

THE BIOMAGNIFICATION IMPACT OF DISCHARGING ELEVATED LEVELS OF IRON IN  
MINE EFFLUENT ON HUMAN HEALTH: A CASE OF NAMPUNDWE MINE, CENTRAL  
PROVINCE OF ZAMBIA.

BY

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Dissertation submitted to the University of Zambia in Partial Fulfilment of the requirement  
for the award of Master of Science Degree in Sustainable Mineral Resource Development.

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## **DECLARATION**

I, KAMBAFWILE MWITWA do hereby declare that this report represents my own clear personal effort and experiences during field research data collection and that it had not been submitted at any university for assessment or award of any academic qualification. All published and unpublished material used in this report have been acknowledged and references provided here forth.

Sign: \_\_\_\_\_

Date: \_\_\_\_\_

## CERTIFICATE OF APPROVAL

This dissertation of Mwitwa Kambafwile is approved as fulfilling part of the requirement for the award of the Master of Science Degree in Sustainable Mineral Resources Development.

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## ABSTRACT

This study investigated the biomagnification impact of elevated iron levels in mine effluent on human health around Nampundwe Mine area in Shibuyunji District of Zambia. Nampundwe Mine produces massive Pyrite ( $\text{FeS}_2$ ) with minor Pyrrhotite ( $\text{FeS}$ ) and chalcopyrite ( $\text{CuFeS}_2$ ) used in copper smelters. Pyrite plays a crucial function in copper smelting process, providing a source of sulfur, iron, and fuel that aids copper recovery, reduce slag viscosity and increases smelting efficiency. Due to processing of the mineral, iron is transformed from ferrous to ferric form which is bioavailable to plants when released to the environment through the mine effluent and dust fallout from the tailings dam. The study was motivated by the extensive use of land and mine effluent for small scale farming by the local community. The study objective was to assess the impact of elevated levels of iron in mine effluent on the health of the Nampundwe Mine community.

The use of land near mines and mine effluent for agriculture purposes could result in biomagnification of heavy metals. Biomagnification is the increase in concentrations of a substance, in the tissue of an organism in a higher level of a food chain. The study used a mixed-method approach which included laboratory data collection of iron content in soil, mine effluent, vegetable leaves, vegetable soup and blood samples. Blood samples were collected from guinea pigs to represent the animal model. The study discussed the prevalence of diseases associated with elevated iron levels such as acute hepatitis A, B and C, liver damage, cancer, haemolytic anaemia, liver injuries, and dry skin disorder using a questionnaire. The study used stata and excel in analysing laboratory data, and descriptive statistics of iron content in serum and vegetable in determining the iron content.

The finding was that iron content in effluent was within the ZEMA threshold of 2mg/l for effluent water but above the WHO/FAO threshold of 0.5mg/l for irrigation water. Further, the iron content showed an increase in the leafy vegetables from 24.67 mg/l to 32.8 mg/l, from 26.31mg/l to 45.4mg/l and from 27.13mg/l to 32.1mg/l. Furthermore, iron levels in guinea pig serum equally increased by 23 per cent, 67 per cent, 13.5 per cent and 55 per cent. The results confirmed a pass-through effect of iron biomagnification in shoots of fresh vegetables via the food chain. Similarly, the guinea pigs exhibited signs of iron overload in form of a dry skin disorder.

With these findings, of effluent water results being above the WHO/FAO threshold for irrigation water, it is recommended that authorities such as Zambia Environmental

Management Agency consider enforcing compliance with Mining Regulations at source. With the observed biomagnification in effluent irrigated vegetables, ZEMA to consider restricting the use of plant effluent for agriculture purposes. In a multi-sectoral intervention, ZEMA is recommended to work with Ministry of Health, and to sensitize the catchment population on the dangers of consuming mine effluent irrigated vegetables and improve the cancer status in mining communities.

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## ACRONYMS

FAO	Food Agriculture Organization
GI Tract	Gastrointestinal Tract
GRI	Global Reporting Initiative
HFE	High Iron Genes
HDPE	High Density Polyethylene
KCM	Konkola Copper Mines
MoH	Ministry of Health
UNZA	The University of Zambia
WHO	World Health Organization
FAO	Food Agriculture Organization
ZEMA	Zambia Environmental Management Agency
SP	Specimen

## CHAPTER ONE

### INTRODUCTION AND BACKGROUND

#### 1.0 Introduction and Background

Nampundwe Mine is one of four business units operated by Konkola Copper Mine PLC. It is located approximately 41.16 km from Lusaka Central Business District. Nampundwe Mine is an underground mine operating a pyrite processing plant. Pyrite is used in copper smelting and roast leach electro-winning is a common metal sulphide mineral in tailings and valuable mineral raw material Peppas et al., (2000). Toxic heavy metals of pyrite leachate could enrich in organisms and enter human body via food chain and are harmful to people's health.

Mining operations generate huge volumes of waste in form of effluents and tailing discharged to the natural environment. The effluent is a combination of mine dewatering, process plant wastewater, storm water runoff and tailing dam discharge. The effluent is released to the environment through two ZEMA licenced points as shown in figure 1.0. Environmental monitoring is conducted as prescribed in licence conditions (ZEMA, 2011).

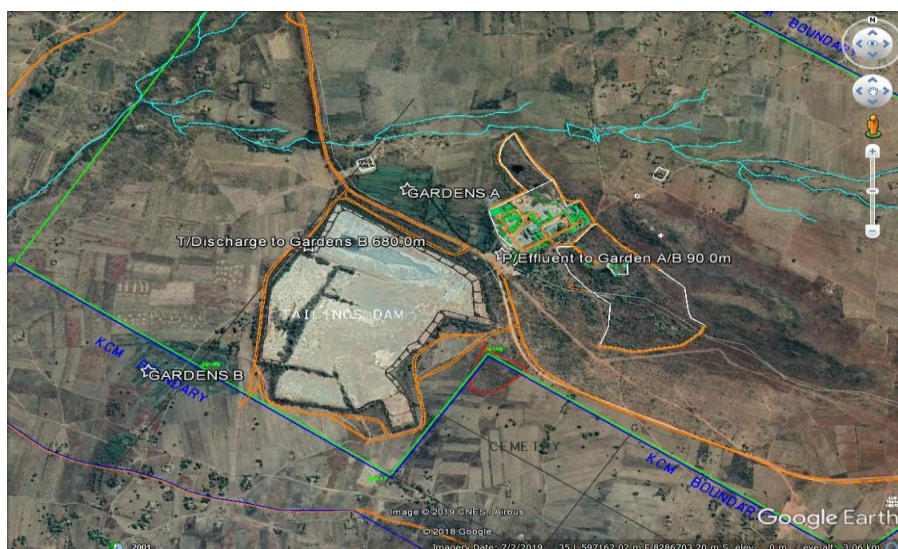


Figure 1.0: Effluent Discharge Points *Source: Google Earth*

The tails are transported in slurry form using high density polyethylene (HDPE) pipelines to the Tailing storage facility. Pyrite is exposed to the environment through pipeline failures and spillages during handling of concentrate. When pyrite is exposed to the environment, it undergoes a process called oxidation. Oxidation is the chemical process which involves the loss of electrons. Oxidized pyrite released to the environment is potentially absorbed by plant

roots from the soil solution most readily as ferrous (Fe<sup>2+</sup>) but also in some cases as ferric (Fe<sup>3+</sup>) (Kobayashi and Nishizawa, 2012).

Nampundwe has recently experienced an unprecedented population increase, tied to the creation of Shibuyunji District, which has increased water usage in the district (NWASCO, 2021). Access to water is a human right under Sustainable Development Goal number six on clean water and sanitation (United Nations, 2023). The observed phenomenon in Nampundwe is that agriculture is the main economic activity. Social Management of water is changing, as numerous companies and consultant studies report that failure to earn a social license (Gehman et al., 2016), approval of a mining project by the community, has lately poised the most significant risk in mining companies. The main agricultural activity is vegetable growing, which utilizes the water from the mine for crop production. These gardens are located near the main plant and the tailing storage facility. The oxidized pyrite released to the environment is potentially absorbed by the vegetables grown and irrigated using the plant effluent.

The Mine effluent discharged by Nampundwe Mine finds its way to the Kacheta stream and other usable water sources. As per the Nampundwe Global Reporting Initiative (GRI) report of 2017, the first quarter of 2017 generated non-compliant results averaging 2.28 mg/l of iron with maximum iron values at 5.03 mg per litre, as shown in Figure 1.1. Nampundwe recorded high iron content from boreholes BH 3 and BH 4. This iron level stood at 3.03 mg/l above the ZEMA threshold of 2 mg/l (i.e., 51% above the limit). According to WHO/FAO (2003), the permissible limit of iron in water for irrigation is 0.5 mg/l.

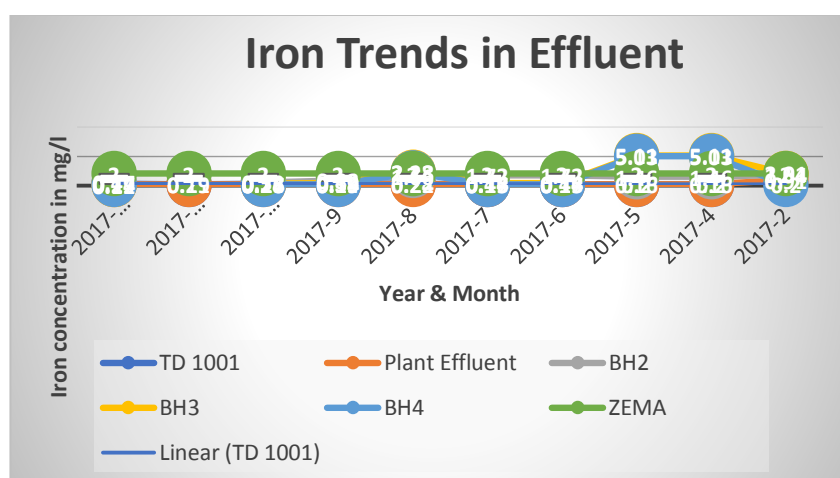


Figure 1.1: Iron Trends in Effluent

The vegetable rape, scientifically called *brassica napus*, and Chinese Cabbage, also known as *brassica rapa*, are commonly grown vegetables in Nampundwe near the mine site. *Brassica napus* is mostly grown using the fertilization-based strategy called biofortification, in which chemical stimulants aid the uptake and storage of essential nutrients. Iron is a micronutrient and its availability to the plant roots depends on the pH level of soil. Becker et al., (2017) argues that this brassica species, *rapa*, has high affinity for microelements and can be grown both in dry and wet season. The bioavailability of some minerals to humans, for example iron, may be increased by cooking (Lee & Clydesdale, 1981), Kałużewicz et al., (2018) and Popko et al., (2018). This could be the situation in the Nampundwe community. The elevated level of iron in the effluent used to water the vegetables may have a pass-through effect by consuming the brassica specie.

The effluent is used by the community for irrigating vegetable gardens (Figure 1.2). The national concern on the access to water is being deepened by mineral deposits and successful mineral explorations increasingly being in water-scarce regions (Rockström, 2014). Nampundwe is located on a highly water-scarce area, where KCM is conducting its mineral extraction. Nampundwe has been facing competing water demands, such as, domestic, agriculture, subsistence farming, livestock herding and fisheries.

The exhibited pictures in Figures 1.2 (a and b) and figure 1.3 (a and b) show some of the main agricultural activities that the host community of Nampundwe Mine are engaged in, using the plant effluent. The main activities include vegetable productions, such as, pumpkin leaves, rape, Chinese Cabbage, and animal rearing.



(a) Rape fields



(b) Tomato field

Figure 1.2: (a) and (b): Small Scale Vegetable Farming fields by the community



(a) Cattle drinking mine effluent

(b) Cattle drinking mine effluent

Figure 1.3: (a) and (b): Small Scale Animal Rearing cattle by the community.

Studies on heavy metal poisoning such as Mulanga, (2016) and Corley & Mutiti, (2017), despite determining the lead concentration and its poisoning effects in patients, did not Carry out either the pass-way analysis of lead in humans or conduct the animal model experimental design. This study employed a mixed-method approach to not only determine the pass-way of iron but also the possible effect of elevated iron on human health.

### 1.1 Statement of the problem

Wastewater generated at Nampundwe Mine is from underground dewatering activities, process plant wastewater and stormwater runoff which is discharged into Kacheta Stream as Mine effluent. However, before the Mine effluent gets into the Kacheta stream, it is used for the irrigation of vegetables by the local community. Research suggests that elevated iron in water may be associated with an increased risk of several health conditions (Anne et al., 2003), Capell, (2004) and Kalyani & Srinivas, (2018). The most likely cause of public health concern are coronary heart disease (CHD), cancer, elevated blood sugar and type 2 diabetes (Bruce & Robert, 2006), Corley & Mutiti, (2017). The iron intake in humans is mainly through a wide range of meat, refined foods and vegetables. The iron level in the human body can also be traced through the biomagnification of iron elements across the food chain (Sayadi et al., 2020). However, there is no convincing evidence that elevated iron status increases the risk of coronary heart disease, type 2 diabetes, cancer, hepatitis fibrosis, hepatic cellular carcinoma and bronze diabetes. The link between the biomagnification of iron via the food chain and its

associated health risks must therefore be determined to contain the escalating levels of non-communicable diseases.

## 1.2 Study objectives

### 1.2.1 Main objective

The study's main objective is to empirically examine the impact of elevated levels of iron content in effluent on the health of Nampundwe Mine Community. To achieve this, the research study specific objectives are as given below.

### 1.2.2 Specific objectives

- i. To determine the water quality of mine effluent used in irrigating vegetables in Nampundwe.
- ii. To evaluate the iron level in the local soil structure in Nampundwe mine area.
- iii. To establish the iron levels in the vegetables grown by subsistence irrigation agriculture, using the Mine effluent water.
- iv. To assess the biomagnification of iron in the food chain through the vegetable to the human route.
- v. To establish the effect of elevated iron levels on the health of the Nampundwe community. This is through investigating evidence of symptoms associated with elevated iron in human. This will be done by examining local clinic medical records.

## 1.3 Research questions

- i. Does the effluent at Nampundwe Mine meet the regulator's quality standards?
- ii. Is Bioconcentration of iron exhibited in the vegetables grown by subsistence irrigation using Mine effluent?
- iii. Does elevated iron level transfer occur from effluent water via irrigation to vegetables to the shoots of rape and Chinese Cabbage in Nampundwe?
- iv. Does iron overload (above statutory limit) in water have implications on the health of the community?

#### 1.4 Research hypotheses

Elevated iron levels causes non-communicable diseases, acute hepatitis A, B and C, liver damage, cancer, haemolytic anaemia, liver injuries, diabetes mellitus type 1 and 2, Acquired haemolytic anaemia-unspecified, and dry skin disorder in Nampundwe Area.

#### 1.5 Significance of the study

There is no known existing convincing and clear-cut empirical evidence from studies in Zambia and other countries that have shown that elevated iron in water increases acute hepatitis A, B and C, liver damage, cancer, haemolytic anaemia, liver injuries, diabetes mellitus type 1 and 2, acquired haemolytic anaemia-unspecified, and dry skin disorder.

There is a need to accurately determine the health risks associated with elevated iron levels in the water. Such a correlation would help Mines manage and mitigate their business risk against the health of the surrounding communities that utilize Mine effluent. Further, the study's outcome will aid and guide health policymakers to design health policy interventions that mitigate the escalating problem of non-communicable diseases and improve the health of the Zambian population. Furthermore, the problem of high heavy metal content in mine effluent could inform environmental regulators of the need to increase the frequency of inspections and redesign the environmental legal framework, to enhance community safety and compliance.

#### 1.6 Limitation of the Study

The shortcomings of the study included but were not limited to the following:

- i. The research study was limited to using guinea pigs to mimic the physiology of humans. The blood samples should have been collected from humans to determine the effect of elevated iron levels in Mine effluent on the community's health.
- ii. This study was operationalized on the assumption that other causes of acute hepatitis A, B and C, coronary heart disease (CHD), cancer, hemolytic anaemia, liver injuries, Diabetes mellitus type 1 and 2, Acquired hemolytic anaemia-unspecified, Dermatitis or eczema, unspecified and dry skin disorder, such as congenital and opportunistic infections due to chronic diseases and other infectious hepatitis B and C were not put into consideration. This is particularly so that only cases directly related to iron overload were looked at.

## 1.7 Structural Outline

In Chapter 1, the study has been introduced with the research objectives and questions highlighted. The value of such research has been argued and the limitations discussed. The rest of the thesis is structured as follows: Chapter 2 will focus on the theoretical and empirical literature review, while chapter 3 will delve into the study's methodology. Specifically, section 3.1 will discuss the research design. Chapter 4 will detail the estimation, presentation and interpretation of the results. Section 4.1 provides descriptive statistics, while section 4.2 discusses the trend analysis of the serum and vegetable samples. Finally, chapter 5 will discuss the conclusion and areas for further research and make policy recommendations.

## CHAPTER TWO

### LITERATURE REVIEW

#### 2.0 Introduction

This section provides the theoretical and empirical literature underpinnings. The study in this section highlights empirical findings of the effect of biomagnification of iron via the food chain on human health.

#### 2.1 Theoretical literature review

Animal models in medical and scientific investigations have been widely used. Rodents, rexes monkeys and guinea pigs have been valuable instrumental specimens in medical and scientific works in determining underlying genetic and pharmacological compositions (Van et al., 2020), (Frank & Igor, 2014), (Lynda et al., 2007) & (Charles & Eliot, 2005).

##### 2.1.1 Hepatic Fibrosis in Experimental Iron Overload Model

Chang et al., (1959) developed an experimental iron overload model and found portal fibrosis in rabbits and dogs receiving long-term parenteral injections with iron chelates. In patients, the development of fibrosis and cirrhosis are the major clinical manifestations of the disease Terrault et al., (2018) and Frank & Igor, (2014).

Many experimental protocols have been unsuccessful in producing hepatic fibrosis and cirrhosis after administration of iron to mice, rats, guinea pigs, rabbits, dogs and rhesus monkeys due to inadequate degree of elevated iron (Ibid). However, there are a few reports, such as, Van et al., (2020), and Starley et al., (2010) of hepatic fibrosis being produced in experimental models of iron overload. In baboons, the clinical trial produced mild portal fibrosis in association with a transient increase in hepatic Prolyl-hydroxylase activity after long-term injections of iron-polymaltose.

Iron is a carrier of oxygen to the tissues from the lungs via red blood cell haemoglobin as a medium for electrons within cells and is an integrated part of enzyme systems in various tissues (Gupta, 2014). However, it was concluded that excess tissue iron could provide a direct stimulus to collagen biosynthesis and hepatic fibrosis without prior iron-mediated cellular injury. The other effect of elevated iron was the patches of sores on the body of the rats coupled

with damaged heart organs. Therefore, elevated iron level causes some forms of cancer and heart injuries Chang et al., (1959).

## 2.2 Empirical Literature Review

This section delves on a review of empirical literature and their findings on heavy metal biomagnification, specifically iron. This focus review of literature is basically on biomagnification in terrestrial ecosystem through the following route: transfer of heavy metals from soils to plant and subsequently plants to animals (Zeng et al., 2022).

### 2.2.1 Biomagnification in Terrestrial Ecosystem

Sharma et al., (2020), Kałużewicz et al., (2018) and Popko et al., (2018) argue that the application of foliar improves the acquisition of microelements from the soil to the plant which causes bio-concentration in plants and subsequently biomagnification of heavy metals in terrestrial ecosystem,

Heavy metals are mostly rapidly acquired by plants in their development stages from germination Kosakivska et al., (2021) and Vclko & Ohnoutkova et al., (2019). The terrestrial transfer of heavy metals from plants to humans is via consumption of cooked vegetable soups which has been empirically verified to contain bioconcentrated levels of heavy metals in shoots and leafy vegetables Sadee et al., (2023), Niebla-Canelo et al., (2022), Wang et al., (2022), Mutiti & Corley (2017) and Mulanga (2016).

Corley & Mutiti, (2017) on a study on “The Effects of Lead Species and Growth Time on Accumulation of Lead in Chinese Cabbage in Zambia”, argues that the primary pathway for heavy metal exposure in contaminated areas is via consumption of locally produced food. The study investigated the accumulation of lead in Chinese Cabbage grown in contaminated soils and estimated the weekly dietary intake. Experiments were conducted to determine the effects of different growth times, concentrations, and lead species (carbonate, nitrate, and sulphide) on the uptake of lead in shoots. Results showed that Chinese Cabbage accumulated up to 38 mg/kg in the shoots. This deduction entails that the heavy metal in vegetables increases as the plant grows and is linked to the increase in lead level in the soil via biomagnification.

Jara-Marini et al., (2020) and Liangqian et al., (2016) argues that sulphide rich tailings oxidize spontaneously when exposed to air and water, in instances that tailings have low organic matter content with no buffering capacity, easily releases metals such as iron to the environment.

When iron is released to the environment, it is absorbed by plant depending on the iron availability in the soil. Iron availability in soil refers to the amount of iron accessible by plant for uptake and use. High levels of iron in plants causes necrotic spots, yellowish plant leaves, dark roots and inhibition of plant growth through indirect and direct iron toxicity Ghosh et al. (2022) & Martinez et al., 2021.

In the environment, iron is available in two biologically important forms that include ferric and ferrous. In acidic environment, iron acts as a reducing factor while in basic environment iron acts as an oxidant Cesar et al., (2016) and Jones et al., (2011).

### 2.2.2 Health Risks Associated with Biomagnification

Empirical findings have shown that there is an array of health risks associated with the consumption of food stuffs with heavy metal concentrations, such as, vegetable shoots and leaves by herbivores as well as cooked vegetable soups and fresh meats by carnivores Sadee et al., (2023), Wang et al., (2022) and Ankar-Brewoo et al., (2020). Elevated levels of iron in organisms used as specimen and laboratory determined heavy iron content presence have revealed to be stimulants for human skin diseases such as skin cancer Giani et al., (2021), Rauf et al., (2020) and Chavatte et al., (2020). Increased intestinal absorption of iron causes health impairments, such as liver damage and skin hyper-pigmentation (Capell, 2004).

Giani et al., (2021) on a study on “Heavy metals in the environment and thyroid cancer” chronic exposure to heavy metals, even at slightly increased environmental concentrations that cause no harm to mature thyrocytes, may alter the biology of stem/precursor thyroid cells, leading to a predisposition to malignant transformation.

Naddafi et al., (2022) and Witkowska et al., (2021) argue that the cancer cases, deaths and death rate increased with the exposure to drinking water that contained elevated levels of iron content. Further, the studies revealed that metals disturb native proteins’ by catalysing the oxidation of amino acid side chains, perturbing protein folding as well as displacing essential metal ions in enzymes.

Kalyani & Srinivas, (2018) argue that hemochromatosis is associated with porphyria cutanea tarda because of iron load, which affects the quantity and activity of uroporphyrinogen decarboxylase by which uroporphyrinogen accumulates in the skin and present as blisters.

Maki et al.,(2017) on a study on “The Influence of Hydrous Ferric Oxide, Earthworms, and a Hyper Tolerant Plant on Arsenic and Iron Bio-availability fate, and Transport in Soils” in America argues that the application of arsenate pesticide increase the mobility and bio-availability of iron, evidenced by increased iron concentration in earthworms ,plants and leachate.

Current studies, such as Kalyani & Srinivas, (2018) on a study of “Primary Hemochromatosis Presenting as Type 2 Diabetes Mellitus: A Case Report with Review of Literature”, argues that iron overload causes tissue damage in liver, pancreas, skin, joints, heart, and gonads. The authors further argue that approximately 50 per cent of patients diagnosed with hemochromatosis had either type 1 or type 2 diabetes mellitus, due to iron overload which leads to impaired insulin synthesis, release, and insulin resistance. This revelation clearly shows that elevated iron content causes skin impairment, heart disease and diabetes. Sandros et al., (2021) and Sousa et al., (2020) argue that non-alcoholic fatty liver disease and steatosis are also associated with alteration in iron metabolism and hepatic iron accumulation in which excess free iron triggers oxidative stress and switches towards macrophage pro-inflammatory status.

### 2.3 Summary

The theoretical and empirical literature has revealed that biomagnification of heavy metals occurs across the food chain. For instance, four weeks old vegetable shoots showed lower iron level content than fully grown eight weeks old vegetable leaves. Further, cooked vegetable soups revealed high levels of heavy metal content such as lead and iron. Hfe genes in human beings have a high affinity for water and iron absorption. Furthermore, the determination of iron levels in human beings and the associated health impairment has been carried out using animal models as opposed to actual experiments on human beings. Finally, literature has revealed that there exists a linkage between high iron and a wide range of cancers like cirrhosis, liver injuries and dry skin condition.

Research on the implications of heavy metals on agricultural productivity and soil health has been extensively carried out in Lusaka (Kapungwe, 2015), Zambia’s mining areas (Matthews et al.,2023), Kabwe (Simukanga, 2017) and Copperbelt province (Muimba et al.,2022). However, most studies have focused effects of heavy metals on crop yields (Schelenker & Roberts, 2009). Few studies in Zambia such as, Mutiti & Corley (2017) and Mulanga (2016), revealed the effect of lead on health of communities in Kabwe. This study on the other hand

specifically addresses the effect of elevated iron on health via the food chain in Nampundwe. The gap is particularly evident in the context of smallholder farming systems that use the mine effluent in the irrigation of a wide range of crops. If elevated iron is present, it has potential to cause devastating consequences for livelihoods and health outcomes. The findings of this study will aid mines manage and mitigate their business risk against the health of the surrounding communities that utilize mine effluent. Further, the study's outcome will guide health policymakers to design health policy interventions that mitigate the escalating problem of non-communicable diseases and improve the health of the Zambian population.

## CHAPTER THREE

### METHODOLOGY

#### 3.0 Introduction

This chapter details the methodology used to carry out the estimations and data collection, both primary and secondary data. The study used four-tier primary data sources, namely: Mine effluent sampling, experimental vegetable field data, experimental animal data from guinea pigs and the health facility data on the prevalence of cancers, diabetes and hepatic diseases in the Nampundwe area. The chapter further describes the nature of the data to be used and variable labelling. This chapter covers topics that include model specification, data sources and estimation procedures.

#### 3.1 Research Design

##### 3.1.1 Study design and sites

The study conducted a descriptive analysis using mixed methods among other thing, vegetable iron analysis in fresh leaves, cooked leaves and cooked vegetable soup for purposes of quantitatively determining the iron level present. Further, a data collection tool was constructed based on the diseases reported in the standard Ministry of Health (MoH) of Zambia disease reporting form. The standard diseases form was used to streamline identifiable non-communicable diseases, such as, acute hepatitis A, B and C, liver damage, cancer, hemolytic anaemia, liver injuries, diabetes mellitus type 1 and 2, acquired hemolytic anaemia-unspecified and dry skin disorders as per the standard diseases reporting form.

The study was conducted in Shibuyunji District of the Central Province, specifically in Nampundwe, where the field treated with effluent water was located within a 5-kilometre radius of the effluent discharge point. The control field was situated in Mwembeshi, 35 kilometres from the plant effluent discharge point, which had similar conditions except for the source of water for irrigation. Mwembeshi control field was irrigated using a ground water aquifer through a borehole.

##### 3.1.2 Research Approach

The study adopted a mixed approach procedure in obtaining evidence and implications of elevated iron levels on human health in Nampundwe. The information was obtained from peer-

reviewed literature, ZEMA, WHO and other policy documents on environmental management. The prevalence rate of diseases, such as acute hepatitis A, B and C, liver damage, cancer, hemolytic anaemia, liver injuries, diabetes mellitus type 1 and 2, acquired hemolytic anaemia-unspecified, and dry skin disorder was determined using data analysis from the study questionnaire. Purposive sampling was used for health personnel, among them medical doctors, nurses, clinical officers and laboratory technicians. A total of thirty participants took part in the online survey that was emailed to them and completed electronically. SurveyMonkey, for online data collection and analysis platform, was utilized in designing the survey and collecting the survey responses.

### 3.1.3 Conceptual Framework

Figure 3.0 exhibits the interlink between a person's iron status and the subsequent effect of the elevated iron level on the health of human beings. Hemochromatosis, a condition that may develop because of increased iron absorption in the Gastrointestinal (GI) tract, which includes the mouth, oesophagus, stomach, small intestine, large intestine, and anus, causes hepatic parenchymal. This could be a rare tumor of vascular origin with a dilated and elongated hepatic artery with a non-specific symptom such as right upper quadrant pain and weight loss, liver failure, and hepatic failure as a result of a direct effect of elevated iron level and increased collagen synthesis as well as lipid peroxidation of organ membrane Van et al., (2020) , Bruce & Robert, (2006) & Maurizio et al., (1996). It manifests itself as skin cancers and liver cancers. Peroxidation or the membrane burning dysfunctional, causes alteration in enzymes, causing cell injuries and death in cases where there is confirmed elevated iron in humans or laboratory experiments on animal models. Decreased degradation due to increased collagen may result in fibrosis, cirrhosis and hepatocellular carcinoma, a group of cancers.

### 3.1.3.1 Conceptual Framework Diagram

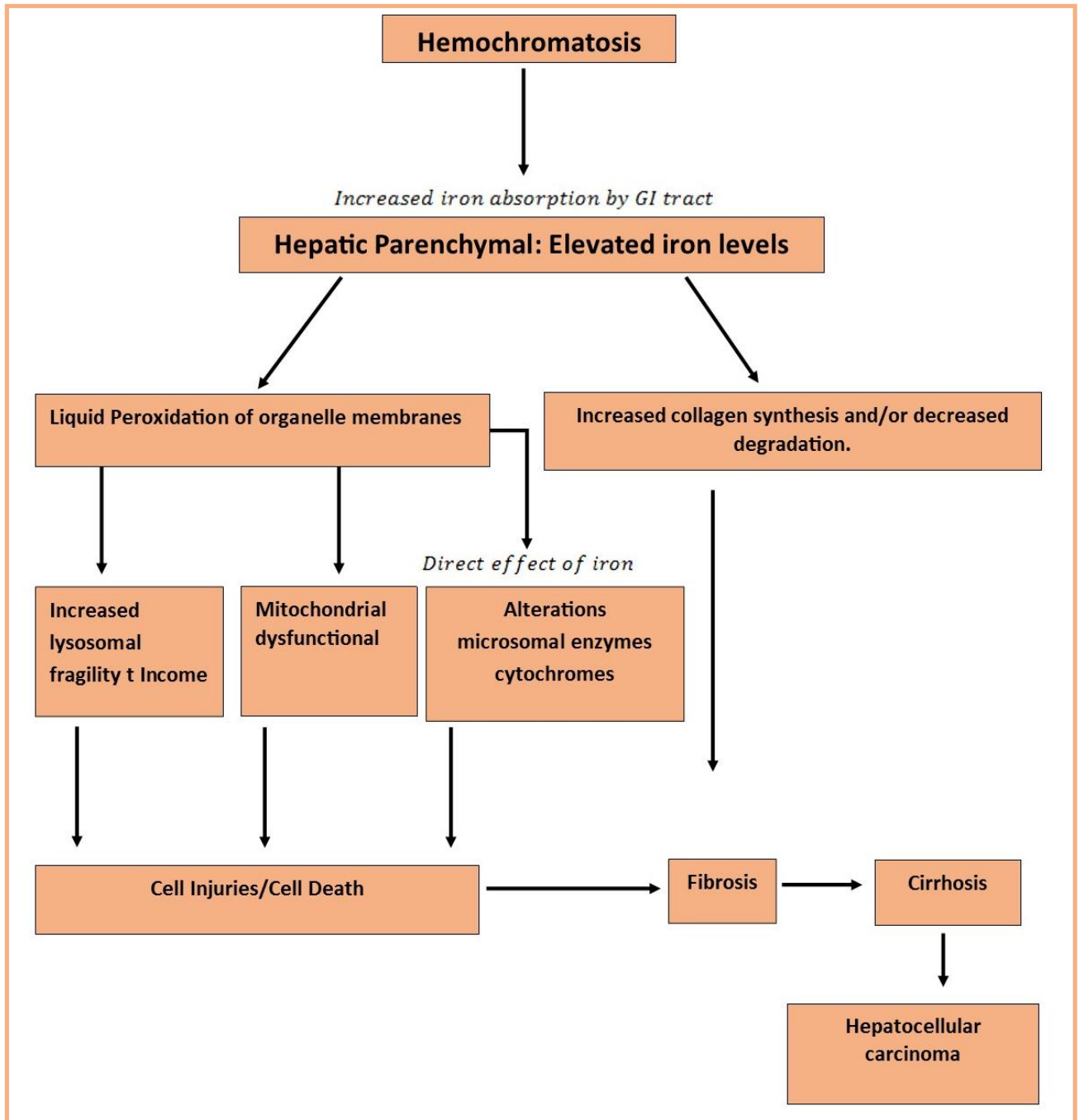


Figure 3.0: Relationship between iron content and health status

Source: Bruce and Robert (2006) "The Pathology of Hepatic Iron Overload: A Free Radical-Mediated Process" Shreveport, Louisiana.

#### 3.1.4 Mine Effluent Data Collection and Variable Definition

Water samples were collected from various points of the plant effluent sampling points, namely, the tailing dam discharge sampling point and Mwembeshi control water source. The water samples were taken for analysis at the University of Zambia's Environmental Engineering laboratory (School of Engineering). Based on these assessments, vegetable fields with two brassica species blocks were assigned codes as effluent water-treated fields at Nampundwe, and the control-water-treated field was erected at Mwembeshi. The codes were used to summarize the findings of elevated iron as shown in Table 3.0. According to the variable definition, crops irrigated with mine effluent were labelled Effluent Water Treated Rape (EWT-R (Rape) and Effluent Water Treated Chinese Cabbage (EWT-C (Chinese Cabbage). The field which was irrigated by control water was labelled (CWT- R (Rape) and CWT-C (Chinese Cabbage). Laboratory data were collected to ascertain the extent of iron concentration in the crops cultivated, and the results were coded.

Table 3.0. Coding and Variable Definition

<b>CODE</b>	<b>EXPLANATION</b>	
$EWT_R$	Irrigating rape using water from Mine effluent	
$Fe_{EWT_R}$	1	First sample iron level for treated rape
	2	Second sample iron level for treated rape
$EWT_C$	Irrigating Chinese Cabbage using water from Mine effluent	
$Fe_{EWT_C}$	1	First sample iron level for treated Chinese Cabbage
	2	Second sample iron level for treated Chinese Cabbage
$CWT_R$	Control plants: Irrigating rape using non-mine effluent water	
$Fe_{CWT_R}$	1	First sample iron level in control rape
	2	Second sample iron level in control rape
$CWT_C$	Irrigating Chinese Cabbage using non-mine waste effluent	
$Fe_{CWT_C}$	1	First sample iron level for control Chinese Cabbage
	2	Second sample iron level for control Chinese Cabbage
$Fe_{S_C}$	Control soil iron level	
$Fe_{S_T}$	Treated soil iron level	
$Fe_{H_2O_T}$	Iron level in mine effluent used to irrigate rape and Chinese Cabbage	
$Fe_{H_2O_C}$	Iron level from the control used to irrigate rape and Chinese Cabbage	
$Cok_{Sop}$	Cooked rape and Chinese Cabbage soups.	
$Fe_{Sop_R}$	Iron level in cooked treat rape soup	
$Fe_{Sop_C}$	Iron level in cooked treated Chinese Cabbage soup	
$Fe_{Sop_C_{Contrl}}$	Iron level in cooked Chinese Cabbage from the control field	
$Fe_{Sop_R_{Contrl}}$	Iron level in cooked Rape from the control field	

### 3.1.5 Field Experimental Design for Brassica Napus and Brassica Rapa

The study used the brassica plant garden which contained each treatment for Rape and Chinese Cabbage. This procedure is adopted from Corley & Mutiti, (2017) on the study on “The Effects of Lead Species and Growth Time on Accumulation of Lead in Chinese Cabbage” in Kabwe.

Chinese Cabbage plants (Rapa type, with a 40-day maturity growing period) were transplanted from the nursery at Nampundwe for effluent water treated vegetables and Mwembeshi for the control water treated vegetables. Two hundred (200) plants of rape and Chinese Cabbage seedlings respectively were transplanted into effluent water treated field and control water treated fields. The treated field had 200 plants of rape and Chinese Cabbage and 200 plants of each Brassica species were transplanted into the control fields. In this portion of the study, plants were harvested and processed after 40 days of growth to allow for maximum growth. During harvest, entire Chinese Cabbage and rape plants were uprooted and the shoots separated from the roots.

According to the Ibid (2017) experimental procedure, Chinese Cabbage was harvested and tested for lead concentration at the 28<sup>th</sup> day and 40<sup>th</sup> day of the shoot development. This study replicated this procedure and harvested both rape and Chinese Cabbage at this interval to determine iron concentration. This study was however designed to determine the iron concentration in rape and Chinese Cabbage on both the fresh leaves as well as the cooked soups. Figure 3.1 and Figure 3.2 shows the two fields of the effluent water treated rape and Chinese Cabbage at four weeks and eight weeks and the control water treated rape and Chinese Cabbage at the same stages.



(a) Chinese Cabbage at 4 weeks



(b) Rape at 4 weeks

*Figure 3.1:* (a and b): Vegetable fields Chinese Cabbage and rape at four weeks



(a) Chinese Cabbage at 8 weeks



(b) Rape at 8 weeks

*Figure 3.2:* (a and b): Vegetable fields Chinese Cabbage and rape at eight weeks

### 3.1.6 Experimental Design on Guinea Pigs

A total of eight guinea pigs were used in the experiment and divided as follows: Four for the control experiment, and the remaining four were placed under treatment by feeding them EWT- Rape and EWT – Chinese Cabbage. At each stage of data collection of the iron level in the crops, the treated were fed on rape and Chinese Cabbage from Nampundwe for the same period of days. Blood samples were tested for serum iron content before the guinea pigs were fed treated rape and Chinese Cabbage and after eight weeks of treatment.

### 3.1.7 Primary Data Collection from Health Facilities

Primary data from health facilities on the trends and disease prevalence was collected from health facilities within a 35-kilometer radius of the Mine effluent discharge point, as shown in figure 3.3. The clinics sampled include Nampundwe mine clinic (3.5km), Nampundwe Rural Health clinic (5.2 km), Masiteki Rural Health Centre (15.8 km), Sichobo Rural Health Centre (23 km), Mwembeshi Rural Health Centre (27 km), and Public Health Department within KCM premises.



Figure 3.3: Location of the Health Facilities in Nampundwe Source: Google Maps

The participants at the health facility level were active health workers including two medical doctors, eight clinical officers, sixteen nurses and four laboratory technologists. The target interviewees were highly knowledgeable on the laboratory analysis and occurrences of diseases, such as acute hepatitis A, B and C, liver damage, cancer, haemolytic anaemia, liver injuries, diabetes mellitus type 1 and 2, acquired haemolytic anaemia-unspecified and dry skin disorder, which was very helpful for the study. A structured questionnaire with two sections attempted to ascertain the prevalence of diseases associated with an elevated iron level.

### 3.1.8 Data Source and Type

Primary field data were used in the study, particularly on iron concentration in the two segmented fields, one in Nampundwe and the other in Mwembeshi defined as:  $EWT_R$ ,  $Fe_{EWT_R}$ ,  $EWT_C$ ,  $Fe_{EWT_C}$ ,  $CWT_R$ ,  $Fe_{CWT_R}$ ,  $CWT_C$ ,  $Fe_{CWT_C}$ ,  $Fe_{S_C}$ ,  $Fe_{S_T}$ ,  $Fe_{HO_2-T}$ ,  $Fe_{HO_2-C}$ ,  $Cok\_Soup$ ,  $Fe_{Sop_R}$  and  $Fe_{Sop_C}$  in Table 3.0. The quantitative content of iron was analyzed from the University of Zambia Environmental Engineering laboratory from germination between the 22.10.2020 and 14.12.2020. Further, the incidences of epidemiological trends were collected using a structured electronic questionnaire administered to a total of 30 health professionals from Nampundwe mine clinic, Nampundwe government clinic, Masiteki Rural Health Centre, Sichobo Rural Health Centre, Mwembeshi Rural Health Centre, Nampundwe Rural Health Centre and Public Health Department KCM Nampundwe of Shibuyunji District of the Central Province, Zambia.

### 3.1.9 Ethical considerations

Informed consent to conduct the study was obtained from the University of Zambia School of Mines Research Ethics Committee as well as the National Health Research Authority (NHRA). Further, Shibuyunji District Health Office was availed the NHRA authorization document, and the research study was allowed to be conducted.

### 3.2 Limitation of the Study

The shortcomings of the study included but were not limited to the following:

- i. The research study was limited to using guinea pigs to mimic the physiology of humans. The blood samples should have been collected from humans to determine the effect of elevated iron levels in Mine effluent on the community's health.
- ii. This study was operationalized on the assumption that other causes of acute hepatitis A, B and C, coronary heart disease (CHD), cancer, hemolytic anaemia, liver injuries, Diabetes mellitus type 1 and 2, Acquired hemolytic anaemia-unspecified, Dermatitis or eczema, unspecified and dry skin disorder, such as congenital, opportunistic infections due to chronic diseases and other infectious hepatitis B and C were not put into consideration. This is particularly so that only cases directly related to iron overload were considered.

### 3.3 Summary

The study employed a mixed method approach in determining the possibilities of biomagnification of elevated iron in mine effluent and its subsequent effects of iron overload on the health of Nampundwe Mine community. On the one hand, the study extracted guinea pigs' serum to determine the iron level content for both guinea pigs fed on effluent water treated vegetables and control water treated vegetables. The serum was tested for iron level content at Pine Ridge Laboratory Services. Further, fresh leafy and boiled vegetable soups were tested for iron levels at the University of Zambia's Environmental Engineering Laboratory and Geo-Chemical Analytical Laboratory in the School of Mines. Water and soil samples were analyzed for iron content at the University of Zambia's Environmental Engineering Laboratory. On the other hand, the disease prevalence rate was determined by the responses from the clinicians at selected clinics in Shibuyunji District Health Offices by way of a structured questionnaire.

## CHAPTER FOUR

### RESULTS AND DISCUSSION

#### 4.0 Introduction

This chapter focuses on estimating the models specified in examining the effects of elevated iron levels on the health outcomes of the Nampundwe population. The estimated results of elevated iron by the laboratory blood samples taken from guinea pigs mimic the almost sure morphology of human beings. The justification and relevance of the explanatory variables are discussed, as well as the influence of the results of iron levels of the treated rape and Chinese Cabbage on the health outcomes of the treated guinea pigs. The descriptive statistics of the variables of interest will be discussed in conjunction with the health facility data collected.

#### 4.1. Descriptive Statistics

The statistical output summarizes the distribution of variables used in the study. The descriptive statistics are summaries of both treated and non-treated guinea pigs, soil, rape and Chinese Cabbage to have a clear trend analysis.

##### 4.1.1 Soil Laboratory Findings

Table 4.0 shows the soil sample results that were collected for analysis of iron to establish the iron level of the environment in which the plants were planted. The soil iron content was determined using the atomic absorption spectrometry procedure. The results for iron content were reported in milligrams per litre (mg/l) or parts per million (ppm).

Table 4. 0 Soil Sample Results for iron content

Soil Sample Results (Iron Content)			
	Field 1	Field 2	Field 3
Sample Date	01.10.2020	01. 10.2020	23.10.2020
Iron Level (mg/l)	1.098	2.206	0.9383

The soil samples analysis revealed that the soil sample dated 01.10.2020 for field 1 contained 1.10 mg/l of iron which was within the WHO/FAO (2003) threshold for iron concentration in agricultural soil.

Conversely, the soil sample dated 01.10.2020 for field 2 contained 2.206 mg/l of iron and within the WHO/FAO (2003) threshold concentration in agricultural soil.

The control field soil sample 3 recorded iron content of 0.9383 mg/l which was within the WHO/FAO (2003) threshold for iron concentration in agricultural soil. The level of iron recorded is far below the WHO/FAO (2003) maximum permissible limit of 5000 mg/l. Factors that contribute to iron levels in soil include excess water and aeration. Acidic soils and poorly aerated soils increase iron availability.

#### 4.1.2 Water Laboratory Findings

##### 4.1.2.1 Iron levels in Water

The water samples tests for the study were carried out using a multiplicity water sampling strategy at three different time periods as shown in table 4.1.

Table 4.1 Water Sample Results for iron

Water Sample Results (Iron Content)					
S No.	Effluent				Control Water
	Sample Date	UG	Plant	TD	Mwembeshi
1	30.09.2020	0.9 mg/l	0.6 mg/l	0.01mg/l	0.01mg/l
2	01.10.2020	1.3 mg/l	0.8 mg/l	0.01mg/l	-
3	15.10.2020	0.7 mg/l	0.42 mg/l	0.01mg/l	-
	Average	0.97 mg/l	0.61 mg/l	0.01 mg/l	0.01mg/l

There were varying iron levels between underground (UG) and plant effluent as various environmental conditions during sampling, such as ph. (levels of ph. shown in table 4.2) influences the iron content. UG effluent, on average, for the period recorded an iron level of 0.97 mg/l representing a 51.5 per cent iron below the recommended threshold of ZEMA. Further, the plant effluent recorded an average of 0.61 mg/l iron content which indicated a 69.5 per cent iron content below the ZEMA threshold 2mg/l (ZEMA, 2011) iron requirement. Furthermore, the Tailing Dam (TD) overflow recorded an average of 0.01 mg/l of iron, representing 99.5 per cent of iron below the recommended iron level by ZEMA. Finally, the control water sample revealed a constant iron level of less than 0.01 mg/l, which is also 99.5 per cent in iron content below the recommended ZEMA threshold.

The summary results indicate that all the average iron content were below the recommended ZEMA threshold. The differences in the reading were attributed to the conditions at sampling time which affect heavy metal content such as ph.

However, all the recorded iron levels for the UG and Plant effluent were above the WHO/FAO permissible values (0.5 mg/l) for irrigation waters as shown in figure 4.0. Because vegetables are irrigated using plant effluent, the use of effluent which is above permissible limits is therefore inappropriate.

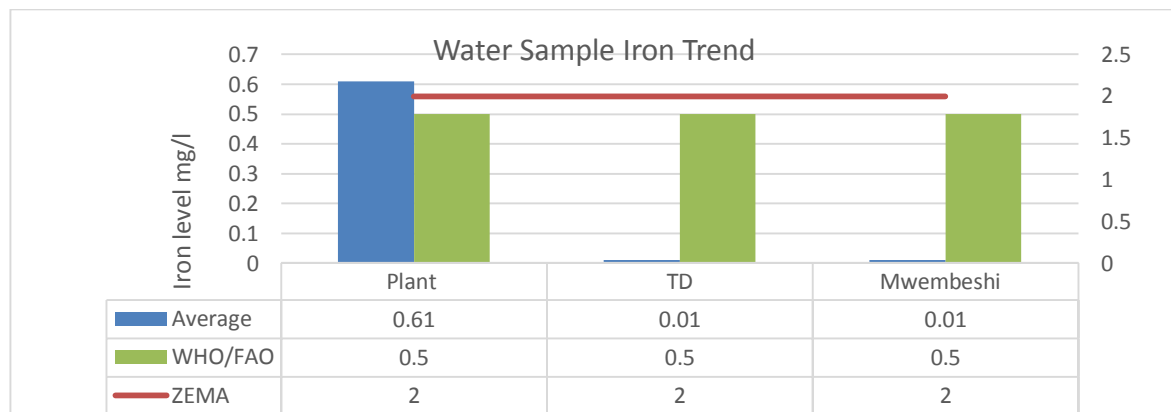


Figure 4.0: Iron Content in water Samples

#### 4.1.2.2 pH levels in Water

The pH level results sampled between 12.09.2022 and 17.10.2022 2022 averaged 7.81 for plant effluent and 7.92 for TD, respectively. The pH average falls within the WHO/FAO (2003) maximum permissible range of 6.5-8.4. A pH of 7 indicates neutrality of the effluent, a pH greater than 7 gravitates towards alkaline while a pH of less than seven designates acidity of the effluent. Acidic condition of effluent entails an increase in the bioavailability of iron according to (Nanae et al., 2016), (Peng et al., 2019) & (Hekmatfar et al., 2018) which increases the rate of absorption by the plant, thereby leading to bioconcentration. Therefore, the absence of the acidic condition entails that the bioavailability of iron is reduced.

The content of iron averaged 0.285mg/l, 0.11mg/l and 0.01 mg/l for the Plant effluent, TD effluent and control, respectively. The iron results in table 4.2 were compliant with both the recommended ZEMA threshold (2mg/l) (ZEMA, 2011) for effluent and the WHO/FAO (2003) threshold for irrigation water is (0.5mg/l).

Table 4.2: Water Sample results for iron and pH level

Water Sample Results (Iron and pH Content)								
S No.	Mine Effluent					Control Water		
	Sample Date	Plant		TD		Sample Date	Mwembeshi	
		Iron (mg/l)	pH	Iron (mg/l)	pH		Iron (mg/l)	pH
1	12.09.2022	0.26	7.84	0.24	7.97	12.09.2022	0.01	6.80
2	19.09.2022	0.32	7.81	0.19	7.98	19.09.2022	0.01	6.87
3	26.09.2022	0.29	7.84	0.20	7.83	-	-	-
4	03.10.2022	0.30	7.82	0.01	7.93	-	-	-
5	10.10.2022	0.26	7.75	0.01	7.86	-	-	-
6	17.10.2022	0.28	7.79	0.01	7.92	-	-	-
	Average	0.285	7.81	0.11	7.92	-	0.01	6.84

#### 4.2 Effluent Treated Raw Leaf Rape and Chinese Cabbage Laboratory Results

The vegetable samples were analysed at the University of Zambia School of Mines Geo-Chemical Analytical Laboratory using the Atomic Absorption Spectrophotometry method.

The results obtained are summarised in table 4.3 and detailed in appendix 3.0.

Table 4.3: Vegetable Leaves and Soup Results (Fe levels mg/l)

Sampling Period	EWT		EWT- Soup		CWT		CWT- Soup	
	Rape (mg/l)	C- Cabbage (mg/l)	Rape (mg/l)	C- Cabbage (mg/l)	Rape (mg/l)	C- Cabbage (mg/l)	Rape (mg/l)	C- Cabbage (mg/l)
Week 4	24.67	26.31	20.1	20.94	26.31	27.13	19.49	18.24
Week 8	32.8	45.4	*	*	13.8	32.1	*	*

\* - Not Analysed

#### 4.2.1 Raw Rape and Raw Chinese Cabbage

The analysis of iron content in fresh vegetable leaves was conducted at a time space of 4 weeks and 8 weeks. The effluent water treated rape recorded 24.67 mg/l level of iron at four weeks, while at eight weeks recorded 32.8mg/l. The iron levels increased by approximately 32.9 per cent signifying that biomagnification occurred. The control water treated rape recorded 26.31mg/l at four weeks and 13.8mg/l at eight which can be attributed to sampling contamination.

On the other hand, effluent water treated Chinese Cabbage recorded 26.31 mg/l level of iron at 4 weeks and recorded 45.4mg/l of iron at 8 weeks. While Chinese Cabbage grown in control water treated fields recorded 27.13 mg/l of iron content at 4 weeks and 32.1mg/l at 8 weeks. An increase of 72.38 per cent and 18.32 per cent respectively.

The biomagnification of iron can thus be quantitatively deduced as having evidently occurred in both the rape and Chinese cabbage as shown in figure 4.1. The finding agrees with (Sayadi et al., 2020), who traced the biomagnification of iron at different germination stages and argue that shoots of Chinese Cabbage absorb a significant amount of heavy metals such as iron.

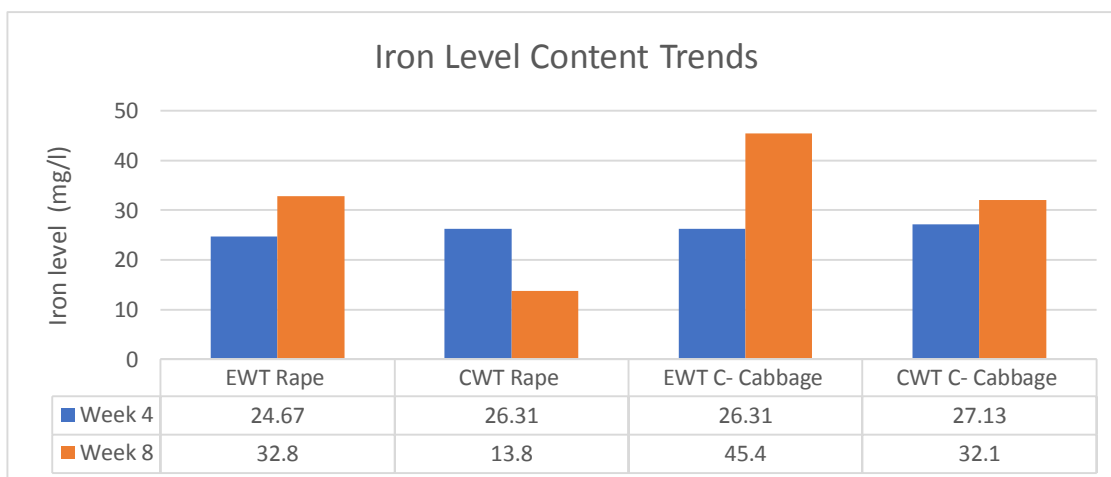
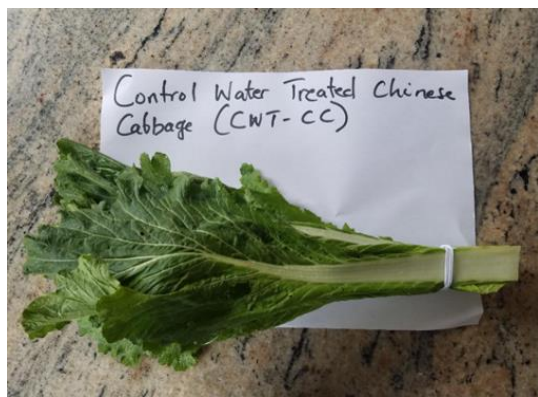


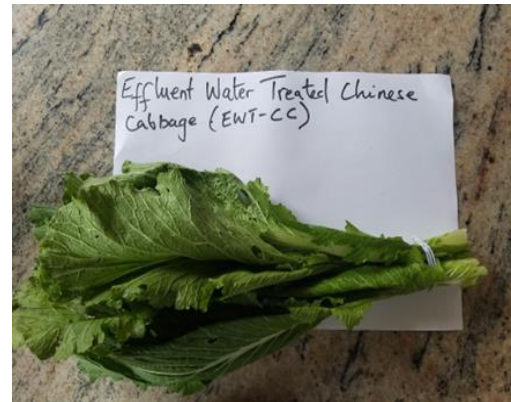
Figure 4.1: Iron level Content trends in fresh vegetables

This biomagnification may have serious plant growth implications such as inhibition of growth, yellowish leaves and dark roots. Figure 4.2 and Figure 4.3 show the differences in plants growth size and colouration for those grown in control water treated field and those grown in effluent water treated field. Both Rape and Chinese cabbage grown in effluent

water treated fields showed necrotic spots, yellowish leaves with leaves being smaller in size (Figure 4.2. b and Figure 4.3.b) compared to the leaves of plants grown in control water fields for both species which were greener and bigger in size (Figure 4.2.a and Figure 4.3.b). The findings agree with the results by Ghosh et al. (2022) & Martinez et al., (2021) who found that iron overload leads to dark root development, yellowish leaves, and inhibited growth.



a) Control water Treated Chinese Cabbage



b) Effluent Water treated Chinese Cabbage

Figure 4.2 (a and b): Fully grown Chinese cabbage CWT- CC and EWT- CC.



a) Control Water Treated Rape



b) Effluent treated Rape.

Figure 4.3 (a and b): Fully grown Rape CWT-Rape and EWT- Rape.

#### 4.2.2 Control Water Treated Chinese Cabbage and Rape Boiled Samples

Boiled Control water treated rape and Chinese Cabbage recorded iron levels of 20.1 mg/l and 20.94 mg/l, respectively, while the control water-treated rape recorded iron levels of 19.49mg/l and Chinese Cabbage recorded 18.24mg/l (Figure 4.4 and Table 4.3). Both the control water

and effluent water recorded a high level in comparison with the baseline of 2.0 mg/l of iron as per the ZEMA recommended threshold, which is an indication that biomagnification had occurred.



*Figure 4.4: Boiled vegetable Soup Samples*

It can therefore be deduced that there is evidence of biomagnification in both raw and boiled samples of Chinese Cabbage and rape for the treated and control samples.

The findings are concurrent with Corley & Mutiti, (2017), who found similar results and argue that heavy metals such as lead, and iron accumulate, exponentially in boiled Chinese Cabbage soups as growth progresses.

#### 4.3 Guinea Pig Serum laboratory results

Iron concentration in human beings was determined using the pseudo-serum specimen of guinea pig species scientifically called *cavia porcellus*. The specimen were kept separately, those that were fed on effluent water treated rape and Chinese Cabbage (Figure 4.5) from those that were fed on control water treated rape and Chinese Cabbage (Figure 4.6). Blood was then collected for analysis (Figure 4.7) before feeding the guinea pigs with the vegetables treated with either effluent or control treated water and after the feeding was done for 8 weeks.



*Figure 4.5: Guinea pigs fed on effluent water treated rape and Chinese Cabbage.*



*Figure 4.6: Guinea pigs fed on Control Water Treated Rape and Chinese Cabbage.*



*Figure 4.7: Blood Extraction from the Guinea Pig*

Seven blood Specimen denoted as specimen (SP) were extracted from guinea pigs fed on both the effluent water treated vegetables and control water treated vegetables Table 4.4 details the change in iron level in the serum. The guinea pig serum iron levels increased by 23 per cent for specimen 1, specimen 2 reduced by 9 per cent, Specimen 3 increased by 68 per cent, Specimen 4 increased by 14 per cent, specimen 5 increased by 56 per cent, specimen 6 and specimen 7 was not analysed due to extraction of insufficient serum sample size.

Table 4.4: Serum Sample Results (Iron Content -  $\mu\text{g/dL}$ )

<b>Blood Sample Results (<math>\mu\text{g/dL}</math>)</b>								
<b>S No</b>	<b>Date</b>	<b>SP 1</b>	<b>SP 2</b>	<b>SP 3</b>	<b>SP 4</b>	<b>SP 5</b>	<b>SP 6</b>	<b>SP 7</b>
1	02.11.2020	167.5	252.5	204.4	245.8	232.9	127.9	254.7
2	11.12.2020	206.3	205	343	278.9	362.6	Not Analysed ***	Not analysed ***

*Source: Laboratory Results from Pine Ridge Laboratory Services*

\*\*\* Denotes insufficient serum sample extraction

The sample results in micrograms per deciliter were converted to micrograms per litre to uniformly determine whether biomagnification could have occurred in comparison with the baseline ZEMA threshold of 2.0 mg/l. The conversion was done as provided for on the online conversion of  $1\text{mg/l} = 100\ \mu\text{g/dL}$  The results of the conversion are tabulated in table 4.5.

Table 4.5: Serum Sample Results (Iron Content in mg/l)

<b>Blood Sample Results (mg/l)</b>								
<b>S No</b>	<b>Date</b>	<b>SP 1</b>	<b>SP 2</b>	<b>SP 3</b>	<b>SP 4</b>	<b>SP 5</b>	<b>SP 6</b>	<b>SP 7</b>
1	02.11.2020	1.675	2.525	2.044	2.458	2.329	1.279	2.547
2	11.12.2020	2.063	2.05	3.43	2.789	3.626	NA	NA

The changes in serum iron are further shown in figure 4.8, where the  $x$  – axis represented the sample analysed and the  $y$  – axis is the change in iron content in the serum. From the initial analysis of iron level content on the 02.11.2020 revealed that SP 1 increased from 167.5  $\mu\text{g/dL}$  to 206.3  $\mu\text{g/dL}$  an indication that biomagnification did occur by 23 per cent through the absorption mechanism in the gastrointestinal tract.

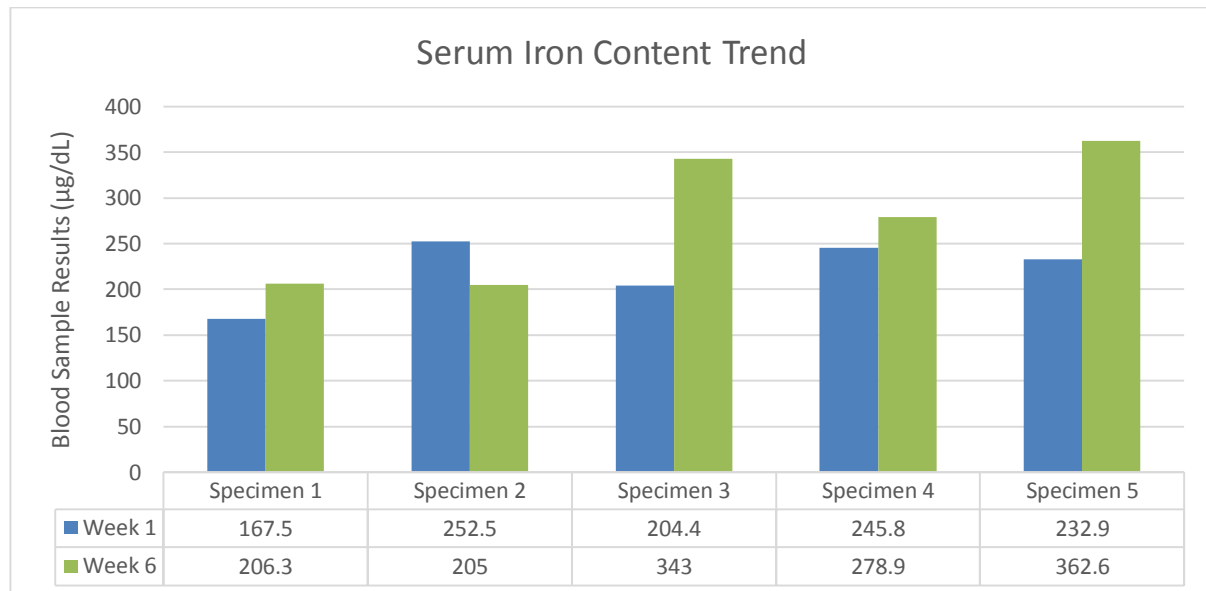


Figure 4.8: Trend Analysis of Change in serum iron

There is evidence of iron bioconcentration in guinea pigs *cavia porcerillus* which can mimic the iron level bioconcentration in human beings. Further, Specimen SP 6 was observed to have developed dry skin which is one of the signs of iron overload, while both specimen 6 and specimen 7 exhibited insufficient blood and could not be drawn for analysis. This can be attributed to anaemia. an indication of iron overload had occurred.

This observed rise iron levels agree with Bruce & Robert, (2006), who also found evidence for liver injuries, fibrosis and cirrhosis when iron levels increase in the range of 8-18  $\mu\text{g/L}$ . This finding is also supported by the findings of Sandros (2021) that there is a link between elevated iron levels and symptoms of fibrosis and hepatic diseases.

The observed rise in iron concentration due to biomagnification may be linked to the possible prevalence of non-communicable illnesses, such as hemochromatosis, in the Nampundwe area. This is so because two of the specimen exhibited signs of haemolytic anaemia, and dry skin disorder and this resulted in the death of the specimen, as found by Kalyani & Srinivas, (2018) and Bruce & Robert, (2006), who also found evidence of liver injuries, fibrosis and cirrhosis when iron levels increase in the range of 8-18  $\mu\text{g/L}$ .

#### 4.4 Responses from Questionnaires

The questionnaires were administered to various clinics within a radius of 35 km from the plant effluent discharge point. A total of 15 questionnaires were administered to the Nampundwe Mine Clinic, which recorded 73.3 per cent response rate, while 20 questionnaires were administered to government clinics, and they gave a response rate of 95 per cent as shown in table 4.6.

Table 4.6. Number of respondents from health facilities

Description	Parameter	
	Nampundwe Mine Clinic	Nampundwe Government Clinics
Institutions		
Questionnaires submitted	15	20
Questionnaires responded to	11	19
% Response rate	73.3%	95%
Total respondents	30	

##### 4.4.1 Prevalence rate of Hepatitis A, B and C

Figure 4.9 indicates that a prevalence rate of hepatitis A, B and C as a likely consequence of iron overload was confirmed by 73 per cent of the respondents. The results were controlled for hepatitis that was hereditary, as indicated in the MoH standard disease reporting form. This finding is also in agreement with empirical findings by Wang et.al. (2006) & Maurizio et al., (1996) that prevalence rate of hepatitis increases with elevated iron levels.

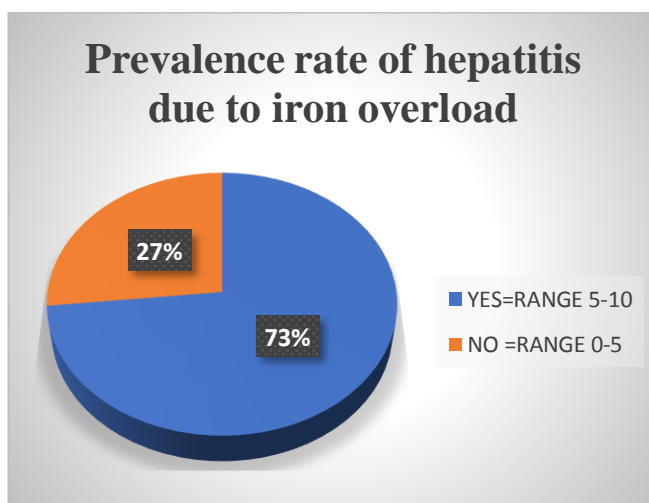


Figure 4.9: Incidences of hepatitis

#### 4.4.2 Prevalence rate of Hepatocellular-Carcinoma

73 percent of the interviewees confirmed that hepatocellular carcinoma could have been caused by above threshold intake of iron, as revealed in Figure 4.10. This shows that there is a causal/effect between elevated iron intake and Hepatocellular carcinoma. This agrees with membrane Van et al., (2020) & Bruce & Robert, (2006), who also found that iron intake has a causal implication of Hepatocellular carcinoma.

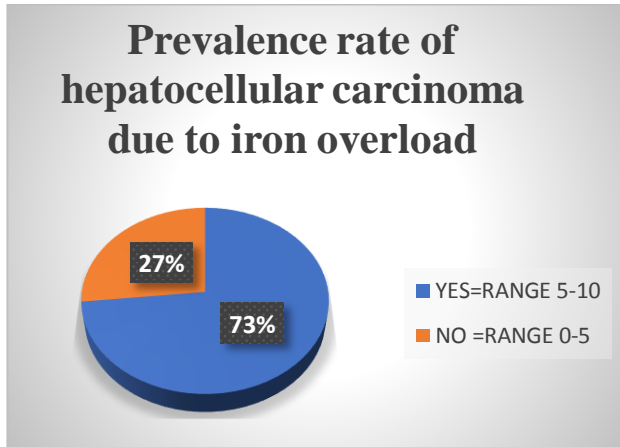


Figure 4.10: Incidences of Hepatocellular carcinoma.

#### 4.4.3 Prevalence rate of Hemochromatosis

The study results revealed that a paucity of 27 per cent of the respondents did not confirm that hemochromatosis was caused by the over-intake of iron, as shown in figure 4.11. However, the research definition of prevalence to be 50 per cent or more reveals that 73 per cent of respondents confirmed a causal link between iron overload and hemochromatosis. The results are in line with the findings by Kalyani & Srinivas (2018), Capell (2004) & Fleming et al., (2001) that hemochromatosis is due to iron overload.

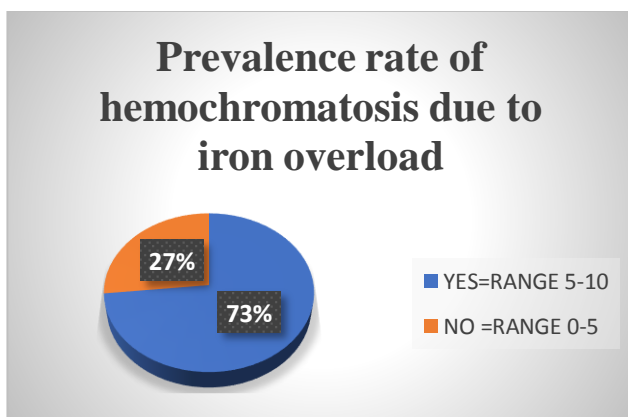


Figure 4.11: Incidences of hemochromatosis

#### 4.4.4 Prevalence rate of Diabetes

The results show that 67 per cent of the interviewees indicated that there is a link between the high intake of iron and diabetes. This linkage revealed in figure 4.12 is supported by findings by Capell (2004) & Gira et al., (2003), who found positive implications of iron overload on diabetic conditions.

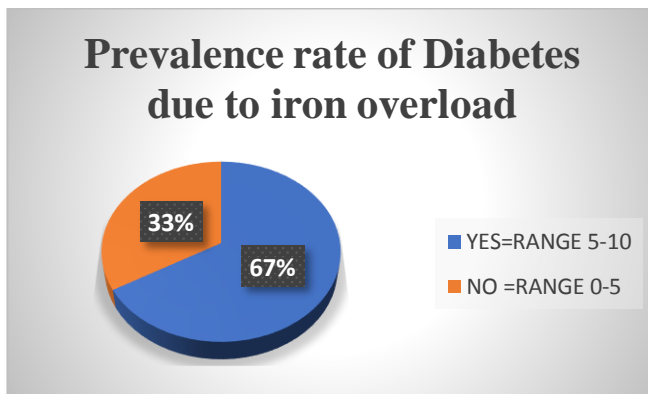


Figure 4.12: Incidences of diabetes

#### 4.4.5 Prevalence Rate of Cancer

The study results, as revealed in figure 4.13, that 66 per cent of the respondents had a strong revealed indication that an overload of iron has an indirect effect on an array of cancers. This is particularly interesting that studies by Van et al., (2020); Kalyani & Srinivas, (2018); Bruce & Robert, (2006) and Maurizio et al., (1996), revealed that hemochromatosis is associated with porphyria cutanea tarda because of iron load which affects quantity and activity of uroporphyrinogen decarboxylase by which uroporphyrinogen accumulates in the skin and present as blisters.

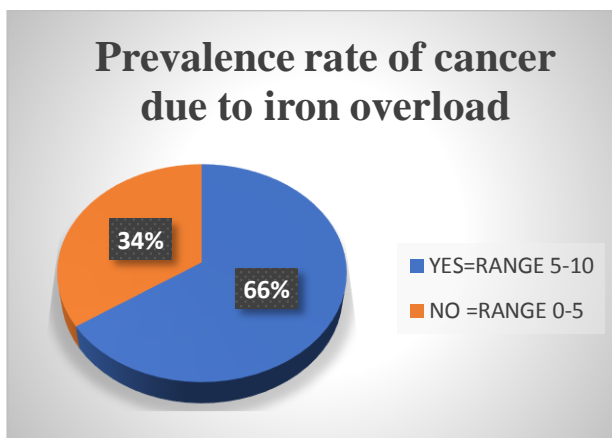


Figure 4.13: Incidences of cancers

#### 4.4.6 Prevalence rate Liver-Damage

Figure 4.14 revealed that 60 per cent of the medical officers directly involved in the health delivery process confirmed that iron-overload has a causal effect on liver damage. This confirms the experimental design results by (Starley et al., 2010; Bruce & Robert, 2006), who found a similar link between elevated iron in guinea pigs and liver damage.

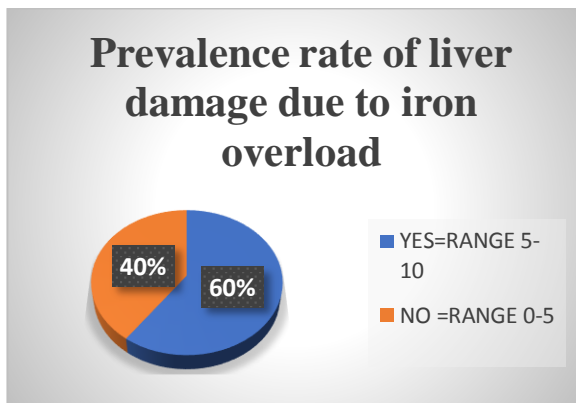


Figure 4.14: Incidences of liver-damage

#### 4.4.7 Prevalence rate skin-disease

Results, as shown in figure 4.15, indicate that approximately 57 per cent of the clinicians indicated that iron overload causes dry skin disease. This agrees with membrane Van et al., (2020)) Bruce & Robert, (2006) and Maurizio et al., (1996), who also argue that elevated iron levels cause dry skin disease.

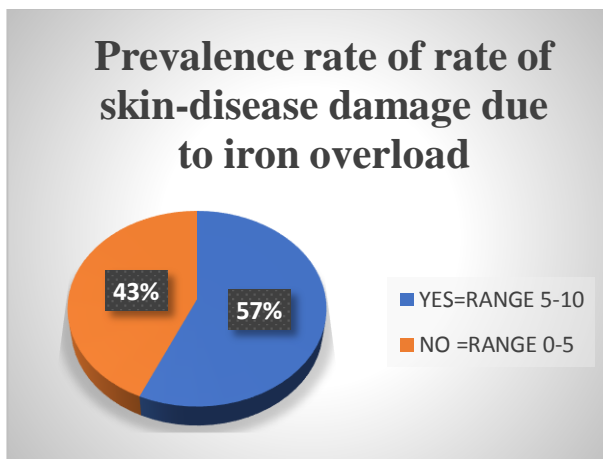


Figure 4.15: Incidences of skin disease

#### 4.5 Summary

The study revealed that mine effluent was within the ZEMA statutory requirement. The levels of iron in vegetables grown were established and indicated that, indeed, biomagnification took place from the germination to consumption of fully-grown vegetable shoots in Chinese Cabbage and rape. Further, fresh shoots from rape and Chinese Cabbage revealed that biomagnification had occurred. Furthermore, the guinea pig average iron serum in milligrams per litre exhibited bio-concentration of 23 per cent, 68 per cent, 13.3 per cent and 55.7 per cent for Specimen 1, Specimen 3, Specimen 4 and Specimen 5 in that order, while Specimen 6 and Specimen 7 exhibited signs of elevated iron levels of dry skin disorder. Finally, close to 58.8 per cent of the respondents confirmed that iron overload causes dry skin diseases, liver injuries, cirrhosis, hepatocellular carcinoma, hepatitis, hemochromatosis, and diabetes. Therefore, there is a causal link between the ascertained biomagnification of iron across the food chain and the highly perceived iron overload-related diseases from the survey results.

## CHAPTER FIVE

### CONCLUSION AND RECOMMENDATION

#### 5.0 Conclusion

It is evident from the results obtained that Nampundwe Mine effluent does meet the ZEMA statutory requirement of 2mg/l. However, the effluent is not compliant with the WHO/FAO limit for irrigation water of 0.5mg/l. This entails that the water used for irrigation by the vegetable farmers is not fit for use in vegetable irrigation activities. Further, the soil sample analysis within the Mines 35 km radius revealed that the results are within WHO/FAO statutory limit. However, biomagnification occurred in raw rape and Chinese Cabbage shoots. The boiled Chinese Cabbage and rape soups combined revealed biomagnification as well. The biomagnification was revealed in the shoots of fully developed rape and Chinese Cabbage. The average iron results recorded for vegetables in this study were all lower than the WHO/FAO permissible value of 425 mg/l yet still did indicate that biomagnification had occurred due to the increase in iron levels in comparison with the baseline.

Furthermore, guinea pig serum iron for the four samples confirmed a combined average of 40 per cent biomagnification in guinea pigs' serum. Equally, two of the guinea pigs exhibited signs of elevated levels of iron in serum which was anaemia for both the specimen and dry skin disorder for one of the specimen.

The study baseline evidence that the mine effluent, which was not compliant with WHO/FAO irrigation water standards, contributed to the biomagnification of iron in the vegetable. Moreover, guinea pigs' laboratory results link iron overload to possible health implications such as hepatitis, hemochromatosis, dry skin disease and haemolytic, which was because of the vegetables consumed. This, therefore, closes the gap earlier identified on the source of iron that links elevated iron levels to its associated health impacts on humans.

However, the study did not link directly using exit poll interviews on the prevalence of iron associated cases using clinical records. This was due to Covid-19 measures employed by the health facilities. Therefore, the study utilized an online perception study on the causal link between elevated iron levels and hepatitis, hemochromatosis, dry skin disease and haemolytic anaemia administered to the clinicians in health facilities in Nampundwe that revealed around 58.8 per cent confirming that elevated iron level causes the aforementioned disease conditions.

## 5.1 Areas for further research

Due to budgetary constraints and limited financial resources, the study used only seven guinea pigs as the sample size, which could have been sixty. The timeframe for this research could have been done on a large scale and with multiple data collection of serum, soil, mine effluent and vegetable samples for 24 months to unearth possible unobserved heterogeneities.

The determination of the iron levels and possible direct causal link of hepatitis, hemochromatosis, dry skin disease and haemolytic anaemia in patients in Nampundwe should be clinically investigated at their first reporting stage at the health facilities to rule out other possible causes.

## 5.2 Recommendations

The research findings may be useful to health policy designers and implementers, as the evidence adduced can be a serious baseline study for further research for policy. For instance, the research gaps, such as actual clinical laboratory examinations on observed patients with hepatitis, hemochromatosis, dry skin disease and haemolytic anaemia, should be carried out. In the medium term, the regulator should consider a more proactive approach in extending inspections and conducting independent water analysis for heavy metal content in communities utilizing mine effluent for irrigation to determine and mitigate biomagnification. More importantly, the prohibiting use of mine effluent and land in mining areas for agricultural purposes as it is evident that biomagnification of heavy metals such as iron occurs in leafy vegetables. This finding can be used as an opportunity for mining houses to clean up areas that are contaminated with heavy metals by using leafy vegetables that are hyperaccumulators for bioremediation. Enforcement by ZEMA could, among other things, demand for mining houses to implement available best practise at source to ensure compliance with requirements on Mine effluent. Furthermore, the regulator could consider extending monitoring soil specifications for agriculture land use requirement.

Despite KCM upholding best practices by dealing with heavy metal content, such as iron at source, as evidenced in the results of effluent which remained within 2mg/l ZEMA threshold, agricultural use of mine effluent, does ignite biomagnification and health concerns as revealed in the study.

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- <https://www.convertmeasurementunits.com/convert+Microgram+per+deciliter+to+Milligram+per+liter.php>

## APPENDICES

### 6.0 Laboratory Report-Clinical Chemistry: Pine Ridge Laboratory Services



# PINE RIDGE LABORATORY SERVICES



Molecular Level Precision

**Plot No: 916 Water works,  
Libala South,  
LUSAKA  
ZAMBIA.**

**Contact: +260-974 054 620,  
WhatsApp: 962-238719**

**Email: pineridgelab2020@gmail.com**

**Website: pine-ridge-laboratory-services-vetlab.business.site**

### LABORATORY REPORT

LAB ID: PR06

#### CLINICAL CHEMISTRY

**Owner/Animal Name:** M/S Mwitwa. K/  
**Species:** Guinea pig (*Cavia porcellus*)  
**Specimen:** Serum (red-top vacutainer)  
**Exam required:** Total Serum Iron

**Date Submitted:** 2<sup>nd</sup> November, 2020  
**Person/vet Submitted:** M/S Mwitwa. K  
**Customer ID:** MK01  
**Contact:** 0977188006

**History:** For research

Analyte (SI UNITS)	RESULTS							
	LAB No. GL	13421	13422	13423	13424	13425	13426	13427
	Sample ID:	1	2	3	4	5	6	7
<b>Total Serum Iron (µg/dL)</b>		167.5	252.5	127.9	204.4	254.7	245.8	232.9
<b>Reference Range (µg/dL)</b>	Yet to be established at lab level- ( <i>Cavia porcellus</i> )							

For all your laboratory diagnostic tests (Biochemistry, Haematology, Microbiology, endocrinology, pathology), Water quality assessments, etc.

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**TEST REMARKS**

Kindly correlate clinically.

**Note:**

**Higher-than-normal iron level may be a sign of:**

- Too much iron in the body (hemochromatosis)
- Anemia due to red blood cells being destroyed too quickly (hemolytic anemia)
- Liver tissue death
- Inflammation of the liver (hepatitis)
- Iron poisoning
- Frequent blood transfusions

**Lower-than-normal level may be a sign of:**

- Long-term digestive tract bleeding
- Heavy menstrual bleeding, in some species.
- Intestinal conditions that cause poor absorption of iron
- Not enough iron in the diet
- Pregnancy

**ANALYST:** Emmanuel. S. Kabwali  
(MSc. OHEA-cand, BSc. Biomed)

**DATE REPORTED:** 05/11/2020

For all your laboratory diagnostic tests (Biochemistry, Haematology, Microbiology, endocrinology, pathology), Water quality assessments, etc.



## 6.1 Laboratory Report-Clinical Chemistry: Pine Ridge Laboratory Services



# PINE RIDGE LABORATORY SERVICES



Molecular Level Precision

Plot No: 916 Water works,  
Libala South,  
LUSAKA  
ZAMBIA.

Contact: +260-974 054 620,  
WhatsApp: 962-238719  
Email: [pineridgelab2020@gmail.com](mailto:pineridgelab2020@gmail.com)  
Website: [pine-ridge-laboratory-services-vetlab.business.site](http://pine-ridge-laboratory-services-vetlab.business.site)

### LABORATORY REPORT

LAB ID: PR99

### CLINICAL CHEMISTRY

**Owner/Animal Name:** M/S Mwitwa. K/  
**Species:** Guinea pig (*Cavia porcellus*)  
**Specimen:** Serum (red-top vacutainer)  
**Exam required:** Total Serum Iron

**Date Submitted:** 11<sup>th</sup> December, 2020  
**Person/vet Submitted:** M/S Mwitwa. K  
**Customer ID:** MK01  
**Contact:** 0977188006

**History:** For research

Analyte (SI UNITS)	RESULTS							
	LAB No. GL	14746	14747	14748	14749	14750	14751	14752
	Sample ID:	1	2	3	4	5	6	7
Total Serum Iron (µg/dL)		206.3	205.0	Insufficient sample	343.0	Insufficient sample	278.9	362.6
Reference Range (µg/dL)	Yet to be established at lab level- ( <i>Cavia porcellus</i> )							

For all your laboratory diagnostic tests (Biochemistry, Haematology, Microbiology, endocrinology, pathology), Water quality assessments, etc.

1

**COMMENTS**

Samples # 3 and #5 were quite insufficient, repeat testing is advised with at least 3mL serum

**TEST REMARKS**

Kindly correlate clinically.

**Note:**

**Higher-than-normal iron level may be a sign of:**

- Too much iron in the body (hemochromatosis)
- Anemia due to red blood cells being destroyed too quickly (hemolytic anemia)
- Liver tissue death
- Inflammation of the liver (hepatitis)
- Iron poisoning
- Frequent blood transfusions

**Lower-than-normal level may be a sign of:**

- Long-term digestive tract bleeding
- Heavy menstrual bleeding, in some species.
- Intestinal conditions that cause poor absorption of iron
- Not enough iron in the diet
- Pregnancy

**ANALYST:**

Emmanuel. S. Kabwali  
(MSc. OHAE-cand, BSc. Biomed)

**DATE REPORTED:**

16/12/2020

For all your laboratory diagnostic tests (Biochemistry, Haematology, Microbiology, endocrinology, pathology), Water quality assessments, etc.



## 6.2 Laboratory Report: Geo-Chemical Analytical Laboratory



**THE UNIVERSITY OF ZAMBIA**  
SCHOOL OF MINES  
GEO-CHEMICAL ANALYTICAL LABORATORY

BOX 32379  
LUSAKA  
ZAMBIA

TEL/FAX: +260-1-294086/290165

ATTENTION : MWITWA KAMBAFWILE  
DATE SUBMITTED : 21/12/20  
DATE ANALYSED : 31/12/20  
DATE REPORTED : 29/01/21

### LABORATORY REPORT

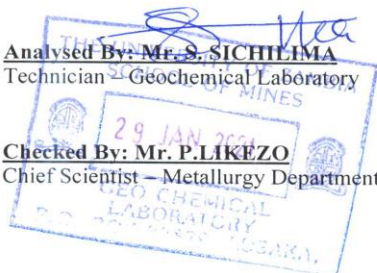
Sample Id	Method	Iron mg/L
<b>BOILED SAMPLES</b>		
CONTROL WATER TREATED CHAINESE CABBAGE	Atomic Absorption Spectrophotometry	18.24
EFFLUENT WATER TREATED RAPE	Atomic Absorption Spectrophotometry	20.10
EFFLUENT WATER TREATED CHAINESE CABBAGE	Atomic Absorption Spectrophotometry	20.94
CONTROL WATER TREATED RAPE	Atomic Absorption Spectrophotometry	19.49
<b>CHEMICAL ANALYSIS</b>		
EFFLUENT WATER TREATED CHAINESE CABBAGE	Atomic Absorption Spectrophotometry	26.31
EFFLUENT WATER TREATED RAPE	Atomic Absorption Spectrophotometry	24.67
CONTROL WATER TREATED CHAINESE CABBAGE(CWT-C)	Atomic Absorption Spectrophotometry	27.13
CONTROL WATER TREATED RAPE	Atomic Absorption Spectrophotometry	26.31

**NB.**

- i. *This laboratory report should not in any way be used as a certificate for goods not sampled and sealed by our officers.*
- ii. *The results reported above pertain only to the sample submitted for analysis and not necessarily to any other samples of similar nature*
- iii. *If this document is transmitted electronically, it will only be valid if it is supported by the original document.*

*[Signature]*  
**Analysed By: Mr. S. SICHILIMA**  
Technician – Geochemical Laboratory

**Checked By: Mr. P. LIKEZO**  
Chief Scientist – Metallurgy Department



### 6.3 Laboratory Report: Department of Soil Science Services Laboratory

The University of Zambia  
 Department of Soil Science  
 Service Laboratory



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

ATTN: Ms. Mwitwa Kambafwile  
 Lusaka

#### Plant Tissue Analysis Results - December, 2022


Lab	Sample	Sample					Fe
no.	Id	number					Ash/ $\text{HNO}_3$ mg/kg
20221143	Control water treated chinese cabbage	3					321.00
20221141	Control water treated rape	1					138.00
20221142	Effluent water treated chinese cabbage	2					454.00
20221144	Effluent water treated rape	4					328.00

G. Musukwa  
 Chief Technician  
 09/01/2023



Head,  
 Dept of Soil Science

## 6.4 School of Engineering, Environmental Engineering Laboratory Results

  
 SCHOOL OF ENGINEERING  
 CIVIL ENGINEERING DEPARTMENT  
 ENVIRONMENTAL ENGINEERING LABORATORY  
 P.O. Box 32379, Lusaka

**PHYSICAL/CHEMICAL EXAMINATION OF WATER**

Attn : Mwitwa Kambafwile  
 Nampundwe Mine  
 Sampled by : Client  
 Report date : 30.12.2020

**Laboratory Results (WATER)**

Sample ID	Sampling Date	Iron (mg/l)
KCM UG Effluent	30.09.2020	0.90
KCM Plant Effluent	30.09.2020	<0.01
KCM TD Overflow	30.09.2020	1.30
KCM UG Effluent	01.10.2020	<0.01
KCM TD Overflow	01.10.2020	<0.01
KCM TD Overflow	01.10.2020	0.80
KCM Plant Effluent	01.10.2020	0.70
KCM UG Effluent	15.10.2020	0.42
KCM Plant Effluent	15.10.2020	<0.01
KCM TD Overflow	15.10.2020	<0.01
Mwembeshi Garden	23.10.2020	


**Laboratory Results (SOIL)**

Sample ID	Sampling Date	Iron (ppm)
KCM Soil Sample 1	01.10.2020	1.10
KCM Soil Sample 2	01.10.2020	2.21
Mwembeshi Garden Soil	23.10.2020	0.94

UNIVERSITY OF ZAMBIA  
 SCHOOL OF ENGINEERING  
 30 DEC 2020  
 DEPT. OF CIVIL ENGINEERING  
 P.O. BOX 32379 LUSAKA

Tested by: E. Mutati  
 Lab. Technician

Checked & Approved by: Joshua Liyanga  
 Lab. Manager / Co-ordinator

  
 SCHOOL OF ENGINEERING  
 CIVIL ENGINEERING DEPARTMENT  
 ENVIRONMENTAL ENGINEERING LABORATORY  
 P.O. Box 32379, Lusaka

**PHYSICAL/CHEMICAL EXAMINATION OF WATER**

Attn : Mwitwa Kambafwile  
 KCM - Nampundwe  
 Sampled by : Client  
 Report date : 03.11.2022

**Laboratory Results**

Parameter	Water Sample	Water Sample	WHO Guideline (Maximum Permissible value for drinking water)
Sampling Date	12.09.2022	19.09.2022	
pH	6.80	6.87	
Iron (mg/l)	<0.01	<0.01	0.30

Tests carried out in conformity with "Standard Methods for the Examination of water and Wastewater APHA, 1998".

Tested by: E. Mutati  
 Lab. Technician

Checked & Approved by: Joshua Liyanga  
 Lab. Manager / Co-ordinator

UNIVERSITY OF ZAMBIA  
 SCHOOL OF ENGINEERING  
 03 NOV 2022  
 CIVIL AND ENVIRONMENTAL  
 ENGINEERING LABORATORY  
 P.O. BOX 32379 LUSAKA

**PHYSICAL/CHEMICAL EXAMINATION OF WATER**

Attn : Mwtwa Kambafwile  
 KCM - Nampundwe  
 Sampled by : Client  
 Report date : 07.11.2022

Parameter	Laboratory Results			WHO Guideline (Maximum Permissible value for drinking water)
	Plant Effluent	Plant Effluent	Plant Effluent	
Sampling Date	12.09.2022	19.09.2022	26.09.2022	
pH	7.84	7.81	7.84	6.5 - 8.5
Iron (mg/l)	0.26	0.32	0.29	0.30

Tests carried out in conformity with "Standard Methods for the Examination of water and Wastewater APHA, 1998".

Tested by: E. Mutati  
 Lab. Technician

Checked & Approved by: Joshua L. Mwanza  
 Lab. Manager /Co-ordinator



**PHYSICAL/CHEMICAL EXAMINATION OF WATER**

Attn : Mwtwa Kambafwile  
 KCM - Nampundwe  
 Sampled by : Client  
 Report date : 07.11.2022

Parameter	Laboratory Results			WHO Guideline (Maximum Permissible value for drinking water)
	Tailing Dam	Tailing Dam	Tailing Dam	
Sampling Date	03.10.2022	10.10.2022	17.10.2022	
pH	7.93	7.86	7.92	6.5 - 8.5
Iron (mg/l)	<0.01	<0.01	<0.01	0.30

Tests carried out in conformity with "Standard Methods for the Examination of water and Wastewater APHA, 1998".

Tested by: E. Mutati  
 Lab. Technician

Checked & Approved by: Joshua L. Mwanza  
 Lab. Manager /Co-ordinator



**PHYSICAL/CHEMICAL EXAMINATION OF WATER**

Attn : Mwtwa Kambafwile  
 KCM - Nampundwe  
 Sampled by : Client  
 Report date : 07.11.2022

Parameter	Laboratory Results			WHO Guideline (Maximum Permissible value for drinking water)
	Tailing Dam	Tailing Dam	Tailing Dam	
Sampling Date	03.10.2022	10.10.2022	17.10.2022	
pH	7.93	7.86	7.92	6.5 - 8.5
Iron (mg/l)	<0.01	<0.01	<0.01	0.30

Tests carried out in conformity with "Standard Methods for the Examination of water and Wastewater APHA, 1998".

Tested by: E. Mutati  
 Lab. Technician

Checked & Approved by: Joshua L. Mwanza  
 Lab. Manager /Co-ordinator



**PHYSICAL/CHEMICAL EXAMINATION OF WATER**

Attn : Mwtwa Kambafwile  
 KCM - Nampundwe  
 Sampled by : Client  
 Report date : 07.11.2022

Parameter	Laboratory Results			WHO Guideline (Maximum Permissible value for drinking water)
	Plant Effluent	Plant Effluent	Plant Effluent	
Sampling Date	03.10.2022	10.10.2022	17.10.2022	
pH	7.82	7.75	7.79	6.5 - 8.5
Iron (mg/l)	0.30	0.26	0.28	0.30

Tests carried out in conformity with "Standard Methods for the Examination of water and Wastewater APHA, 1998".

Tested by: E. Mutati  
 Lab. Technician

Checked & Approved by: Joshua L. Mwanza  
 Lab. Manager /Co-ordinator



## 6.5 Questionnaire



### THE UNIVERSITY OF ZAMBIA SCHOOL OF MINES

#### THE BIO-MAGNIFICATION IMPACT OF DISCHARGING ELEVATED LEVELS OF IRON EFFLUENT ON HUMANS: CASE OF NAMPUNDWE

##### University of Zambia: Awareness and Perception Survey of Iron Overload

I am conducting this research as a minimum requirement for the award of MSc in Sustainable Mineral Resource Development in the School of Mines. With the support from the School of Mines Dean and the Directorate of Research and Graduate Studies am conducting a survey on the “The Bio-Magnification Impact of Discharging Elevated Levels of Iron Effluent on Humans: Case of Nampundwe.” The study seek to empirically examine the impact of above threshold and statutory iron content in waste water on health status in Nampundwe. Elevated iron status may be associated with increased risk of a number of health conditions such as coronary heart disease (CHD), cancer, elevated blood sugar, liver damage and type 2 diabetes just to mention a few.

You have been selected to participate as health practitioner. The Information you provide in this survey is **STRICTLY CONFIDENTIAL** and will only be used for analysis.

**PLEASE DO NOT WRITE YOUR NAME ON THIS QUESTIONNAIRE.**

Your participation is greatly appreciated.

Kambafwile Mwitwa

Principal Investigator

Questionnaire No:
-------------------

Post-graduate Student

University of Zambia School of Mines

Researcher:

--

Mobile: +260977188006

Date:

SECTION A: BACKGROUND INFORMATION

Question	Select ONE Best Answer	Circle the Best Answer							
1. Health facility name									
2. Level of health provision of the facility	1. Tertiary hospital 2. First Level hospital 3. Clinic 4. Health Post	1	2	3	4				
3. What is the category of your medical profession?	1. Medical Doctor 2. Clinical Officer 3. Nurse 4. Laboratory Technologist 5. Other Specify.....	1	2	3	4	5			
4. What are your number of years of experience?	1. Below one year 2. 1-4 years 3. 5-10 years 4. Above 10 years	1	2	3	4				

5.What health care services does the facility offer?	1. Outpatient care 2. Outpatient and Inpatient 3. Laboratory services 4. Other care.....	1	2	3	4														
6.Does your facility have a fully functional laboratory?	1. Yes 2. No	1	2																
7.If Yes to above, how many personnel directly work in the laboratory?	1. 1-2 2. 3-5 3. 6-10 4. Above 10	1	2	3	4														
8.Of the following noncommunicable disease investigated in the past 1 year, how prevalent are the following diseases: 1=lowest 10=highest	A. PREVALENCE RATE	1	2	3	4	5	6	7	8	9	10								
	1.Cancers																		
	2. Acute hepatitis A																		
	3. Acute hepatitis B																		
	4. Acute hepatitis C																		
	3.Liver damage or disease																		
	4.Hepatocellular Carcinoma																		
	5. Diabetes mellitus type 1																		
6. Diabetes mellitus type 2																			

	6.Hemochromatosis													
	7. Acquired haemolytic anaemia -unspecified													
	8.Dry skin disorder													

SECTION B: EPIDEMIOLOGICAL TRENDS OF CANCER DIABETES BLOOD SUGAR AND HEART-DISEASE

The following table lists a number of diseases and diagnostic tests. Please note which tests are performed in your laboratory or are referred to other laboratories for investigations. For each disease, note whether or not you test any of the named specimens by any of the listed tests. Please give the approximate number/month of each test you perform.

A.Disease	B.Specimen Type	C.Assay Performed	D.Yes=1	E. No=2	F. Do you think the this condition is caused by over-intake of iron	
					YES=1	NO=2
1. Acute Hepatitis A, B and C	B1. Serum	B.1.1 Anti-HAV IgM				
		B.1.2 Anti-HBc IgM				
		B.1.3 Anti-Hbs Ag				
		B.1.4 Anti-HCV IgM				
		B.1.5 Anti-HEV IgG				
2..Hepatocellular Carcinoma	B2.blood sample	B2.1 alkaline phosphatase (ALP)				
3.Hemochromatosis	B3. blood sample B4 Serum Iron	B.3.4.1Total iron-binding capacity (TIBC)				
		B.3.4.2Transferrin saturation				
		B.3.4.3 Ferritin				

		B.3.4.4 Transferrin saturation				
4. Diabetes mellitus type 1,2 and 3	B4 Urine	B.4.1 ALT, which stands for alanine transaminase,				
	B5 blood sample	B5.1 Glucose level				
5.Cancers	B6 Complete blood count (CBC) B7 Tumor marker tests B8 Blood protein testing	B6.1 marrow biopsy				
		B7.1(PSA) for prostate				
		B7.2 (CA 125) for ovarian cancer				
		B7.3 alpha-fetoprotein (AFP) for liver cancer				
		B8.1 (electrophoresis) to examine various proteins				
6.Liver damage or disease	B9 Blood	B.9.1 liver panel				
		B.9.2 MRI				
		B.9.3Liver biopsy				
7.Dry skin disease	B10 Blood	B.10.1 Cardiac Troponins				
		B.10.2 Lipids				
		B.10.3 Natriuretic peptides				

END OF SURVEY THANK YOU FOR YOUR TIME.