the usage of wastewater for irrigation in vegetable growing in the Kafue lagoon areas along Ngwerere River, In April, May, June and July 2004 the average discharges from Manchinchi was 72, 545 m3/day, 58,805 m3/day, 39, 357 m3/day and 32, 803 m3/day, respectively. The design capacity of the treatment plant was 36, 000 m3/day. Therefore for April, May and June 2004 the design capacity was exceeded (Lusaka Water and Sewerage Company). As a result of overloading the treatment plant, the final effluent lost its quality to 59 % removal efficiency. The study results by Charles (2004), assumes that the management of municipal services in Lusaka, which is inclusive of wastewater treatment, is either inadequate or poorly functional, rendering the wastewater not usable for irrigation purposes.

2.4 CHARACTERIZATION OF HEAVY METALS

Metals are said to be normally present in relatively low concentrations, usually less than a few mg/l, in conventional irrigation waters and are called trace elements. The Trace elements are not normally included in routine analysis of regular irrigation water, however attention should be accorded to them when using sewage effluents, particularly if contamination with industrial wastewater discharge is suspected (Liang, 2011). Sardar 2013 noted that trace elements include Aluminium (A1), Beryllium (Be), Cobalt (Co), Fluoride (F), Iron (Fe), Lithium (Li), Manganese (Mn), Molybdenum (Mo), Selenium (Se), Tin (Sn), Titanium (Ti), Tungsten (W) and Vanadium (V) (WHO,2008).

Trace elements create definite health hazards when taken up by plants. Under this group of toxic metals include : Arsenic (As), Cadmium (Cd), Chromium (Cr), Copper (Cu), Lead (Pb), Mercury (Hg) and Zinc (Zn), while Heavy metals of highest public concern are Arsenic (As), Lead (Pb), Cadmium (Cd), Copper (Cu) , and Mercury(Hg), these metals are widely dispersed in the environment through different causative factors. The elements mentioned above have no beneficial effects in humans, and there is no known homeostasis mechanism for them (Sracek et al. 2012). They are generally considered the most toxic to humans and animals; the adverse human health effects associated with exposure to them, even at low concentrations, are diverse and include, but are not limited to, neurotoxic and carcinogenic actions (UNICEF/WHO, 2015).

2.5 UPTAKE OF HEAVY METALS IN VEGETABLES AND SOIL

The application of wastewater has led to changes in soil physicochemical characteristics and heavy metal uptake by food crops, particularly vegetables. The soil pH changes depends on the pH of the wastewater used for irrigation, and the soil pH has a great influence on the mobility and bioavailability of heavy metals (Tembo & Ceranak, 2006). Shah et al. (2012), noted that heavy metal contents vary among different vegetables at different sites, thus the variations in heavy metal concentrations in vegetables may be ascribed to the differences in their morphology and physiology for heavy metals uptake, exclusion, accumulation and retention.

Vegetables accumulate heavy metals in their edible and non-edible parts (Kribek et al. 2010), leafy parts of vegetables accumulate higher amounts of heavy metals than their fruits (Liu et al. 2013). Fasinu *&* Orisakwe (2013), Determined that the concentrations of heavy metals in vegetables per unit dry matter generally follow the order: leaves > fresh fruits > seeds.

Ikenka et al. (2013), pointed out that heavy metal accumulation in leafy vegetables is possibly high owing to the fact that the leaves are the main parts of the vegetables used for photosynthesis, for the reason that higher metal mass flows to the leaves due to strong transpiration and atmospheric heavy metal deposition. Khan et al. (2008), reported that vegetables grown at environmentally contaminated sites could take up and accumulate metals at concentrations that are probably toxic to human health. Their study was conducted to analyse the metal contents of some vegetables in Sahre Rey-Iran with emphasis on their toxicological implications. Recently matured leaf and fruit samples of Shahrerey vegetable farms were sampled and analysed to determine heavy metals. The results from their study showed that metal uptake differences by the vegetables are attributed to plant differences intolerance to heavy metals and vegetable species.

According to Akota et al. (2008), the extent to which vegetables take up heavy metals differs from one vegetable to the next. There are some vegetables that accumulate higher levels of heavy metals than others. The extent of accumulation differs within the type of heavy metals where by one vegetable may accumulate the highest concentration of one or more heavy metals but not all of them. However, within the *Brassica genus*, there also exist some other species which show the tendency to accumulate high metal concentrations, and which can be characterized as metal accumulators. Some of these species grow fast and produce a high biomass (Sardar et al. 2013). Examples are *Brassica juncea* (mpilu), *Brassica rapa* (Chinese cabbage) and *Brassica napus* (rape). *Brassica juncea* was shown to accumulate high levels of heavy metals including Cd, Cr, Cu, Ni, Pb and Zn under certain conditions which particularly enhance the solubility of metals in the soil (Mazumder, 2008). Nadeem *et al*. (2009), found that two *Brassica* species (*Brassica juncea and* Raphanus *sativus*) as well as (*Cucurbita moschata)* are moderately tolerant when grown on a multi-metal contaminated soil.

2.5.1 HEAVY METALS IN SOIL

According to Mora & Sanchez (2009) several experiments have been conducted to study the effect of soil parameters and other elements on mobility and bioavailability of heavy metals from soil to crop system. Mora & Sanchez (2009) stated that, the mobility and bioavailability of heavy metals in contaminated soil is affected by a few biological processes and physiochemical properties like soil pH, organic matter (OM), caution exchange capacity (CEC), soil texture, and soil microbiota.

Wu (2009) pointed that, Heavy metal concentrations are greatly affected by soil organic contents as soils with high organic waste concentrations are generally confined to heavy metal concentrations of less than 1000 mg kg−1 soil, while industrial waste-contaminated soil contains more than 10,000 mg kg−1 soil (Mahar et al. 2013). Increasing concentrations and variation in distribution of heavy metal in metal-amended soil is the main source of contamination of food the chain with heavy metals, as the soil is used as an important tool in the prime route of heavy metal intake into the human body through soil–crop uptake (Liu et al. 2007).

2.6 PERMISSIBLE LIMITS OF HEAVY METALS IN DIFFERENT MEDIA

The most appropriate wastewater to be used in irrigation should meet the recommended chemical quality guidelines both at low cost and with minimal operational and maintenance requirements. Vegetables and soil are equally recommended to have a certain threshold of chemical accumulations following the World Health Organisation Standards

Table 1: WHO guidelines for heavy metals and threshold values for soil, water and crops (mg/kg and mg/l)

Element	**Recommended Maximum Concentration for Heavy Metals in Agricultural soil, Wastewater and Crops**		
	Soil (mg/kg)	**Water (mg/l)**	**Crop (mg/kg)**
Cadmium	0.8	0.1	0.02
Copper	36	2.0	10.0
Lead	85	0.1	2.0

Zinc	50	5.0	5.0

Source: Adapted from National Academy of Sciences (1972) and Pratt (1972).(WHO, 2008)

2.7 POTENTIAL HUMAN HEALTH RISKS

Nhapia et al. (2012), reported that inorganic chemicals usually exist in municipal wastewaters at very low concentrations and ingestion over prolonged periods could lead to detrimental effects on human health. The principal of health hazards associated with the chemical constituents of wastewaters, therefore, arise from the contamination of crops or groundwater in high concentrations (UNICEF/WHO, 2015). Hillman (1988) noted that particular concern should be attached to cumulative chemicals, principally heavy metals as they give rise to the greatest health concern in agricultural use of wastewaters. Few epidemiological studies have established definitive adverse health impacts attributable to the practice. Heavy metal association as causative agents in diseases is underestimated due to the degree and length of time of exposure to the heavy metals.

2.7.1 Health Risk Assessment

Dietary intake of heavy metals through contaminated vegetables may lead to various chronic diseases. Kribek et al. (2010) noted that, Bioaccumulation of heavy metals in soft tissues interferes with normal physiological functions and generally exert their toxic effects by forming complexes with organic compounds. Their toxicity can damage or reduce mental and central nervous function, lower energy levels, and damage to blood composition, lungs, kidneys, liver and other vital organs. Long term exposure may result in slowly progressing physical, muscular, and neurological degenerative processes that cause muscular dystrophy, and multiple sclerosis (Liang, 2011).

One of the methods of measuring potential health risks of heavy metals is through conducting a human health risk assessment. A comprehensive health risk assessment process consists of four distinct phases: (1) hazard identification; (2) exposure assessment; (3) dose-response assessment; and (4) risk characterization. This method is considered as the characterization of the potential adverse health effects to humans as either non-carcinogenic or carcinogenic due to exposures from environmental hazards (USEPA, 2012). Non-carcinogenic chemicals are assumed to have a threshold; a dose below which no adverse health effects will be observed where an essential part of the dose-response portion of a risk assessment includes the use of a reference dose (RfD). Carcinogens are assumed to have no effective threshold. This assumption implies that there is a risk of cancer developing with exposures at low doses and, therefore, there is no safe threshold for exposure to carcinogenic chemicals (Francis et al. 2017).

Dietary exposure is one of the ways through which human beings are exposed to heavy metals thus to ascertain the dose ingested, the chronic daily intake (CDI) and hazards quotient (HQ) of heavy metals for the vegetables are calculated based on the equations for the hazard quotient using oral toxicity reference dose (RfD) values provided by US EPA (2005). If the value of HQ is less than 1 then the exposed population is said to be safe. In general, the oral Reference Dose (RfD) is an estimate (with uncertainty spanning perhaps an order of magnitude) of a daily exposure to the human population (including sensitive subgroups) that is likely to be without an appreciable risk of deleterious effects during a lifetime. The RfD is based on the assumption that thresholds exist for certain toxic effects such as cellular necrosis and is expressed in units of mg/kg/day (Francis et al., 2017 & Liu et al., 2013).

The Blacksmith institute project completion report of 2014 revealed that in the U.S normal blood levels of lead where less than 10 g/dl (micrograms per deciliter). They however noted that in Kabwe, blood concentrations of lead where 300 g/dl and recorded in children which posed carcinogenic adverse health effects. The records showed that average blood levels of children were between 60 and 120 g/dl. They also noted that children were especially vulnerable to acute, sub-acute and chronic effects of ingestion of chemical pollutants as well as cancer, as they are likely to consume more (twice of the amount) of food per unit of body weight as adults.

While studies in China, attributed the use of contaminated industrial wastewater for crop production to be associated with 36% increase in hepatomegaly (enlarged liver), and a 100% increase in both cancer and congenital malformation (Rajanker et al., 2014). In Japan, Itai-itai disease, a bone and kidney disorder, was associated with chronic cadmium pollution of paddy water coming from the Jizu River (Liu et al., 2013) thus leading to both non- carcinogenic and carcinogenic adverse health effects.

Yube et al. (2010), Studied the potential health risk of heavy metals in vegetables grown in agricultural soil irrigated with polluted water of the Shitalakhya river in Narayangonj, Bangladesh. Agricultural soil and vegetables were analysed for copper (Cu), nickel (Ni), cadmium (Cd), chromium (Cr), lead (Pb) and zinc (Zn).The study highlighted that daily intake metals (DIM) values compared with oral reference dose, suggested that the consumption of vegetables grown in agricultural soil is nearly free of risks. Health risk index (HRI) values of the studied heavy metals were <1, indicating that there is a relative absence of health risks

CHAPTER THREE
METHODOLOGY

3.1 STUDY SITE AND POPULATION

The study was conducted in Zambia in Lusaka being the capital city and one of the fastest growing cities in sub-Saharan Africa. The country's population as of 2018 is estimated at 17.61 million, while Lusaka city's population is 2.5 million. Less than 50% live in the metropolitan area, the rest on the urban fringe or satellite areas. The city is quite widespread and covers a surface area of about 400 square kilometres. Chunga Township is among the many peri-urban areas located in the north east side of Lusaka District. The study site was purposively selected as it houses the Chunga wastewater treatment plant and the Chunga stream. The Chunga wastewater treatment plant comprises a biological filtration system serving mainly the domestic catchment within Lusaka. The plant no longer functions properly, mechanically or process wise, resulting in little treatment of the influent flows. The Chunga stream is one of the most polluted streams due to improper discharge of end effluents from the Chunga treatment plant which deteriorates the quality of the water in the Chunga stream, thus the wastewater is secondary wastewater and is easily accessed by the farmers as the stream is located along the gardens. Control samples for vegetables were collected from a garden 40 metres before Chunga wastewater treatment plant on the side of the plant where the waste water is not discharged. For this study wastewater is based on the fact that the dilution ratio of the effluent in to the Chunga stream is high. Thus the stream has a small capacity to hold the volume of wastewater released from the Chunga treatment plant. The common vegetables grown in this area are *Brassica napus (*Rape), *Brassica juncea* (Mpilu), *Brassica rapa* (Chinese cabbage) and *Cucurbita moschata* (Pumpkin leaves) .The vegetables are grown in the area throughout the year and are used either for home consumption or supplied to the local market in Chunga and Soweto market.(CS0, 2011).

Figure 2 Depicts the 9 sampled gardens for the study from the control garden.

3.2 STUDY DESIGN

This was a cross sectional study. It was carried out at one time point or over a short period. The Data was collected from April to July 2019 to provide a 'snapshot' of the outcome and the characteristics associated with it. Wastewater samples, soil samples and vegetable samples were collected from Chunga gardens.

3.3 STUDY VARIABLES

This section describes the types of variables which were included in the study being: dependent variable and independent variables as can be seen in the table 2:

Table 2: Study Variables

Variable	**Indicator**	**Scale of Measurement**
Heavy Metal	Presence of Heavy Metal	Concentration of Heavy Metals in mg/kg/mg/l. Yes/No Binary
• Heavy metal concentration in vegetables	• In accordance with WHO/FAO permissible limits of metal accumulation in vegetables Refer to table 1 for details	Compliant/Non-Compliant Binary
• Concentration of heavy metals in soil	• In accordance with WHO/FAO permissible limits of metal accumulation in vegetables	Compliant-/Non-Compliant- Binary

Variable	Indicator	Scale of Measurement
	Refer to table 1 for details	
• Concentration of heavy metals in wastewater	• In accordance with WHO permissible limits of metal accumulation in vegetables Refer to table 1 for details	Compliant/Non-Compliant Binary
• Bioaccumulation factor	• BF > 1 • BF< 1	Accumulator/ Excluder Continuous
• Health risk	• HQ> 1 HQ < 1	Risk/Non Risk Continuous

Cucurbita moscahta- Pumpkin Leaves: *Brassica rapa*- Chinese cabbage: *Brassica napus*- Rape: *Brassica juncea*- Mustard Green Mpilu

3.4 STUDY POPULATION

Wastewater, soil and mature vegetables grown using wastewater during the study around Chunga garden’s in Lusaka.

3.4.1 Inclusion Criteria

Only mature vegetables and soil around Chunga gardens as well as wastewater not more than 10km away from the Chunga treatment plant.

3.4.2 Exclusion Criteria

All vegetables sprayed with pesticides or fertilizers at the time of the study and soil whose owners did not consent to be part of the study were excluded in addition to wastewater less than 2m depth.

3.5 SAMPLE SIZE AND SAMPLING METHODS

3.5.1 Vegetable Sampling

The gardens along the Chunga stream of not less than 10km from the Chunga treatment plant were picked, for this study only nine gardens were available at the time of the data collection. The available gardens were stratified into strata of four different types of leafy vegetable samples. The vegetables, were then collected based on the beds of the gardens available at the time of the study. The beds included in the study were sampled using simple random sampling. The raffle method was used to pick a bed from each garden that consisted of more than two beds of the same type of vegetable. Vegetable samples were collected at the four edges and at the middle to create a composite sample. The vegetables were collected within 10km radius from the Chunga wastewater treatment plant. Leafy vegetables were preferred for sampling as past research indicates that they accumulate heavy metals at a greater capacity than other vegetables (US EPA, 2012).Three to four types of leafy vegetables were collected from the available gardens. The gardens were sampled within a radius of 4km from the beginning point of the reference garden irrigated without wastewater, as well as after the Chunga treatment plant. A final composite sample was constituted by the laboratory. A total of 28 vegetables samples were collected from 9 vegetable gardens, the vegetable samples included *Brassica napus* (rape), *Brassica rapa* (Chinese cabbage), *Brassica juncea* (Mpilu) and *Cucurbita moschata* (Pumpkin leaves).

3.5.2 Soil Sampling

Eight surface soil (0-20 cm) samples were collected at the same point as the vegetable samples, 1kg soil samples were collected at the middle point of the beds, according to EPA (2012) standards and packed into labelled polyethylene bags.

3.5.3 Wastewater Sampling

Wastewater samples were sampled 1 meter off shore from the stream. The sampling point was picked based on the midpoint of the garden, at a uniform distance measured using the beginning and end point of the garden. 500mls of nine wastewater samples were collected at a depth of 2m. The water samples were transported for analysis at the University of Zambia School of Agriculture laboratory on the same day.

3.6 DATA COLLECTION, MANAGEMENT AND QUALITY CONTROL

3.6.1 Data Collection

3.6.1.1 Vegetables Collection

Samples of commonly grown vegetables, eight samples of Rape (*Brassica napus*), four samples of Mpilu (*Brassica juncea*), eight samples of Chinese cabbage (*Brassica rapa*) and eight samples of pumpkin leaves (*(Cucurbita moschata)* were collected in pre-cleaned zipper bags. For metal analysis, only the edible parts of the vegetable samples were used. The freshly harvested mature vegetables were taken to the laboratory and thoroughly washed with running tap water and rinsed with deionized water to remove any soil particles attached to the plant surfaces. The vegetable samples were transported to the University of Zambia School of Agriculture laboratory for analysis on the same day.

3.6.1.2 Soil Collection

Surface soil (0-20 cm) samples were collected using a spade. The spade was inspected for possible cross contamination and cleaned with ambient water for individual sample collection. The soil was sampled from the central part of the bed in the gardens and transferred to a pre-cleaned plastic container (US EPA, 2012). The eight surface soil samples were collected in pre-cleaned zipper polythene bags, which were kept in airtight large plastic containers. The soil samples were transported for analysis at the University of Zambia School of Agriculture laboratory on the same day.

3.6.1.3 Wastewater Collection

Wastewater samples were collected from the Chunga stream which is used to dispose of end effluent from the Chunga treatment plant. Wastewater was collected in 500mls bottles obtained from the laboratory. A contraption was used that was attached to a 3.5m pole to collect the wastewater at the mid- point using a uniform distance. Approximately 100 to 200ml of wastewater were filled in the bottle samplers.

For the reference group 100 to 200mls of fresh water were filled in the bottle samplers. The fresh water was collected at a distance further away from the Chunga treatment plant. The wastewater and freshwater was collected at a depth of 3m and stored into labelled polyethylene bags at room temperature for laboratory analysis.

The wastewater samples were transported for analysis at the University of Zambia School of Agriculture laboratory day.

3.7 MEASUREMENTS OF THE PARAMETERS

3.7.1 Laboratory Analysis

The samples were analysed at the soil and crops laboratory of the School of Agriculture at the University of Zambia. The samples were transported to the laboratory within 7 hours of sampling. This was to ensure quality control of the samples. The parameters analysed are shown in Table 3 including the methods of analysis used. The heavy metals were analysed using the Atomic Absorption Spectrophotometer.

3.7.2 SAMPLE PREPARATION

Sample preparation was used to ensure the removal of organic impurities from the samples and thus prevent interference in analysis. Preparation of the samples was one of the storage steps taken to preserve the samples from bacterial activities and to release metals into the analytical solution (Simutanyi, 2008).

3.7.2.1 Vegetable Digestion

The vegetable samples were washed with distilled water and subsequently rinsed to eliminate all contaminants including air-borne pollutants. All washed vegetable (edible part) samples were chopped into small pieces and then dried using an electric oven at the temperature range between (60-70) ˚C for 24 hours. The dried samples of vegetables were then grounded using an electric grinder in order to obtain the final powder. The obtained powder was sieved through a 0.4 mm polystyrene sever and kept well into labelled polyethylene bags ready for measurements. The vegetable samples were weighed using a digital balance to obtain 6.0 g of the sieved powder for vegetable samples. The samples were mixed with cellulose in the binder in for proper binding. In order to get the exact weight of the mixtures, 1g for vegetable leaf sample was weighed to make the analyst of approximately 7.5 g respectively.

3.7.2.2 Soil Digestion

The soil samples were air dried for 72 hours and oven dried for 24 hours at 25˚C in the laboratory, at room temperature to obtain constant weight. The dried soil samples were then grounded using an electric grinder and sieved through 2.2 mm polystyrene sieve and kept into labelled polyethylene bags ready for further analysis. The soil samples were weighed using a digital balance to obtain 12.0g of the sieved powder. The samples were mixed with a cellulose binder in the ratio 4:0:9 to obtain 2.7g weight of the soil samples to make an analyst of approximately 14.7g. A final sub-sample of the soil was obtained by quartering (Van & New, 2004).

3.7.2.3 Atomic Absorption Spectrometry of the Vegetable and Soil Samples

The mixed powders of the vegetables and soil samples were put into a pulveriser for mixing and homogenising purposes. A manual hydraulic press machine was used, the pellets of the soil and vegetable leaves samples were put at a pressure of 15 tonnes and 10 tonnes respectively in the hydraulic press machine. The pellets used were of intermediate thickness with outer diameter of 32 mm. The pellets were finally kept into the labelled sample holders and then placed in the Atomic Absorption Spectroscopy machine for analysis.

The heavy metal concentrations were measured using the Atomic Absorption Spectroscopy which provides a sensitive means of determining more than seventy elements. About 1 g of the vegetable and soil sample was weighed, Aqua Regia solution was added to about 30ml (a mixture of one part concentrated Nitric acid and three parts concentrated Hydrochloric Acid). The crushed vegetable and soil samples were heated on a hot plate above 400 degrees Celsius for 15 minutes or when it neared dryness. A conical flask was used after being washed to avoid contamination, after which the samples were filtered into 100ml volumetric flask. Finally, the Atomic Absorption Spectrophotometer was calibrated with Known standards to ensure a linear curve was obtained and finally run the samples on the Atomic Absorption Spectrophotometer for heavy metal concentration.

3.7.2.4 Wastewater Digestion

All the water samples were filtered after adding 5 drops of 10% nitric acid. The filtrates were then used to determine the different elements on the AAS. Concentration of each trace element was determined by Atomic Absorption Spectrometer using relevant standards for each element on a PERKIN ELMER Model Analyst 400 AAS with suitable lamps for Cu, Cd, Zn and Pb. Relevant dilutions were made when needed. The standard concentrations used to calibrate the AAS for each of the analysed elements are given below. Cu: 0, 5.0, 15.0, 30.0 mg/L, Zn: 0, 0.5, 1.5, 3.0 mg/L. Pb: 0, 5 mg/l, Cd: 0, 2mg/l.

In view of data quality assurance; average values of three replicates were taken for each determination and after every 10 samples, Blanks were prepared to check for background contamination by the reagents used.

The above information has been provided by the school of agriculture laboratory at the University of Zambia.

3.8 DATA MANAGEMENT

The data was entered in excel spreadsheet and exported to STATA software Version 14 for analysis. Data normality was checked using a histogram plot in Stata. Descriptive statistics such as measures of central tendency were reported for continuous data using medians and interquartile ranges to summarize heavy metal concentrations. Frequencies were reported to summarize categorical data which included the type of vegetables and heavy metal elements as well as number of gardens, wastewater samples, and soil in addition to vegetables samples. Non-parametric statistical tests were used as the heavy metal concentrations were skewed. Kruskall Wallis test was used to compare metal concentrations between the wastewater samples, vegetable and soil samples. All the tests were done at 95% confidence level.

Bioaccumulation factor (BF) for the vegetables irrigated with wastewater was calculated using the equation below adapted from the US EPA 2011:

$BF = Cv/Cs$

Where: Cv is the mean metal concentration in the vegetable sample (mg/kg) and Cs is the Mean metal concentration in the soil sample (mg/kg). If the BF is grater > 1 then the vegetables are found be accumulators of heavy metals, while a bio-accumulation factor less than < 1 means that the vegetable is an excluder of accumulation of heavy metals.

Human health risk assessment was calculated using the equations from the U.S. Environmental Protection Agency’s (USEPA) Risk Assessment Guidance for Superfund, Volume I, and Human Health Evaluation Manual (1989). Values used in the Equation were taken from the Environmental Protection Agency's Exposure Factors Handbook (1997) (US EPA, 2004).

Equation $$HR1 = C\,\frac{CF \times IR \times FI \times \ EF \times ED}{BW \times AT}$$

Where *CF*= Contaminant concentration in food (mg/kg), *IR*= Ingestion rate (kg/meal), *FI*= Fraction ingested from contaminated source (unit less), *EF*= Exposure frequency (meals/year), *ED*= Exposure duration (years), *BW*= Body weight (kg), and *AT*= Averaging time (period over which exposure is averaged – days).

The Human health risk assessment consisted of assessing non-carcinogenic risk assessment and Carcinogenic risk assessment. Non-carcinogenic risk referred to chemical substances which are characterized by a threshold below which the body is able to cope with or recover from the exposure. A brief or low exposure leaves no consequence until the next exposure. Non-carcinogenic risk assessment was determined through the calculation of chronic daily intake (CDI) using the equation above and the values for the USEPA depicted in table 3.The non-carcinogenic hazards quotient (HQ) were determined by using the formula: HQ = CDI/ RfD. Where the oral toxicity reference dose (RfD) values were provided by United States Environmental Protection Agency (US EPA, 2012). If the value of HQ is less than 1 then the exposed population is said to be safe.

Table 3: The following Parameters were used for estimating exposure assessment in vegetables (Koki et al., 2015; Liu et al., 2007; Wongsasuluk et al, 2013)

Parameter	**Resident**	**Worker**
CR	2 L/day drinking water	1 L/day drinking water
	100 mg/day soil and dust ingestion	50 mg/day soil and dust ingestion
	30 m3/day air inhalation	30 m3/day air inhalation
EF	350 days/year	250 days/year
ED	Actual event duration	Actual event duration
	or 30 years if chronic	or 25 years if chronic
BW	70 kg (adult), 15 kg (child)	70 kg
AT	Actual event duration if not carcinogenic	
	or 365 days/year x 70 years if carcinogenic	

Source: USEPA, 1989; Nazaroff & Alvarez-Cohen, page 571)

Table 4: The toxicity responses (dose response) to heavy metals and the oral reference dose (RfD) and oral slope factor (SF) (USEPA, 2011)

Heavy Metal	**Oral *RfD* (mg/kg/day)**	**Oral *SF* (mg/kg- day)-1**
Cadmium	0.005	0.38
Copper	0.04	n.d
Lead	0.0005	8.5
Zinc	0.3	n.d

n.d = not determined

For cancer risk the equation below was employed

Cancer risk =CDI ×CSF ((Liang et al., 2011; Wu et al., 2009; Asare‐ Donkor et al., 2016).

The linearized multistage model assumptions estimates the risk of getting cancer, which is not necessarily the same as the risk of dying of cancer, so it should be even more conservative as an upper-bound estimate of cancer deaths. Potency factors used in this study were found in the EPA database on toxic substances called the Integrated Risk Information System (IRIS). Thus, Cancer risk in our study represents the probability of an individual lifetime health risks from carcinogens.

CDI is the chronic daily intake of carcinogens (mg kg -1 d -1); CSF is the cancer slope factor of hazardous substances (mg kg -1 d -1). The newly Cadmium and Lead cancer slope was provided by US EPA being 0.38 and 8.5 (USEPA, 2004; Asare‐ Donkor et al., 2016). The cancer slope factor (CSF) unlike the reference dose for non-carcinogenic health risk assumes that exposure to any amount of a carcinogen will increase the risk of cancer, i.e., there is no safe or threshold dosage (Liu et al., 2013). A cancer slope factor (CSF) is an upper bound, approximating a 95% confidence limit, on the increased cancer risk from a lifetime exposure to a toxicant by ingestion, dermal or inhalation exposure route (Liu et al., 2013). The acceptable or tolerable risk for regulatory purposes is within the range of 10 -6 to 10 -4 (i.e., 1 case of cancer per every 1,000,000 to 1 case of cancer per every 10,000) (US EPA, 2011).

Therefore, on average the probability of developing cancer is that approximately 1 person per 1,000,000 will develop cancer as a consequence of the exposure (Asare‐ Donkor et al., 2016). Slope factors for heavy metals considered in this study have not yet been determined by USA EPA apart for Cadmium and lead. Therefore, only cancer risks (CR) due to exposure to Cadmium and lead were calculated.

Cancer risk was determined using the formula: C = CDI×SF, where SF is the slope factor as provided by US EPA guidelines (Liu *et. al.*, 2013). The acceptable or tolerable risk for regulatory purposes is within the range of 10 -6 to 10 -4 (i.e., 1 case of cancer per every 1,000,000 to 1 case of cancer per every 10,000) (US EPA, 2011).

Hazard quotients and cancer risk for adult males, adult females and children were calculated. Those which had Hazard Quotient above one were recorded. It was important to calculate the Hazard Quotient for each sex as the risks differed according to gender and growth stage. The results were presented in form of tables.

3.9 ETHICAL CONSIDERATION

Ethical approval of the study was obtained from the University of Zambia Biomedical Research Ethics Committee (UNZABREC) before proceeding for data collection. Permission was obtained from Lusaka City Council before commencement of data collection. During the wastewater, vegetable and soil collection, the vegetables were bought at the market price from the garden owners and permission was obtained from the garden owner or any adult who was present and above the age of 18 years to collect the vegetable and soil samples, while permission was obtained from Lusaka Water and Sewerage to obtain the wastewater samples. Participation in the study was voluntary, written consent was attained from the participants willingly. For this study, identification numbers were given to the gardens to aid the process of labelling the sampling bags at each sampling site. There were no foreseeable physical risks to humans involved in this study though it may have invoked anxiety and fear with a view that the study may bring halt to the participants business. However, the participants were assured that the findings of this study were availed to the relevant authorities for appropriate action without compromising their business and for academic purposes. This study was beneficial to the community as it was carried out to provide information on the quality of the wastewater, vegetables and soil with regards heavy metal concentration. Further, the records of the study were kept strictly confidential. All electronic information was coded and secured using a password protected file. In the event the results obtained show that the vegetables pose a health risk, the study results will be published to allow health policy makers and the community members to make informed decisions on the consumption of the vegetables.

4.0 DATA DISSEMINATION

To ensure that the outputs from the research informs and thereby maximise the benefit to the public, National Water and Sanitation Council (NWASCO), Food and Drug laboratory, Ministry of Health, School of Public Health and Lusaka Water and Sewerage Company. The following dissemination strategy will be used, to translate knowledge into practice. This will include interactive workshops with small scale farmers within Chunga on implementation of good practice guidelines on wastewater reclamation. Development of links with key organisations such as United Nations, World Vision, Food Agriculture Agency and Quality Observatories to contribute and capitalise on their networks. Use of electronic media such as websites and social media inclusive of Twitter, Instagram and YouTube/TED will be used to disseminate the information. International conference presentations as well as publications including plain english summary reports of the research, peer review journals, policy brief and local newsletters will be used.

CHAPTER FOUR

PRESENTATION AND INTERPRETATION OF RESULTS

5.1 Description of wastewater, soil and vegetable samples

Table 5 shows the number of gardens and samples, included in the study. Nine gardens were used to collect eight soil samples and 28 vegetable samples which included eight samples of *Brassica napus* as well as eight samples of *Brassica rapa*, four samples of *Brassica juncea* and eight samples of *Cucurbita moschata*. Eight wastewater samples and one fresh water sample were collected from the gardens around the Chunga treatment plant and the Chunga stream, a total of 45 samples were composed for the study. Four heavy metals were analysed being cadmium, copper, lead and zinc in the water samples, soil samples and vegetable samples.

Table 5: Description of number of gardens, types of vegetables, soil and wastewater samples collected per garden.

Garden Number	**Distance m/km**	**Vegetable Samples mg/kg**				**Soil Samples mg/kg**	**Wastewater Samples mg/l**
		Cucurbita moschata	***Brassica rapa***	***Brassica napus***	***Brassica Juncea***		
G1	**40m**	√	√	√	√	×	√
G2	**60m**	√	√	√	×	√	√

G3	**80m**	√	×	√	√	√	√
G4	**200m**	√	√	√	√	√	√
G5	**400m**	√	√	√	×	√	√
G6	**800m**	√	√	√	×	√	√
G7	**860m**	×	√	√	√	√	√
G8	**900m**	√	√	×	×	√	√
G9	**1.2km**	√	√	√	×	√	√

×- Not collected: √- Collected: m- meters: km- kilometres: Brassica -juncea (Mustard Greens/Mpilu): Brassica-rapa (Chinese cabbage): Cucurbita -moschata (Pumpkin Leaves): Brassica-napus (Rape)

Table 6: Median and Interquartile Range concentrations of Heavy Metals in Vegetables, Wastewater and Soil S.

Heavy metal	**WHO standards**			**Vegetables**	**Wastewater**	**Soil**
	Soil	**water**	**crop**	**Median(IQR)**	**Median(IQR)**	**Median(IQR)**
Cadmium	0.8	0.1	0.02	0.1(0.01, 0.09)	0.1 (0.1, 0.3)	5.5 (4.8, 19)
Copper	36	2.0	10.0	32.55(24.97, 42.49)	0.1 (0.1, 0.01)	14.4(12, 38.6)
Lead	85	0.1	2.0	2.255(0.01, 11.2)	0.195(0.19,0.27)	12.7(10.4, 17.9)
Zinc	50	5.0	5.0	37.205(33.96,48.59)	0.01(0.01, 0.03)	21.8(11.5, 68.6)

Interquartile range (IQR)

Table 6 shows the median and interquartile range (IQR) of heavy metal concentrations for wastewater, soil and vegetables. In both wastewater and vegetables copper as well as cadmium levels were below detectable limit. Zinc and Lead were more dispersed in the wastewater, soil and vegetable samples.

5.3 Heavy Metal Concentrations in Vegetables, Soil and Wastewater Samples

Table 7: Heavy Metal Concentrations in Vegetables (mg/kg) from Chunga Gardens Compared To WHO Standards

Site	Vegetable Sample	Heavy metal concentration in vegetables (mg/kg)			
		Cadmium	**Copper**	**Lead**	**Zinc**
	Brassica juncea				
G1	(Control) *Brassica j*	0.01	30.02	2.255	61.325
G3	*Brassica j*	0.01	0.94	12.415	45.78
G4	*Brassica j*	0.01	20.365	9.595	23.15
G7	*Brassica j*	0.09	41.46	16.365	59.38
	Brassica napus				

G1	(Control) *Brassica n*	0.01	30.505	19.185	54.635
G2	*Brassica n*	0.09	25.99	10.27	59.99
G3	*Brassica n*	0.09	32.335	11.85	40.605
G4	*Brassica n*	0.01	23.9	0.01	36.89
G5	*Brassica n*	5.08	81.25	5.08	49.33
G6	*Brassica n*	0.01	94.17	0.01	37.205
G7	*Brassica n*	0.01	21.275	0.01	26.495
G8	*Brassica n*	0.01	20.58	0.01	30.22
G9	*Brassica n*	0.01	61.455	0.01	33.965
	Cucurbita moschata				
G1	(Control) *Cucurbita*	0.01	24.97	11.285	51.99
G2	*Cucurbita m*	0.01	13.905	13.545	48.595
G3	*Cucurbita m*	0.01	34.275	13.545	34.545
G4	*Cucurbita m*	0.565	67.935	0.565	41.155
G5	*Cucurbita m*	0.01	42.49	0.01	35.94
G6	*Cucurbita m*	0.01	33.25	0.01	33.24
G8	*Cucurbita m*	0.01	38.38	0.01	34.37
G9	*Cucurbita m*	0.01	36.975	0.01	37.63
	Brassica rapa				
G1	(Control) *Brassica r*	0.01	23.845	2.255	35.48
G2	*Brassica r*	1.24	416.185	254.515	418.26
G4	*Brassica r*	0.01	25.345	0.01	27.255
G5	*Brassica r*	0.01	85.025	7.335	46.965
G6	*Brassica r*	0.01	54.84	0.01	46.02
G7	*Brassica r*	0.01	25.345	10.72	24.36
G8	*Brassica r*	0.01	32.55	10.57	35.36
G9	*Brassica r*	0.01	34.22	0.01	30.235
Permissible levels as per WHO	WHO-ML	0.2	10.0	2.0	5.0

WHO-ML – World Health Organisation maximum limits: Brassica J- Juncea (Mustard Greens/Mpilu): Brassica R- Rapa (Chinese cabbage): Cucurbita M- Moschata (Pumpkin Leaves): Brassica-Napus (Rape)

The concentrations of heavy metals in vegetables irrigated without wastewater and with wastewater, are given above in table 7. Out of the total 28 vegetable samples analysed in the nine gardens, Concentrations of heavy metals in zinc ranged between 418.26mg/kg – 23.15mg.kg. Copper ranged between 416.185mg/kg - 0.94mg/kg, lead ranges were found between 254.515 – 0.01 with cadmium at 5.08mg/kg – 0.01mg/kg in the vegetable samples.

In the control group for the vegetables zinc values ranged between 61.325mg/kg – 35.48mg/kg, copper ranged between 30.505mg/kg – 23.845mg/kg. Lead was found at 19.185mg/kg – 2.255 with cadmium at 0.1mg/kg.

Heavy metal concentrations in the vegetables irrigated with and without wastewater took the following order *Zn<Cu<Pb<Cd.*

All the vegetables samples with zinc concentrations were not compliant with the World Health Organisation (WHO) Standards as well as 96% (28) of copper, 57% (16) lead and 21% (6) cadmium were non-compliant in the vegetables out of 28 samples.

Table 8: Heavy Metal Concentrations in Wastewater (mg/l) from Chunga Gardens Compared to WHO Standards

Site	**Wastewater Sample**	**Heavy metal concentration in Wastewater (mg/l)**			
		Cadmium	**Copper**	**Lead**	**Zinc**
G1	(control) *freshwater*	0.01	2.59	0.01	0.60
G2	A	0.03	0.01	0.66	0.01
G3	B	0.01	0.01	0.17	0.01
G4	C	0.03	0.01	0.19	0.01
G5	D	0.01	0.01	0.24	0.02
G6	E	0.01	0.01	0.32	0.01
G7	F	0.01	0.01	0.27	0.03
G8	G	0.04	0.01	0.19	0.03
G9	H	0.02	0.01	0.20	0.01
Permissible levels as per WHO	WHO-ML	0.1	2.0	5.0	0.1

WHO-ML – World Health Organisation maximum limits: G- Garden:

Out of the 8 samples tested for the wastewater; Lead was the metal in highest concentration ranging between 0.66mg/l - 0.01mg/.Copper had negligible values in the wastewater being 0.01mg/l. Cadmium and Zinc concentrations values were found between at 0.4mg/l - 0.03mg/l and 0.03mg/l - 0.01mg/l in the wastewater, while copper had the highest value in the control group of fresh water being 2.59mg/l, The lowest value in the reference group was 0.01mg/l for cadmium and lead, with zinc at 0.60mg/l (table 8).Heavy metal concentrations in the water were in the following decreasing sequence *Cu<Pb<Zn<Cd*, 88% (8) of lead, 11% (1) copper,33% (3) zinc and 44% (4) of cadmium out of 9 samples were not compliant with the with the World Health Organisation Standards (WHO) in water.

Table 9: Heavy Metal Concentrations in Soil from Chunga Gardens Compared to WHO Standards

Site	**Soil Sample**	**Heavy metal concentration in Soil (mg/kg)**			
		Cadmium	**Copper**	**Lead**	**Zinc**
G1	A	1.40	7.50	14.20	12.20
G2	B	19.00	14.90	11.20	42.40
G3	C	4.90	12.00	8.70	9.90
G4	D	4.80	113.00	12.70	11.50
G5	E	5.50	14.40	10.40	21.80
G6	F	21.10	14.20	17.90	68.60

G7	G	8.00	38.60	42.20	68.70
G8	H	0.10	12.10	8.20	73.90
Permissible levels as per WHO	WHO-ML	0.8	36.0	85.0	50.0

WHO-ML – World Health Organisation maximum limits: G- Garden:

The highest concentration for soil was copper ranging between 133.00 mg/kg - 2.59mg/kg. Zinc values ranged at a concentration of 73.90mg/kg - 0.87mg/kg. Lead and cadmium values were found in the range of 42.20 mg/kg - 8.20mg/kg and 19.00mg/kg – 0.10mg/kg (Table 9).

Heavy metal concentrations in the soil were in the following order *Cu<<Zn<<Pb<<Cd*. Out of the 8 samples cadmium was 100% (8) non-compliant with World Health Standards (WHO) in soil as well as 25% (2) in copper and 37% (3) in zinc.

5.3 Comparison of Heavy Metal Concentrations in Wastewater, Soil and Vegetable Samples.

Comparison of levels of heavy metals in wastewater, soil and vegetable samples was done using the Kruskal–Wallis one-way analysis-of-variance test.

Table 10: Kruskal–Wallis Comparison of Heavy Metals in Wastewater, Soil and Vegetable Samples

Heavy Metal	H statistics degrees of freedom /P. Value
Cadmium	0.2428
Copper	0.8443
Lead	0.2349
Zinc	0.582

According to table 10 the median concentrations of heavy metals in soil, wastewater, and vegetables were not statistically significant.

5.4 Bio-Accumulation Factors for the Vegetables Irrigated with Waste Water from Chunga Gardens.

Table 11: Bioaccumulation Factors for Each Vegetable Sample from Chunga Gardens

Garden	Vegetable sample	*BF cd*	*BF cu*	*BF pb*	*BF zn*
G2	*Cucurbita moschata*	0.071 (*E*)	1.854 (*A*)	0.909 (*E*)	3.983 (*A*)

G3	*Cucurbita moschata*	0.526 (*E*)	2.300 (*A*)	1.209 (*A*)	0.184 (*E*)
G4	*Cucurbita moschata*	0.115 (*E*)	5.660 (*A*)	0.807 (*E*)	4.157 (*A*)
G5	*Cucurbita moschata*	0.008 (*E*)	3.760 (*A*)	0.787 (*E*)	3.125 (*A*)
G6	*Cucurbita moschata*	0.181 (*E*)	2.309 (*A*)	0.961 (*E*)	1.524 (*A*)
G7	*Cucurbita moschata*	0.125 (*E*)	0.994 (*E*)	0.236 (*E*)	0.500 (*E*)
G8	*Cucurbita moschata*	0.001 (*E*)	3.557 (*A*)	0.012 (*E*)	0.478 (*A*)
G2	*Brassica napus*	0.064 (*E*)	1.401 (*A*)	0.689 (*E*)	4.892 (*A*)
G3	*Brassica napus*	0.004 (*E*)	2.170 (*A*)	1.058 (*E*)	0.957 (*E*)
G4	*Brassica napus*	0.002 (*E*)	1.999 (*A*)	0.001 (*E*)	3.726 (*A*)
G5	*Brassica napus*	1.054 *(A)*	2.160 (*A*)	0.467 (*E*)	4.289 (*A*)
G6	*Brassica napus*	0.181 (*E*)	6.538 (*A*)	0.961 (*E*)	1.706 (*A*)
G7	*Brassica napus*	0.047 (*E*)	1.496 (*A*)	0.558 (*E*)	0.391 (*E*)
G8	*Brassica napus*	0.125 (*E*)	1.496 (*A*)	0.558 (*E*)	0.391 (*E*)
G2	*Brassica juncea*	0.526 (*E*)	1.366 (*A*)	0.856 (*E*)	0.545 (*E*)
G8	*Brassica juncea*	0.001 (*E*)	5.078 (*A*)	0.012 (*E*)	0.509 (E)
G3	*Brassica Juncea*	0.002 (*E*)	0.078 (*E*)	1.773 (*A*)	4.625 (*A*)
G2	*Brassica rapa*	0.885 (*E*)	2.244 (*A*)	0.916 (*A*)	4.283 (*A*)
G3	*Brassica rapa*	0.002 (*E*)	2.112 (*A*)	0.014 (*E*)	2.753 (*A*)
G4	*Brassica rapa*	0.207 (*E*)	6.130 (*A*)	0.577 (*E*)	4.839 (*A*)
G5	*Brassica rapa*	0.181 (*E*)	3.808 (*A*)	0.961 (*E*)	2.111 (*A*)
G6	*Brassica rapa*	0.047 (*E*)	1.784 (*A*)	0.598 (*E*)	0.354 (*E*)
G7	*Brassica rapa*	0.125 (*E*)	0.843 (*E*)	0.250 (*E*)	0.514 (*E*)
G8	*Brassica rapa*	0.001 (*E*)	2.828 (*A*)	0.012 (*E*)	5.499 (*A*)

A- Accumulator: *E*- Excluder: Brassica J- Juncea (Mustard Greens/Mpilu): Brassica R- Rapa (Chinese cabbage): Cucurbita M- Moschata (Pumpkin Leaves): Brassica-N (Rape)

The bioaccumulation factors (BF) of Cadmium (Cd), Copper (Cu), lead (Pb) and Zinc (Zn), for the vegetable samples are given in Table 11. Out of the total 24 samples of vegetables, 20 vegetables were found to be accumulators of Cu, while four vegetable samples were excluders of Cu. 14 vegetable samples out of 24 samples were found to be accumulators of Zn, however 10 were found to be excluders of Zn. Pb was found to accumulate in four vegetables and non -accumulative in 20vegetables. Out of the 24vegetables Cd was found to accumulate in one vegetable sample and excluder in 23vegetable samples. The accumulation trend in the vegetables were in the order of *Brassica napus*>*Brassica rapa*>*Cucurbita moschata*<*Brassica juncea*.

5.5 Human Health Risk Assessment

5.5.1 Chronic Daily Heavy Metal Intake and Hazard Quotients

Table 12: Chronic daily intake (CDI) of heavy metals and hazard quotients in vegetables for Male, Female and Children in Chunga

Heavy Metal	CDI Male	CDI Female	CDI Children	RfD	HQ
Cadmium	0.004	6.47	2.63	(0.005)	0.244 **≤ 1**
Copper	19.53	21.3	85.44	(0.04)	23.8 ≥ 1
Lead	13.55	14.57	59.22	(0.0005)	17.46 ≥ 1
Zinc	223.23	240.40	976.63	(0.03)	25.22 ≥ 1

HQ- Hazard Quotient: ≥ 1- Greater than one: ≤ 1-Less than one

The chronic daily intake mean values of heavy metals in male and female adults as well as children both male and female are seen in the table above. CDI for Cadmium in the adult male was found below the reference dose at 0.004mg/kg, and above the reference dose in female at a value of 6.47mg/kg and 2.63mg/kg in children. Copper values were found between 19.53mg/kg for male, 21.3mg/kg for females and 85.44mg/kg for children. Lead values were 13.55mg/kg for male, 14.57mg/kg for female and 59.22mg/kg for children while Zinc was highly detected beyond the reference dose at a value of 223.22mg/kg for male, 240.40mg/kg for females and 976.63mg/kg for children. Chronic daily intake of heavy metals followed the order *Zn<Cu<Pb<Cd.*

The hazard quotients for adult male and adult female were calculated as seen in table 10. Cadmium was ≤ 1 for both adult male and female as well as children. Copper, Lead and Zinc were ≥ 1 for children. Copper, Lead and Zinc were found to be ≥ 1 in both adult males and females.

5.5.2 Carcinogenic Risk Assessment

Table 13: Cancer risk (CR) due to exposure to Cadmium and Lead in Vegetables.

Heavy Metal	Male(Adult)	Female (Adult/Pregnant)	Children(M/F)
Cadmium	0.228×10^{-3}	2.45×10^{-5}	9.9×10^{-5}
Lead	11.5×10^{-5}	18.2×10^{-5}	50.3×10^{-6}

Table 13 shows that the risk of developing cancer in adult female and children is 2.45 x 10-5 and 9.9 x 10-5 for Cadmium and 0.228 x 10-3. Lead was found to be 11.5 × 10^{-5} for male adult, 18.2 × 10^{-5} female and 50.3 × 10^{-6} for children.

CHAPTER FIVE

DISCUSSION OF FINDINGS

The study revealed that wastewater, soil and vegetable samples analysed from the gardens around the Chunga treatment plant were contaminated with Cadmium (Cd), Copper (Cu), Lead (Pb) and Zinc (Zn) concentrations. The levels of Zn, Cr, Pb and Cd were above the permissible levels of heavy metals in wastewater, soil and vegetables as per WHO Standards. Based on the observed bio-accumulation factors, *Brassica* napus showed more hyper-accumulation potential as compared to the other vegetable samples. The vegetables were found to pose both carcinogenic and non-carcinogenic adverse health effects to male and female adults as well as children.

6.1 Concentrations of Heavy Metals

6.1.1 Cadmium

Cadmium was found at a lower concentration in the vegetables than in the soil and waste water. Cadmium concentrations were found to be 21% in vegetables, 100% in soil and 44% in water not compliant with WHO permissible levels. High levels of cadmium were detected in the reference group in the fresh water. Cadmium bioaccumulation factors in *Brassica rapa, Brassica napus, Curcubita moschata* and *Brassica juncea* were not significantly different in the vegetables.

A study conducted by Rajankar *et al.* (2014), in India showed that cadmium concentrations are usually lower in storage tissues (fruit, roots, and seeds). The ratio of cadmium in roots to cadmium in leaves was determined to be 1:5. Varol & Senb (2012), in their study conducted in Turkey reported that cadmium accumulates mainly in the vegetable leaves as compared to soil and water. Our study opposed the findings of Varol & Senb as we found cadmium concentrations were of negligible concentrations in the vegetable leaves. A study by Kribek *et al*. (2010), in Zambia observed that the range of cadmium in different media varies from soil, water and crop media, ranging from 0.07 to 0.83 with an average of 0.23. For our study Cadmium was found to be highly accumulated in soil as, compared to other heavy metals, as it is assumed to be generally more bioavailable in soil. In Rwanda and Pakistani elevated levels of cadmium in vegetables and crops cultivated in metal-contaminated soils were found (Nhapia et al., 2012; Muhammad et al., 2011). Fascin, (2013) in sub-Saharan Africa reported that the different mineralogy form and the presence of bioleaching microbes could explain the various phyto- and bioavailability of cadmium in plants which could pose health risks.

Despite cadmium concentration being high in the soil, in the vegetables it was under the limit of detection. In contrast with our results, a study by Eruola *et al*. (2011), in Nigeria found elevated levels of cadmium in vegetables that pose health risks. However, the results in our study showed that cadmium was of low potential health risk among the studied heavy metals.

It was noted for our study that low bioaccumulation of Cd in vegetables is due to the fact that hyper accumulation is often metal-selective, and possible diffusion limitations at the soil level reduce the overall suitability of certain vegetables accumulating certain metals with Cd been a probable one for our study.

6.1.2 Copper

The mean concentrations of Copper in the vegetables and soil in this study exceeded the WHO set standards for copper in the different media. However wastewater had low levels of Cu detected and was found to be within the WHO set standards for wastewater, yet Cu in the control water was beyond the permissible WHO standards. Cu was found to be highly accumulated in the vegetables, with *Brassica napus* been the most hyper accumulator of Cu. Non-carcinogenic adverse health effects were established in Cu for the study.

Zhang *et al* (2010), in chin, studied the response of three vegetables to Cu toxicity and found that Cu levels in both root and shoot increased, but root Cu concentration increased more sharply than shoot with increasing Cu levels in growth media. Elinge *et al*. (2011) in their study conducted in kebbi state reported that Cu mainly accumulated in roots while a small fraction (10%) of absorbed Cu was transported to shoot and leaves. Li *et al.* (2010), found that Cu concentration in the shoots was significantly influenced by Cu concentration in soil and increased markedly with an increase in the soil Cu concentration. Mahar & Jahangir (2013), in Pakistan found that of four agronomic species tested, *Brassica rapa* exhibited the highest affinity for accumulating Cu and Pb from the soil, either with or without additional use of mobilizing soil amendments. Sracek *et al.* (2012), in a study in Zambia observed that Cu was a sensitive metal that is easily accumulated by plants as compared to being accumulated in water.

Our study opposed the findings of Zhang *et al*. (2010), as our results showed that copper was highly accumulated in the leafy part of the vegetables and moderately accumulated in the soil, which could be due to geo-graphical spatial movement of the element. For our study this means that Cu levels accumulated more in vegetables as plants are known to be more sensitive to Cu. Low concentrations of Cu in the wastewater can be attributed to the retention duration of cu in water. Comparison of these two studies demonstrates the need to consider the plant species, growth conditions, plant health, plant age, time of exposure, and the growing environment when evaluating Cu accumulation in Vegetables.

6.1.3 Lead

Elevated lead levels beyond WHO permissible limits were detected in a number of water samples and vegetable samples. Lead concentrations in soil were below the set WHO standards. High concentration of lead were found in the water as compared to the vegetables and soil. The bioaccumulation factors of lead in the vegetables did not vary. Out of 24 vegetable samples, only two vegetable samples of *Brassica napus*, one vegetable sample of *Brassica juncea* and *Cucurbita moschata* were found to be hyper-accumulators of lead. For our study lead was found to cause non-carcinogenic and carcinogenic health effects in human beings.

Our results were similar to the results obtained by Ekpo *et al.* (2011) in Nigeria were accumulation of Pb in vegetables and wastewater was high. Cobbina et al., in Ghana (2015) reported high levels of lead above permissible values in the ground and surface water of agricultural sites in residential areas in Ghana. A study by Vaishaly *et al.* (2015), In India analysed the concentrations of Pb in wastewater which ranged from 99.2 to 188 mg/l (mean 133 mg/ l_1) and from 2400 to 16200 mg/l_1 (mean 8870 mg/ l_1). The levels of lead in the wastewater were very high, as opposed to the low solubility of lead that is was reported in normal soils. With the high concentrations of bioavailable Pb present in the wastewater potential hazards to human health were found. A study by Liu *et al.* (2013), in japan determined that copper and zinc have a very short retention time in water, whereas, for cadmium and lead it is longer.

In our study it was observed that Pb accumulation was high in the wastewater as compared to the vegetables and soil, a probable source of Pb being high in wastewater could be attributed to the fact that water functions more or less as a transport media for elements. Therefore it was observed in our study that the retention time may influence the high concentration of Pb in wastewater. This may signal the occurrence of adsorption of Pb in vegetables that are high accumulators of heavy metals. It was observed in our study that Vegetables are likely to accumulate lead in high concentrations as the lead becomes mobile and bioavailable subsequent to changes in soil environmental conditions.

6.1.4 Zinc

The mean concentrations of Zn in the vegetables exceeded the WHO permissible standards in the vegetables. Negligible mean values were found of Zn in the wastewater and soil samples, with a number of samples detected above the WHO standards. The bio-accumulation factor determined for Zn out of the 24vegetables sample, Zn was found to be highly accumulated by

14vegetable samples. A high concentration of Zn was detected in *Brassica rapa*. Zn was found to cause non-carcinogenic health effects in1 human beings.

A study conducted by Bull (2010), in New-York reported that vegetable growing on heavy metal contaminated soils accumulate high concentrations of Zinc to cause serious health risk to consumers, though zinc is considered as an essential element in the human body when accumulated in high levels it becomes toxic. Duda & Blaszcyk *(*2008), in Tanzania reported that heavy metals most often found in vegetables include copper, arsenic, chromium, lead, zinc, cobalt and nickel. When in trace quantities, some of them are micronutrients. However, they can pose a significant health risk to humans, leading to various chronic diseases, particularly in elevated concentrations or in prolonged dietary intakes. In field experiments conducted at two different metal contaminated sites in Belgium and the Netherlands by Momodu (2010), low Zn concentrations in 18 different rape seed accessions ranged between 3.6 to 8.1 mg/kg DW at a total soil Zn concentration of 5.5 mg Cd/kg soil DW for the Belgian site, and between 5.2 and 11.3 mg/kg DW at a total soil Zn concentration of 2.5 mg Cd/kg soil DW for the Dutch site. Sardar *et al.* (2013), reported Cd concentrations in *B. napus* shoots of 3 mg/kg DW after a growing period of 3 weeks in a neutral silty soil with total Cd concentrations of 40 mg/kg. Zinc concentrations in the same plants were about 600 mg/kg. No adverse effects on plant growth were observed at these concentrations.

The findings from other studies were comparable with the findings of our study in which high levels of Zn were found in the edible parts of the vegetables. In the present study, wastewater irrigated vegetable samples were found to contain more Zn concentrations than the fresh water irrigated vegetable samples. Zn was found to accumulate at different concentrations in vegetables irrigated with wastewater and fresh water, thus a difference was observed due to the mobility of Zn. The results of this study showed that bio-accumulation factors (BF) values varied among the vegetable samples in the study for Zn. It was observed that the variations in Zn BF values in the vegetables could plausibly be attributed to the vegetables absorption capacity.

6.2 Comparison across the Different Media

In this study; cadmium, copper, lead and zinc showed a significant difference ($P<0.05$) in the wastewater, soil and vegetable samples. Concentrations of cadmium, copper, lead and zinc differed significantly in the wastewater, soil and vegetables from Chunga gardens. Results of heavy metal ranges were compared with the control group of the vegetables not irrigated with waste water and the vegetables irrigated with fresh water. The comparison showed that there was no difference in heavy metal accumulation with the vegetables irrigated with wastewater.

A study by Francis et al. (2017) revealed significant ($p < 0.05$) heavy metal variation in selected water samples of Chingola township in Zambia. Ikenka et al. (2010), in Zambia reported significant differences in heavy metal concentration with respect to location/media and season of the year. Nhappia et al. (2012), in Rwanda reviewed that heavy metal content in different leafy vegetables as well as wastewater varies significantly. The content varies with time of harvesting and stage of maturity of crops. The significant differences ($P < 0.01$) were observed between the mean metal concentrations in the three vegetables species and wastewater samples.

The source of the heavy metals accumulated in the investigated vegetables was envisaged to be the wastewater used for irrigation. This was conceived to be the source due to the fact that the chunga stream near the Chunga garden's was receiving wastes of different compositions such as batteries, medical waste, industrial waste and waste from the chunga treatment plant, of which are known to be sources of heavy metals and other toxic chemicals (Ettler et al., 2011 & Nyambe et al., 2012). However, the study showed that different sampling locations contributed differently to the mean heavy metal concentrations in the different media. Due to the variations of the heavy metal accumulation in the wastewater, soil and vegetable samples

A plausible reason for the variation in accumulation could be attributed to spatial geographical movement of metals with distance. Thus for our study the researcher assumes that the heavy metals could have been from other sources rather than wastewater such as fertilizers and pesticides (Muzungurie and Mubiyana, 2015).

6.3 Health Risk Assessment

6.4.1 Non-Carcinogenic

Results from Liu et al. (2007) & Liang (2011), revealed that vegetables grown on wastewater-irrigated soils contaminated with heavy metals pose a major health concern. Our study assessed the health risk of the inhabitants of chunga due to heavy metal intake from the vegetable consumption.

The chronic daily Intake of metals (CDI), health risk index (HRI), and target hazard quotient (THQ) were Calculated and the results in our study indicated that the HRI values were >1; therefore, the non-carcinogenic health risks of heavy metal exposure through the food chain was generally assumed to pose a health risk. The estimated dietary intakes of Cd, Cu, Pb, and

Zn were above the tolerable limits. The summed up hazard quotients in adult males, adult females and children were found to be above one, hence a probability of non-carcinogenic effects occurring in both adult males and females with risk greatest to children. The risk sequence for non-carcinogenic health effects for the residents around chunga gardens was laid out as follows: Male>>Female>>Children, The adult male risk was minimal as compared to the adult female.

A study conducted by Francis et al. (2017) in Chingola, Zambia found that HRI and CDI values in drinking water contaminated with heavy metals were above 1, thus the risk of non-carcinogenic risks was more prevalent in children due to arsenic exposure. For our study the hazard quotients for children were higher than the female and male adults. Heavy metal concentration in our study was found to be more in children as they have a lower body mass. Hence the concentration of heavy metals in an adult is twice or thrice in children thus the risk is higher in children regardless of how low the exposure is. A similar study by Khan et al. (2013) reported unacceptable Hazard Quotients in India with highest Cadmium Hazard Quotients of 26.80 in an area of intensive agricultural use posing non-carcinogenic health risks. In this study, Cadmium was below the hazard quotient and was found to pose no adverse health effects in both adult male and female, as well as in children.

The findings of this study regarding the CDI and HRI suggest that the consumption of vegetables irrigated with wastewater contaminated soils are potentially harmful and not free of risk, nevertheless the study acknowledged other sources of metal exposures such as dust inhalation, dermal contact and ingestion (for children) of metal-contaminated soils, which were not included in this study (Badu et al., 2013 and Akoto et al., 2008).

6.4.2 Carcinogenic risks

Vegetable consumption carcinogenic risk due to cadmium was 9.9×10-5 while Cancer risk due to lead was 50.3 ×10-6. This means that the probability of developing cancer for persons who consume the vegetables is 5 persons per 100, 000 and 6 persons per 1,000 for the children. Highest carcinogenic risk was found in lead. Thus according to our study children are more susceptible to developing cancer from ingestion of the vegetables around Chunga gardens, yet the probability for both male and female adults cannot be said to be low as every low dose has a likely hood of causing cancer.

According to Akinola (2006) Lead is causing concern in particular due to the possible impacts on children lead is said to cause cancer of the urinary bladder, skin and lungs, hence lead poses the greatest carcinogenic effect. Due to the additive nature of heavy metals, risk to cancer development in chunga due to heavy metal exposure from consumption of vegetables is likely to be very high.

6.5 LIMITATIONS

1. The source of the heavy metals in the soil can be assumed to be from natural resources such as soil parent material. Confounders were noted at the beginning of the study that could lead to the high concentrations of metals in soil and therefore uptake by vegetables. Fertilizer addition was considered as a probable reason for the high contents of heavy metals in the agricultural soils. Whereas, the other potential source for the heavy metals might have been primarily from long-term usage of pesticides, as the study area is a non-controlled environment, hence there is no control over the addition of fertilisers/pesticides by the individual farmers e.g. in terms of when last applied and how much applied.

2. Information for the risk assessment was limited regarding the toxicity data for cancer slope factors for some of the heavy metals such as Copper and Zinc, furthermore it was noted that the toxicity values are derived from experiments conducted with animals and extrapolated on human organisms. Consequently, there can be varying degrees of uncertainty associated with the toxicity values calculated in the study.

3. No reference sample was used for the soil as the researcher was not given consent to collect the soil samples from the reference garden by its owner.

6.6 STRENGTHS

1. Standardised approaches permit the study to be replicated in different areas or over time with the production of comparable findings.

2. This is the first attempt to quantifying the health risk assessment due to consumption from vegetables irrigated with wastewater in Lusaka District.

3. Confounding was dealt with by inclusion of the control group. The conditions under which the vegetables were grown were assumed to be uniform with variation only being waste water.

CHAPTER SIX

CONCLUSION AND RECOMMENDATIONS

7.1 CONCLUSION

This study revealed that the concentrations of Cadmium, Lead, Copper and Zinc were beyond the World Health Organisation maximum permissible levels in the wastewater, soil and vegetables irrigated with wastewater and without wastewater from the gardens around the Chunga treatment plant. The results from the present study suggest that elemental concentration of both wastewater grown vegetables and fresh water irrigated vegetables was not statistically significant. Elemental concentrations also varied among the four different vegetable types, comparatively, *Brassica napus* (rape) showed highest heavy metal accumulation, with copper being more mobile among the vegetables. Leafy parts of all the vegetable samples depicted more concentration of heavy metals than the soil samples and wastewater samples, Therefore heavy metals which are likely to cause adverse health effects are: cadmium, lead, copper and zinc. According to the health risk assessment conducted in the study high concentrations of heavy metals have a high probability of causing various adverse health effects including non-carcinogenic and carcinogenic health effects. Henceforth, regular monitoring of heavy metals in the vegetables grown in wastewater and freshwater is important to avoid health risks in human beings.

7.2 RECOMMENDATIONS

Ministry of Agriculture

1. These results should be considered in agricultural policy on the basis of regular monitoring and evaluation of vegetables, wastewater and agricultural soil with regards chemical concentrations

Lusaka Water and Sewerage Company and Lusaka City Council

2. Lusaka water and Sewerage Company should improve its treatment methods of wastewater from the Chunga treatment plant as it is discharged into the Chunga stream so as to reduce levels of Cadmium, Copper, Lead and Zinc in the wastewater used during reclamation.
3. Lusaka city council should ensure that awareness programs are implemented and farmers are educated on the consequences of using contaminated wastewater for irrigation purpose.

Further Research

4. Systematic study of the heavy metals in soils and vegetables grown, as well as an assessment of pollution source apportionment is recommended.
5. Preliminary assessment of risk to human health is highly recommended through: identification of the actualised Health risk/attribution/ connecting exposure to actual disease conditions in the population.
6. **Farmers**

   Farmers should inquire with Ministry of Agriculture on safe wastewater irrigation farming systems.

8.0 REFERENCES

Adelekan, b. A. & abegunde, k. D. 2011. Heavy metals contamination of soil and groundwater at automobile mechanic villages in ibadan, nigeria. *International journal of the physical sciences,* vol. 6(5)**,** pp. 1045-1058.

Aderinola, o. J., clarke, o., olarinmoye, o. M., kusemiju, v. & anatekhai, m. A. 2009. Heavy metals in surface water, sediments, fish and perwinkles of lagos lagoon. *American-eurasian j. Agric. & environ. Sci,* 5 (5)**,** 609-617.

Akoto, o., bruce, t. N. & darko, g. 2008. Heavy metals pollution profiles in streams serving the owabi reservoir. *African journal of environmental science and technology, available online at http://www.academicjournals.org/ajest,* vol. 2 (11)**,** pp. 354-359.

Asare-donkor, n. K., boadu,t.a. & adimado,a.a. 2016. Evaluation of groundwater and surface water quality and human risk assessment for trace metals in human settlements around the bosomtwe crater lake in ghana. *Springerplus 5:1812. Doi 10.1186/s40064-016-3462-0*.

Badu, m., wemegah, d. D., boadi, n. O. & brown, f. A. 2013. Assessment of the nutrient load and selected heavy metals in the owabi reservoir and its feeder waters. *Am. J. Sci. Ind. Res,* 4(4)**,** 333-343.

Blacksmithinstitute. 2006. Project completion report: contamination of the kafue river basin. *Available from www.blacksmithinstitute.org, accessed on 6th april, 2016*.

Bull, s. 2010. Copper general information. *Health protection agency, crce hq, hpa, version 1, new york, usa*.

Cobbina, s. J., duwiejuah, a. B., quansah, r., obiri, s. & bakobie, n. 2015. Comparative assessment of heavy metals in drinking water sources in two small-scale mining communities in northern ghana. *Int j environ res public health,* 12**,** 10620-34.

Cso. 2011. Zambia 2010 census of population and housing: preliminary report. *Lusaka, zambia*.

Das, s., patnaik, s. C., sahu, h. K., chakraborty, a., sudarshan, m. & thatoi, h. N. 2013. Heavy metal contamination, physico-chemical and microbial evaluation of water samples collected from chromite mine environment of sukinda, india. *Transactions of nonferrous metals society of china,* 23**,** 484-493.

Duda-chodak, a. B., blaszczyk, u. 2008. The impact of nickel on human health. *J. Elementol, 13(4): 685-696.*

Ekpo, i. A., agbor, r. B., ekanem, a. P., okpako, e. C. & inyang, p. I. 2012. Assessment of heavy metal status and bacteria population in municipal borehole water in landfill and non-landfill areas in calabar municipality, cross river state, nigeria. *International journal of engineering and applied sciences,* vol. 2.

Elinge, c. M., itodo, a. U., i.j., p., u.a., b. Y. & mbongo a, n. 2011. Assessment of heavy metals concentrations in bore-hole waters in aliero community of kebbi state. *Advances in applied science research,* 2 (4)**,** 279-282.

Eruola, a. O., ufoegbune, g. C., eruola, a. O., awomeso, j. A. & abhulimen, s. A. 2011. Assessment of cadmium, lead and iron in hand dug wells of ilaro and aiyetoro, ogun state, south-western nigeria. *Research journal of chemical sciences,* vol. 1(9)**,** 1-5.

Ettler, v., mihaljevič, m., kříbek, b., majer, v. & šebek, o. 2011. Tracing the spatial distribution and mobility of metal/metalloid contaminants in oxisols in the vicinity of the nkana copper smelter, copperbelt province, zambia. *Geoderma,* 164**,** 73-84.

Farooq, m. A., shaukat, s. S., zafar, m. U. & abbas, q. 2012. Variation pattern of heavy metal concentrations during pre- and post-monsoon seasons in the surface water of river indus (sindh province). *World applied sciences journal,* 19 (4)**,** 582-587.

Fasinu, p. S. & orisakwe, o. E. 2013. Heavy metal pollution in sub-saharan africa and possible implications in cancer epidemiology. *Asian pacific journal of cancer prevention,* 14**,** 3393-3402.

Francis hamwiinga., patricia mubita., & chisala, d. Meki. 2017. Heavy metal pollution levels in drinking water sources and and their potential human health risks in chingola district zambia. *Lusaka,* zambia.

Gutiérrez, r., rubio-arias, h., quintana, r., ortega, j. A. & gutierrez, m. 2008. Heavy metals in water of the san pedro river in chihuahua, mexico and its potential health risk. *Int. J. Environ. Res. Public health,* 5(2)**,** 91-98.

Ikenaka, y., nakayama, s., muzandu, k., choongo, k., teraoka, h., mizuno, n. & ishizuka, m. 2010. Heavy metal contamination of soil and sediment in zambia. *African journal of environmental science and technology,* vol. 4(11)**,** pp. 729-739.

Itodo, a. U., elinge, c. M., peni i.j., birnin – yauri u.a. &mbongo a,n. 2011. Assessment of heavy metals concentrations in bore-hole waters in aliero community of kebbi state. *Advances in applied science research, 2 (4):279-282.*

Kavcar, p., sofuoglu, a. & sofuoglu, s. C. 2009. A health risk assessment for exposure to trace metals via drinking water ingestion pathway. *Int j hyg environ health,* 212**,** 216-27.

Khan, s., shahnaz, m., jehan, n., rehman, s., shah, m. T. & din, i. 2013. Drinking water quality and human health risk in charsadda district, pakistan. *Journal of cleaner production,* 60**,** 93-101.

Koki, i. B., bayero, a,s.,umar,a. &yusuf,s. 2015. Health risk assessment of heavy metals in water, air, soil and fish. *Afr. J. Pure appl. Chem, vol. 9(11), pp. 204-210.*

Křibek, b., majer, v., veselovský, f. & nyambe, i. 2010. Discrimination of lithogenic and anthropogenic sources of metals and sulphur in soils of the central-northern part of the zambian copperbelt mining district: a topsoil vs. Subsurface soil concept. *Journal of geochemical exploration,* 104**,** 69-86.

Liang, f., yang, s.g., sun, c. 2011. Primary health risk analysis of metals in surface water of taihu lake, china. *Bull environ contam toxicol 87(4):404–408.*

Li, s. & zhang, q. 2010. Spatial characterization of dissolved trace elements and heavy metals in the upper han river (china) using multivariate statistical techniques. *J hazard mater,* 176**,** 579-88.

Liu, c. W., liang,c,p, lin,k.h., jang,c.s., wang,s.w, huang,y. &yu-mei hsueh .2007. Bioaccumulation of arsenic compounds in aquacultural clams (meretrix lusoria) and assessment of potential carcinogenic risks to human health by ingestion. *Chemosphere, doi:10.1016/j.chemosphere.2007.04.038*.

Liu, x., song,q., tang,y., wanlu li,w., xu,j., wu,j., wang,f.,brookes,p,c. 2013. Human health risk assessment of heavy metals in soil–vegetable system: a multi-medium analysis. *Science of the total environment 463–464, 530–540*.

Mahar, m. T., khuhawar,m.y.,m.a., & jahangir, m. 2013. Health risk assessment of heavy metals in groundwater, the effect of evaporation ponds of distillery spent wash: a case study of southern punjab pakistan. *World applied sciences journal 28 (11): 1748-1756*.

Mazumder.d.n.g. 2008. Chronic arsenic toxicity & human health. *Indian j med res 128, pp 436-447*.

Who. 2008. Guidelines for drinking-water quality. Second addendum to third edition. *Geneva, switzerland*.

Momodu, m. A. & anyakora, c. A. 2010. Heavy metal contamination of ground water: the surulere case study. *Res. J. Environ. Earth sci,* 2(1)**,** 39-43.

Mora, a., mac-quhae, c., calzadilla, m. & sanchez, l. 2009. Survey of trace metals in drinking water supplied to rural populations in the eastern llanos of venezuela. *J environ manage,* 90**,** 752-9.

Morais, m., garcia e costa, f. & de lourdes pereira, m. 2012. Heavy metals and human health. *Environmental health – emerging issues and practice,* www.intechopen.com.

Muhammad, s., shah, m. & khan, s. 2011. Health risk assessment of heavy metals and their source apportionment in drinking water of kohistan region, northern pakistan. *Microchemical journal 98 (2011) 334–343*.

Muzungaire, l. & mubiana, v. K. 2015. Preliminary environmental assessment of metal contamination from mining areas: case of the kafue river, zambia, proceedings of the 14th international conference on environmental science and technology rhodes, greece, 3-5 september 2015.

Nachiyunde, k., ikeda, h., okuda, t. & nishijima, w. 2013. Assessment of dissolved heavy metal pollution in five provinces of zambia. *Journal of environmental protection,* 04**,** 80-85.

Nadeem-ul-haq., arain, m. A., haque, z., badar, n. & mughal, n. 2009. Drinking water contamination by chromium and lead in industrial lands of karachi. *J pak med assoc,* vol. 59.

Naveedullah., h. Z., yu,c., shen,h., duan,d., shen,c., lou,l. & chen,y. 2014. Concentrations and human health risk assessment of selected heavy metals in surface water of the siling reservoir watershed in zhejiang province, china. *Pol. J. Environ. Stud. Vol. 23, no. 3 (2014), 801-811*.

Nazir, r., khan, m., masab, m., rehman, h., rauf, n., shahab, s., ameer, n., sajeb, m., ullah, m., rafeeq, m. & shaheen, z. 2015. Accumulation of heavy metals (ni, cu, cd, cr, pb, zn, fe) in the soil, water and plants and analysis of physico-chemical parameters of soil and water collected from tanda dam kohat. *J. Pharm. Sci. & res,* vol. 7(3)**,** 89-97.

Nhapia, i., walib, u. G., usanzinezab, d., kashaigilic, j. J., banaddad, n. & kimwaga, r. 2012. Distribution of heavy metals in lake muhazi, rwanda. *The open environmental engineering journal,* 5**,** 96-102.

Obiri, s., mattah, p. A., mattah, m. M., armah, f. A., osae, s., adu-kumi, s. & yeboah, p. O. 2016. Assessing the environmental and socio-economic impacts of artisanal gold mining on the livelihoods of communities in the tarkwa nsuaem municipality in ghana. *Int j environ res public health,* 13.

Obodai, e. A., boamponsem, l. K., adokoh, c. K., essumang, d. K., villawoe, b. O., aheto, d. W. & debrah, j. S. 2011. Concentrations of heavy metals in two ghanaian lagoons. *Arch. Appl. Sci. Res,* 3 (3)**,** 177-187.

Ojekunle, o. Z., ojekunle,o.v., adeyemi,a.a., taiwo,a.g., sangowusi, o.r., taiwo,a.d., adekitan,a.a. 2016. Evaluation of surface water quality indices and ecological risk assessment for heavy metals in scrap yard neighbourhood. *Springerplus 5:560. Doi 10.1186/s40064-016-2158-9.*

Paulinus, n. 2015. Assessment of contamination of underground water sources in enyigba pb-zn district, south eastern nigeria using metal enrichment and pollution indices. *International journal of health sciences, vol 4, september 2015 (09).*

Qaiyum, m. S., m. Shaharudin,s., syazwana.i., a. & muhaimin,a. 2011. Health risk assessment after exposure to aluminium in drinking water between two different villages. *Journal of water resource and protection, 3, 268-274.*

Rajagamapathy, v., xavier, f., sreekumar, d. & mandal, p. K. 2011. Heavy metal contamination in soil, water and fodder and their presence in livestock and products: a review. *Journal of environmental science and technology,* 4 (3)**,** 234-249.

Rajankar, p., mohapatra,p.&dubey, a. 2014. A report on toxicity load of yamuna river in delhi. *Toxics link, new delhi, india, available at http://www.toxicslink.org.*

Sardar, k., ali, s., hameed, s., afzal, s., fatima, s., shakoor, m. B., bharwana, s. A. & tauqeer, h. M. 2013. Heavy metals contamination and what are the impacts on living organisms. *Greener journal of environmental management and public safety,* vol. 2 (4)**,** pp. 172-179.

Simutanyi, n. 2008. Copper mining in zambia, the developmental legacy of privatization. *Institute for security studies.*

Shah, m. T., ara,j., muhammad,s., khan,s.,tariq,s. 2012. Health risk assessment via surface water and sub-surface water consumption in the mafic and ultramafic terrain, mohmand agency, northern pakistan. *Journal of geochemical exploration 118, 60–67.*

Sracek, o., kříbek, b., mihaljevič, m., majer, v., veselovský, f., vencelides, z. & nyambe, i. 2012. Mining-related contamination of surface water and sediments of the kafue river drainage system in the copperbelt district, zambia: an example of a high neutralization capacity system. *Journal of geochemical exploration,* 112**,** 174-188.

Tchounwou, p. B., yedjou, c. G., patlolla, a. K. & sutton, d. J. 2012. Heavy metal toxicity and the environment. *Exs,* 101**,** 133-64.

Tembo, b. D., sichilongo, k. & cernak, j. 2006. Distribution of copper, lead, cadmium and zinc concentrations in soils around kabwe town in zambia. *Chemosphere,* 63**,** 497-501.

Unicef/who. 2015. Progress on sanitation and drinking water: 2015 update and mdg assessment. Geneva, switzerland.

Us epa. 1992. Guidelines for exposure assessment, risk assessment forum; 1992 [epa/600/z-92/001].

United states environmental protection agency (us epa). 1997. Exposure factor handbook, ncea-w-005. 1995.107. Usepa: exposure factors handbook, vol. I: general factors. Office of research and development, washington, dc, usa.

Us epa. 2004. Risk assessment guidance for superfund. Volume i*: human health evaluation manual — part e, supplemental guidance for dermal risk assessment.* washington, dc:office of superfund remediation and technology innovation; 2004 [epa/540/r/99/005. Oswer 9285. 7-02ep pb99-963312].

Us epa. 2011. Exposure factors handbook 2011 edition (final); 2011. Http://cfpub.epa.gov/ ncea/risk/recordisplay.cfm?deid=236252.

Usepa. 2012. Waste and clean-up risk assessment.http://www2.epa.gov/risk/waste-andcleanup- risk-assessment

Vaishaly, a. G., mathew, b. B. & krishnamurthy, n. B. 2015. Health effects caused by metal contaminated ground water. *International journal of advances in scientific research,* 1(02)**,** 60-64.

Varol, m. & şen, b. 2012. Assessment of nutrient and heavy metal contamination in surface water and sediments of the upper tigris river, turkey. *Catena,* 92**,** 1-10.

Vieira, c., morais, s., ramos, s., delenie-matos, c. & oliverira, m. 2011. mercury, cadmium, lead and arsenic levels in three pelagic fish species from the atlantic ocean: intra and inter- specific variability and human health risks for consumption. *Food chem toxicol 49(4):923–932*

Violante, a., caporale, a.g. & pigna.m. 2010. Mobility and bioavailability of heavy metals and metalloids in soil environments. *J. Soil. Sci. Plant nutr. 10 (3): 268 - 292.*

Von der heyden, c. J. & new, m. G. 2004. Sediment chemistry: a history of mine contaminant remediation and an assessment of processes and pollution potential. *Journal of geochemical exploration,* 82**,** 35-57.

Who (world health organization). 2008. Guidelines for drinking-water quality. Second addendum to third edition. *Geneva, switzerland*.

Who. 2011. Guidelines for drinking- water quality (4th ed.). http://www.whqlibdoc.who.int/publications/2011/9789241548151_eng.pdf. accessed 09/10/ 2016.

Wongsasuluk, p. C., s. 2013. Heavy metal contamination and human health risk assessment in drinking water from shallow groundwater wells in an agricultural area in ubon ratchathani province, thailand. *Environmental geochemistry and health. Doi: 10.1007/s10653-013-9537-8.*

Wu, b., zhao, d.y., jia, h.y., zhang, y., zhang, x.x., cheng, s.p. 2009. preliminary risk assessment of trace metal pollution in surface water from yangtze river in nanjing section, *china. Bull environ contam toxicol* 82(4):405–409

Yabe, j., ishizuka, m. & umemura1 umemura, t. 2010. Current levels of heavy metal pollution in africa. *J. Vet. Med. Sci*, 72(10)**,** 1257–1263.

Yu, f.c., fang, g.h., ru, x.w. 2010. Eutrophication, health risk assessment and spatial analysis of wa quality in gucheng lake, china. *Environ earth sci 59(8):17411748*

Zhang, q. & li, s. 2010. Spatial characterization of dissolved trace elements and heavy metals in the upper han river (china) using multivariate statistical techniques. *Journal of hazardous materials 176, 57.*

9.0 APPENDICES

9.1 APPENDIX I: INFORMATION SHEET

THE UNIVERSITY OF ZAMBIA

SCHOOL OF PUBLIC HEALTH

CONSENT FORM TO PARTICIPATE IN RESEARCH

Investigator

I Nyirongo Kunda, a student at the University of Zambia is requesting for your participation in the study mentioned below in partial fulfilment of the requirement for the degree of Master of Public Health in Environmental Health.

Title of study

Assessment of Heavy Metals in Wastewater, Soil and Vegetables grown around Chunga Gardens in Lusaka and their Potential Health Risks.

Introduction

- Before you decide whether or not to participate in the study, I would like to explain to you the purpose of the study and what is expected of you.

- Your participation in this study is entirely voluntary and you are, therefore, under no obligation to participate. If you agree to participate, you will be asked to sign the informed consent form.

- Agreement to participate will not result in any immediate benefit.

Purpose

- The purpose of the study is to assess heavy metals accumulation in wastewater, soil and vegetables around Chunga gardens in Lusaka and their potential health risks .
- The information obtained will highlight the recent levels of heavy metal contaminant uptake in the vegetables consumed by the peri urban population.

- The information from this study can be used by the relevant authorities to develop appropriate measures for management of heavy metal contamination in wastewater in order to ensure food safety for the peri-urban in Zambia.

Description of the study Procedure

- The study will involve sampling of the soil and vegetables around chunga gardens. The parameters will be taken to the Laboratory at the University of Zambia school of Engineering for analysis.

Benefits

- The benefits of the research using the vegetables will help in analysing the characteristics of heavy metals and identify their potential implications on health.

Risks

- There are no foreseeable risks to you as your personal information will not be collected.

Confidentiality

- We will not be collecting or retaining any information about your identity. We will not include any information in any report we may publish that would make it possible to identify you.

Thank you for your willingness to contribute to the success of this research. For any clarification feel free to contact me or University of Zambia Research Ethics Committee (UNZAREC).

Contact details for the principle investigator
Nyirongo Kunda
The University of Zambia
Department of Public Health
Cell: 0978087286

The Chairperson UNZABREC
Ridgeway campus, P.O. Box 50110
Lusaka, Zambia.
unzarec@zamtel.zm
Telephone: 260-1-256067

9.2 APPENDIX II: INFORMED CONSENT

Please read each sentence below and think about your choice. After reading each sentence, write "Yes" or "No". **No matter what you decide to do, it will not affect your confidentiality.**

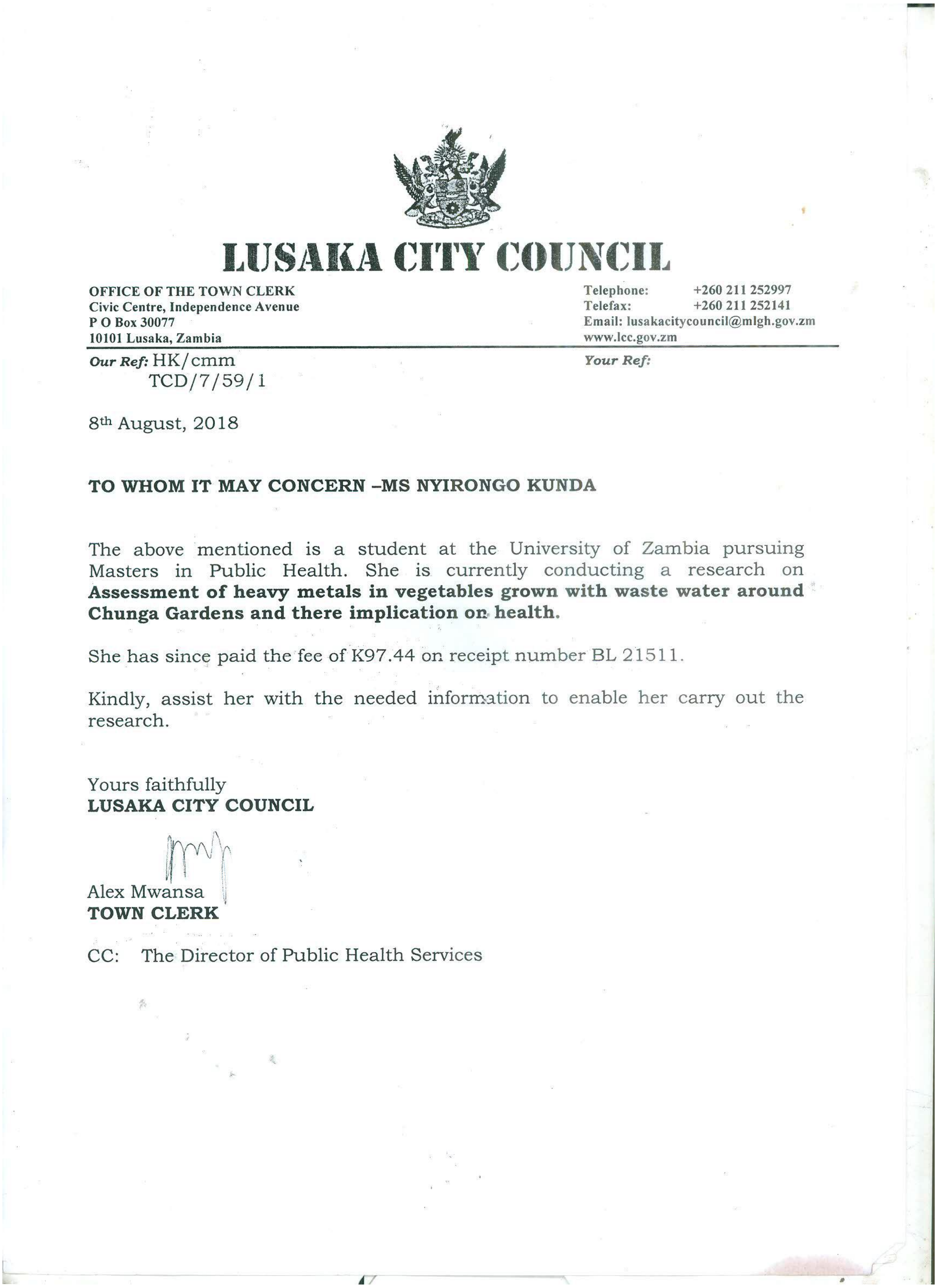
1. I confirm that I have read and understand the information for the above study. I have had the opportunity to consider the information, ask questions and have had these answered satisfactorily. ☐
2. I understand that my participation is voluntary and that I am free to withdraw at any time without giving any reason, without my legal rights being affected. ☐
3. I understand that the relevant data collected during the study, may be looked at by individuals from the University of Zambia, where it is relevant to my taking part in this research. I give permission for these individuals to have access to my records. ☐
4. I agree to take part in the above study. ☐

____________________	____________________
Date	Signature
____________________	____________________
Date	Signature of person taking consent

☐

Thumb print for participant.

9.3 APPENDIX III: PERMISSION LETTER FROM LUSAKA CITY COUNCIL



LUSAKA CITY COUNCIL

OFFICE OF THE TOWN CLERK
Civic Centre, Independence Avenue
P O Box 30077
10101 Lusaka, Zambia

Telephone: +260 211 252997
Telefax: +260 211 252141
Email: lusakacitycouncil@mlgh.gov.zm
www.lcc.gov.zm

Our Ref: HK/cmm
TCD/7/59/1

Your Ref:

8th August, 2018

TO WHOM IT MAY CONCERN –MS NYIRONGO KUNDA

The above mentioned is a student at the University of Zambia pursuing Masters in Public Health. She is currently conducting a research on **Assessment of heavy metals in vegetables grown with waste water around Chunga Gardens and there implication on health.**

She has since paid the fee of K97.44 on receipt number BL 21511.

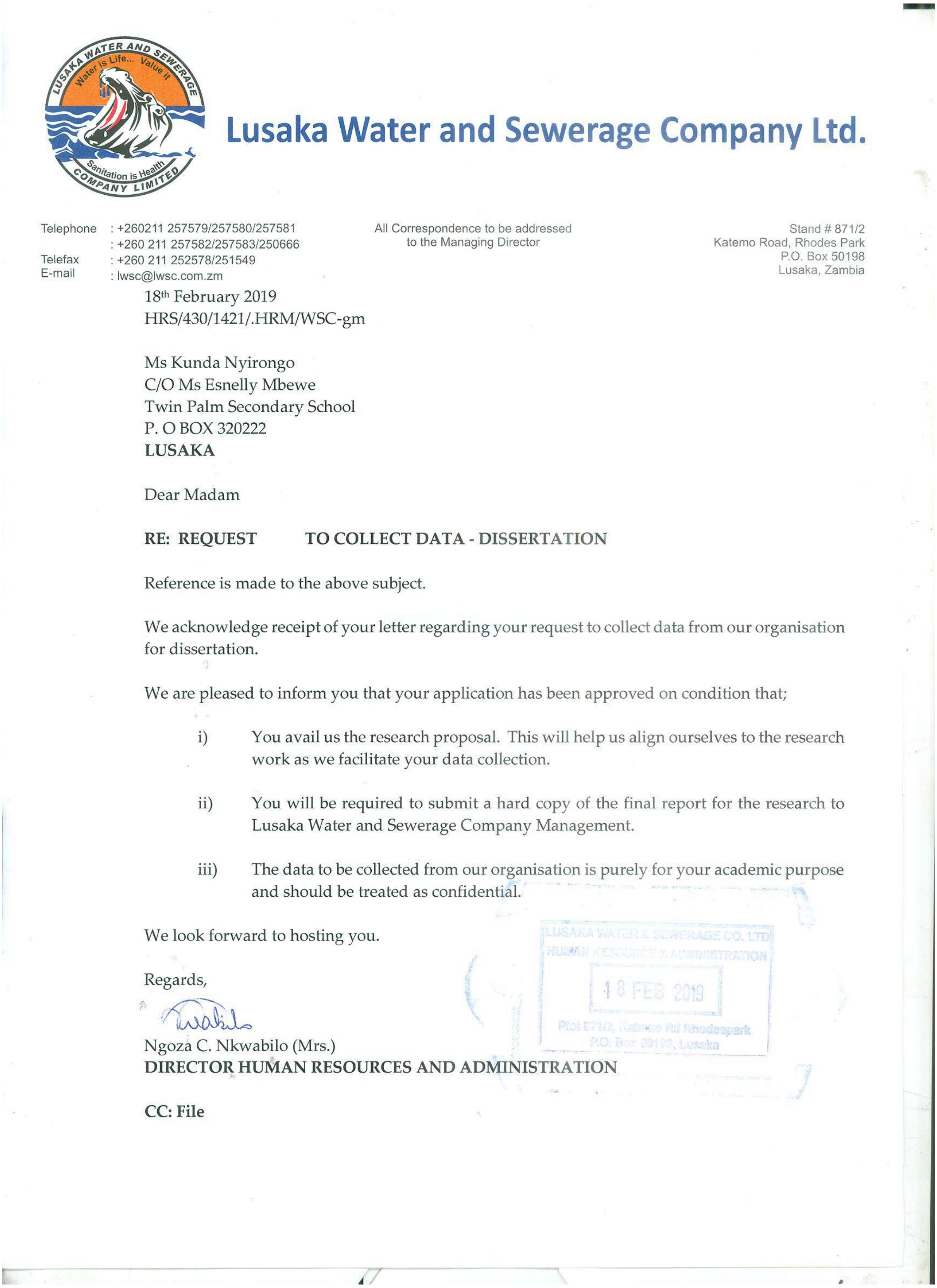
Kindly, assist her with the needed information to enable her carry out the research.

Yours faithfully
LUSAKA CITY COUNCIL

Alex Mwansa
TOWN CLERK

CC: The Director of Public Health Services

9.4 APPENDIX IV: PERMISSION LETTER FROM LUSAKA WATER AND SEWERAGE COMPANY



Lusaka Water and Sewerage Company Ltd.

Telephone : +260211 257579/257580/257581
: +260 211 257582/257583/250666
Telefax : +260 211 252578/251549
E-mail : lwsc@lwsc.com.zm

All Correspondence to be addressed
to the Managing Director

Stand # 871/2
Katemo Road, Rhodes Park
P.O. Box 50198
Lusaka, Zambia

18th February 2019
HRS/430/1421/.HRM/WSC-gm

Ms Kunda Nyirongo
C/O Ms Esnelly Mbewe
Twin Palm Secondary School
P. O BOX 320222
LUSAKA

Dear Madam

RE: REQUEST TO COLLECT DATA - DISSERTATION

Reference is made to the above subject.

We acknowledge receipt of your letter regarding your request to collect data from our organisation for dissertation.

We are pleased to inform you that your application has been approved on condition that;

i) You avail us the research proposal. This will help us align ourselves to the research work as we facilitate your data collection.

ii) You will be required to submit a hard copy of the final report for the research to Lusaka Water and Sewerage Company Management.

iii) The data to be collected from our organisation is purely for your academic purpose and should be treated as confidential.

We look forward to hosting you.

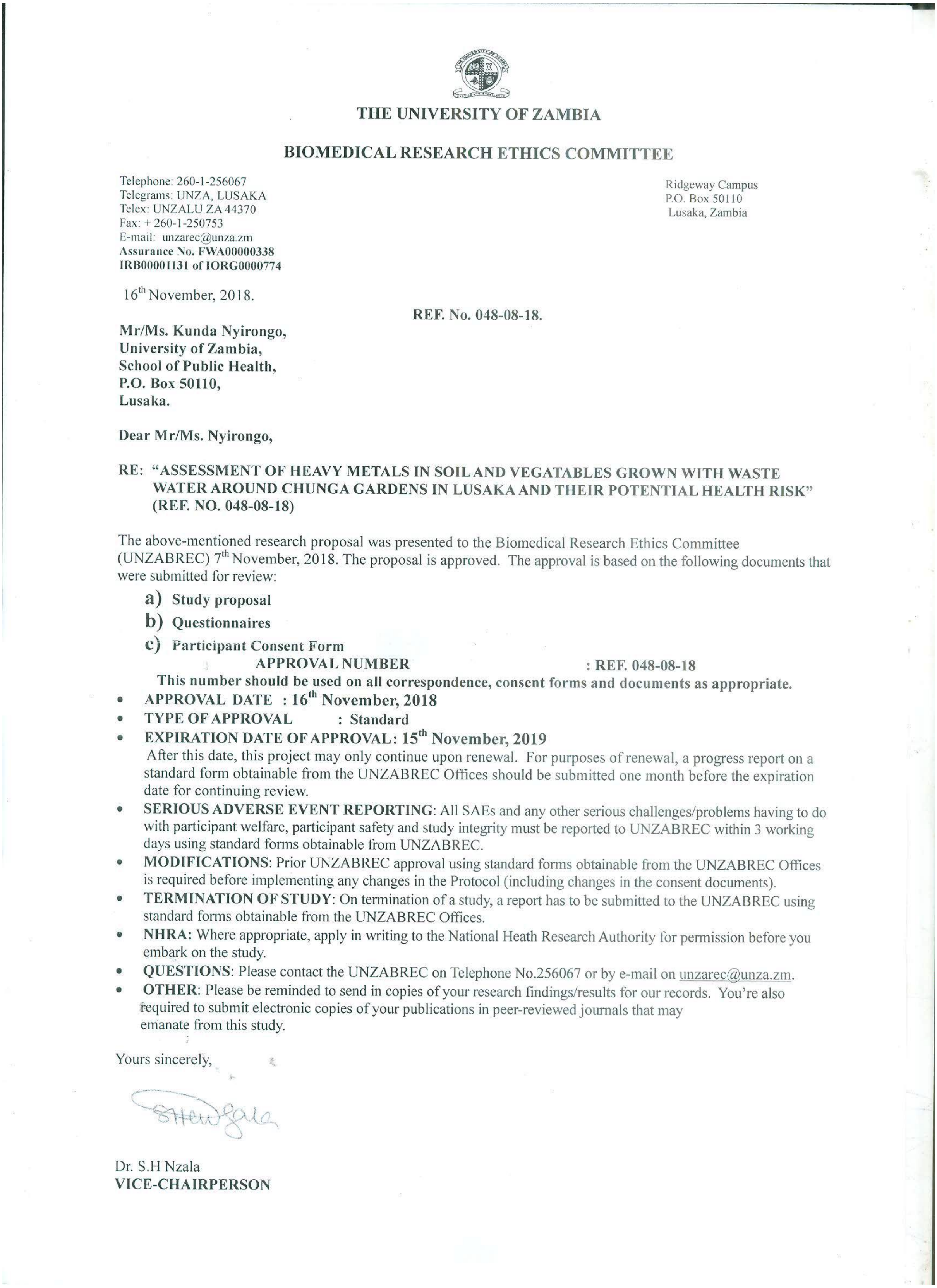
Regards,

Ngoza C. Nkwabilo (Mrs.)
DIRECTOR HUMAN RESOURCES AND ADMINISTRATION

LUSAKA WATER & SEWERAGE CO. LTD
HUMAN RESOURCES & ADMINISTRATION
18 FEB 2019
Plot 871/2, Katemo Rd Rhodespark
P.O. Box 50198, Lusaka

CC: File

9.5 APPENDIX V: APPROVAL LETTER FROM BIOMEDICA RESEARCH ETHICS COMMITTEE



THE UNIVERSITY OF ZAMBIA

BIOMEDICAL RESEARCH ETHICS COMMITTEE

Telephone: 260-1-256067
Telegrams: UNZA, LUSAKA
Telex: UNZALU ZA 44370
Fax: + 260-1-250753
E-mail: unzarec@unza.zm
Assurance No. FWA00000338
IRB00001131 of IORG0000774

Ridgeway Campus
P.O. Box 50110
Lusaka, Zambia

16th November, 2018.

REF. No. 048-08-18.

Mr/Ms. Kunda Nyirongo,
University of Zambia,
School of Public Health,
P.O. Box 50110,
Lusaka.

Dear Mr/Ms. Nyirongo,

RE: "ASSESSMENT OF HEAVY METALS IN SOIL AND VEGATABLES GROWN WITH WASTE WATER AROUND CHUNGA GARDENS IN LUSAKA AND THEIR POTENTIAL HEALTH RISK" (REF. NO. 048-08-18)

The above-mentioned research proposal was presented to the Biomedical Research Ethics Committee (UNZABREC) 7th November, 2018. The proposal is approved. The approval is based on the following documents that were submitted for review:

a) **Study proposal**
b) **Questionnaires**
c) **Participant Consent Form**

APPROVAL NUMBER : REF. 048-08-18

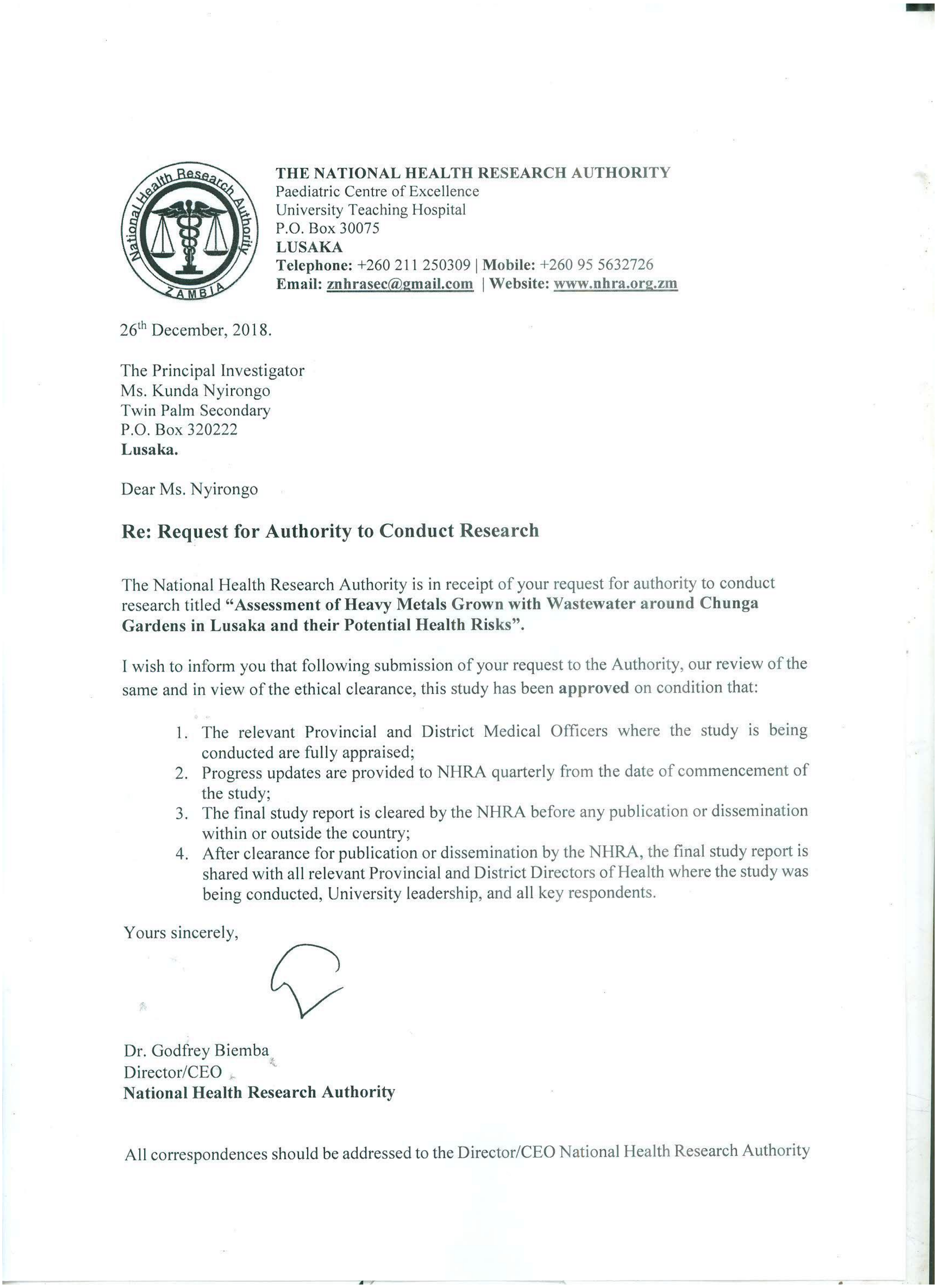
This number should be used on all correspondence, consent forms and documents as appropriate.

- **APPROVAL DATE : 16th November, 2018**
- **TYPE OF APPROVAL : Standard**
- **EXPIRATION DATE OF APPROVAL: 15th November, 2019**
  After this date, this project may only continue upon renewal. For purposes of renewal, a progress report on a standard form obtainable from the UNZABREC Offices should be submitted one month before the expiration date for continuing review.
- **SERIOUS ADVERSE EVENT REPORTING**: All SAEs and any other serious challenges/problems having to do with participant welfare, participant safety and study integrity must be reported to UNZABREC within 3 working days using standard forms obtainable from UNZABREC.
- **MODIFICATIONS**: Prior UNZABREC approval using standard forms obtainable from the UNZABREC Offices is required before implementing any changes in the Protocol (including changes in the consent documents).
- **TERMINATION OF STUDY**: On termination of a study, a report has to be submitted to the UNZABREC using standard forms obtainable from the UNZABREC Offices.
- **NHRA:** Where appropriate, apply in writing to the National Heath Research Authority for permission before you embark on the study.
- **QUESTIONS**: Please contact the UNZABREC on Telephone No.256067 or by e-mail on unzarec@unza.zm.
- **OTHER**: Please be reminded to send in copies of your research findings/results for our records. You're also required to submit electronic copies of your publications in peer-reviewed journals that may emanate from this study.

Yours sincerely,

Dr. S.H Nzala
VICE-CHAIRPERSON

9.6 APPENDIX VI: APPROVAL LETTER FROM NATIONAL HEALTH RESEARCH AUTHORITY



THE NATIONAL HEALTH RESEARCH AUTHORITY
Paediatric Centre of Excellence
University Teaching Hospital
P.O. Box 30075
LUSAKA
Telephone: +260 211 250309 | **Mobile:** +260 95 5632726
Email: znhrasec@gmail.com | **Website:** www.nhra.org.zm

26th December, 2018.

The Principal Investigator
Ms. Kunda Nyirongo
Twin Palm Secondary
P.O. Box 320222
Lusaka.

Dear Ms. Nyirongo

Re: Request for Authority to Conduct Research

The National Health Research Authority is in receipt of your request for authority to conduct research titled **"Assessment of Heavy Metals Grown with Wastewater around Chunga Gardens in Lusaka and their Potential Health Risks".**

I wish to inform you that following submission of your request to the Authority, our review of the same and in view of the ethical clearance, this study has been **approved** on condition that:

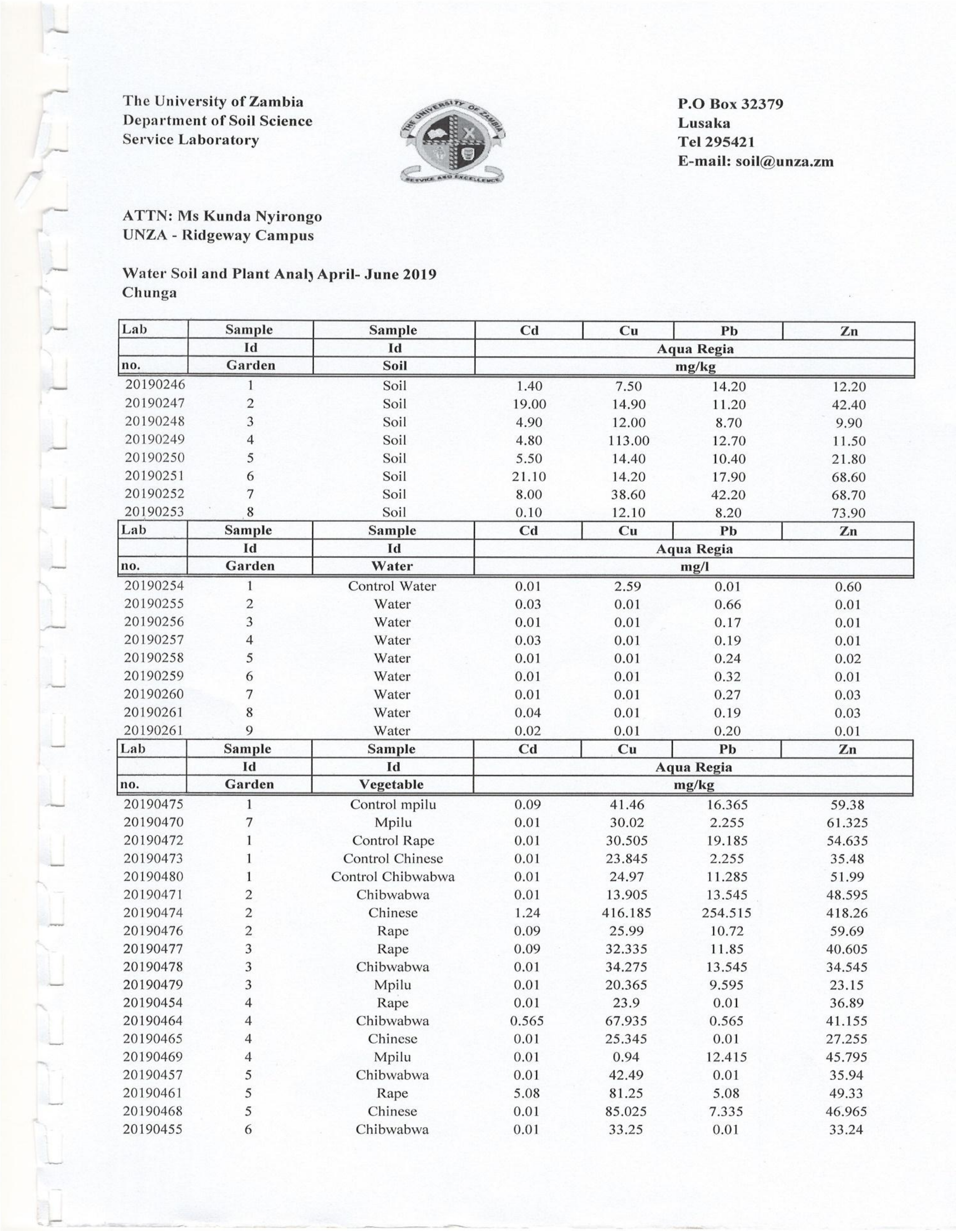
1. The relevant Provincial and District Medical Officers where the study is being conducted are fully appraised;
2. Progress updates are provided to NHRA quarterly from the date of commencement of the study;
3. The final study report is cleared by the NHRA before any publication or dissemination within or outside the country;
4. After clearance for publication or dissemination by the NHRA, the final study report is shared with all relevant Provincial and District Directors of Health where the study was being conducted, University leadership, and all key respondents.

Yours sincerely,

Dr. Godfrey Biemba
Director/CEO
National Health Research Authority

All correspondences should be addressed to the Director/CEO National Health Research Authority

9.6 APPENDIX VII: WASTEWATER, SOIL AND VEGETABLES LABORATORY RESULTS FOR THE STUDY



The University of Zambia
Department of Soil Science
Service Laboratory

P.O Box 32379
Lusaka
Tel 295421
E-mail: soil@unza.zm

ATTN: Ms Kunda Nyirongo
UNZA - Ridgeway Campus

Water Soil and Plant Analy April- June 2019
Chunga

Lab	Sample	Sample	Cd	Cu	Pb	Zn
	Id	Id	Aqua Regia			
no.	Garden	Soil	mg/kg			
20190246	1	Soil	1.40	7.50	14.20	12.20
20190247	2	Soil	19.00	14.90	11.20	42.40
20190248	3	Soil	4.90	12.00	8.70	9.90
20190249	4	Soil	4.80	113.00	12.70	11.50
20190250	5	Soil	5.50	14.40	10.40	21.80
20190251	6	Soil	21.10	14.20	17.90	68.60
20190252	7	Soil	8.00	38.60	42.20	68.70
20190253	8	Soil	0.10	12.10	8.20	73.90

Lab	Sample	Sample	Cd	Cu	Pb	Zn
	Id	Id	Aqua Regia			
no.	Garden	Water	mg/l			
20190254	1	Control Water	0.01	2.59	0.01	0.60
20190255	2	Water	0.03	0.01	0.66	0.01
20190256	3	Water	0.01	0.01	0.17	0.01
20190257	4	Water	0.03	0.01	0.19	0.01
20190258	5	Water	0.01	0.01	0.24	0.02
20190259	6	Water	0.01	0.01	0.32	0.01
20190260	7	Water	0.01	0.01	0.27	0.03
20190261	8	Water	0.04	0.01	0.19	0.03
20190261	9	Water	0.02	0.01	0.20	0.01

Lab	Sample	Sample	Cd	Cu	Pb	Zn
	Id	Id	Aqua Regia			
no.	Garden	Vegetable	mg/kg			
20190475	1	Control mpilu	0.09	41.46	16.365	59.38
20190470	7	Mpilu	0.01	30.02	2.255	61.325
20190472	1	Control Rape	0.01	30.505	19.185	54.635
20190473	1	Control Chinese	0.01	23.845	2.255	35.48
20190480	1	Control Chibwabwa	0.01	24.97	11.285	51.99
20190471	2	Chibwabwa	0.01	13.905	13.545	48.595
20190474	2	Chinese	1.24	416.185	254.515	418.26
20190476	2	Rape	0.09	25.99	10.72	59.69
20190477	3	Rape	0.09	32.335	11.85	40.605
20190478	3	Chibwabwa	0.01	34.275	13.545	34.545
20190479	3	Mpilu	0.01	20.365	9.595	23.15
20190454	4	Rape	0.01	23.9	0.01	36.89
20190464	4	Chibwabwa	0.565	67.935	0.565	41.155
20190465	4	Chinese	0.01	25.345	0.01	27.255
20190469	4	Mpilu	0.01	0.94	12.415	45.795
20190457	5	Chibwabwa	0.01	42.49	0.01	35.94
20190461	5	Rape	5.08	81.25	5.08	49.33
20190468	5	Chinese	0.01	85.025	7.335	46.965
20190455	6	Chibwabwa	0.01	33.25	0.01	33.24

20190456	6	Rape	0.01	94.17	0.01	37.205
20190467	6	Chinese	0.01	54.84	0.01	46.02
20190459	7	Rape	0.01	21.275	0.01	26.495
20190481	7	Chinese	0.01	25.345	10.72	24.36
20190460	8	Rape	0.01	20.58	0.01	30.22
20190462	8	Chibwabwa	0.01	38.38	0.01	34.37
20190482	8	Chinese	0.01	32.55	10.57	35.36
20190458	9	Chibwabwa	0.01	36.975	0.01	37.63
20190463	9	Rape	0.01	61.455	0.01	33.965
20190466	9	Chinese	0.01	34.22	0.01	30.235

NB: to convert to % divide mg/kg by 10000
Trace = <0.01mg/kg

G. Musukwa
Chief Scientist

Head.
Dept of Soil Science