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ANALYSIS OF BEETROOT BULBS (Beta *Vulgaris*) FROM SELECTED GEOGRAPHICAL REGIONS IN KENYA: CONTENTS OF ESSENTIAL NUTRITIONAL ELEMENTS

By

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DECLARATION

This thesis is my original work and has not been presented for a degree or any other award to any other university.

Grace Ndunge, S56/5891/2017

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LIST OF ABBREVIATIONS

ADMCA: Analog to digital multichannel analyser

ANOVA: Analysis of Variance

d.w: Dry weight

EDXRF: Energy Dispersive X-ray Fluorescence

FAO: Food and Agriculture Organization

GPS: Global Positioning System

IAEA: International Atomic Energy Agency

KNBS: Kenya National Bureau of Statistics

LEO: Light Element Optimization

P/B: Peak- to- background ratio

ppm: Part per million

s.d: Standard deviation

SDD: Silicon Drift Detector

SRM: Standard Reference Material

t_{calc}: Student's t-test calculated

t_{tab}: Student's t-test tabulated

USD: United States Dollar

VFN: Vegetable Fact Network

WDXRF: Wavelength Dispersive X-ray Fluorescence

WHO: World Health Organization

ABSTRACT

Beetroot is the taproot part of a Beet plant, which is among one of the several cultivated varieties of Beta *Vulgaris*. It has medicinal value especially for cardiac and cancer treatment. It is also a potential antioxidant and blood cell count booster. Other uses include; source of nutrients, sugars and as a livestock feed. Beetroot is not a popular vegetable among most Kenyans, but is occasionally used as a blend in juices and salads. In this study, thirty-four (34) beetroot bulb (Crimson globe variety) samples were collected, from selected areas in Karatina, Naivasha, Gilgil, Kisumu and Joska-Machakos. The samples were analysed for potassium, calcium, iron, manganese and zinc using Energy Dispersive X-ray Fluorescence (EDXRF). EDXRF method of analysis is used worldwide for elemental analysis since it is a fast, easy, reproducible and non-destructive. Prior to analysis, the bulbs were dried in the oven for 56 hours at 60^{o}C to constant weight, ground, and sieved using a 75-micron sieve to homogenize them. Approximately 0.5-0.6 g of the powder sample was prepared into thin pellets, with a diameter of 25mm and analysed for essential elements using EDXRF. The results of the concentration of the essential elements had a range of: 11000 mg kg^{-1} to 50000 mg kg^{-1} for potassium, 94.0 mg kg^{-1} to 4500 mg kg^{-1} for calcium, 36.0 mg kg^{-1} to 230 mg kg^{-1} for manganese, 12.0 mg kg^{-1} to 770 mg kg^{-1} for iron and 14.0 mg kg^{-1} to 680 mg kg^{-1} for zinc. In general, the trend in the concentration levels of the essential elements was K > Ca > Fe > Mn > Zn. Karatina beetroot samples recorded the highest concentrations for all the elements of interest in the study. The results of Analysis of variance (ANOVA) for all the five sampled regions show that there is a significant difference in the Ca, Fe and Mn concentration levels. In general, the concentration of essential elements in beetroots are comparable to those found in most commonly consumed tubers and leafy vegetables. Beetroots were found to contain sufficient amounts of the essential elements, therefore, are a suitable vegetable source of essential elements and can be used as an immune system booster. In addition, the study recommends for further research to determine the optimum growing conditions and age of harvesting the beetroot bulbs in terms of bio-accumulation of essential elements. This study is supportive to the Government's efforts of improving health care and in the fight against "*hidden hunger-malnutrition*" in the country.

CHAPTER ONE: INTRODUCTION

1.1 General Overview

Beetroot bulb is the taproot part of a beet plant. The plant is among one of the several cultivated varieties of Beta *Vulgaris*, which is good for their edible taproots. Beetroots are used as food colorants, enhancing colour in food products such as jams and jellies, tomato paste, desserts, ice cream, candy and cereals, among others (Grubben & Denton, 2004).

Beetroots can be used either alone, mixed to make a vegetable salad, cold as a condiment or warm incorporating butter as a delicacy. They can be consumed raw, boiled or roasted. For salads, beetroot bulbs are peeled and shredded raw then mixed with other vegetables or eaten alone (Grubben & Denton, 2004).

In addition, beetroot is sold commercially either as boiled, processed beetroot or pickles (VFN, 2017). In different cultures such as India, beetroots are chopped, cooked and spiced, and is a typical dish in the region, while in Eastern Europe, beetroots are used to make soup-borscht (Winner, 1993).

Various health benefits associated with beetroots include; reduce blood pressure in hypertensive patients, and in reducing instances of cardiovascular diseases (George *et al*., 2010). Sports endurance performance has also been found to improve with dietary nitrate supplementation that is found in beetroots (Zafeiridis, 2014)

Research indicates that beetroot contain betanin that can function to reduce the concentration of homocysteine. The latter is a homolog of the naturally occurring amino acid cysteine (Lamberth, 2016). Homocysteine in high levels can cause injure to blood vessels and cause cardiovascular diseases. However, this theory is arguable since it not yet affirmed if homocysteine itself is harmful or it is a signal of increased risk for cardiovascular (Cacciapuoti, 2012). Beetroots also contain inorganic nitrate that is converted to nitric oxide and is important for physiological intercellular messenger and cytotoxic effector molecule (Moat, 2008).

Additionally, 100 g of beetroots contain carbohydrates: 6.8 g of sugar, approximately 2.8 g dietary fibre and a source of fructose which are short-chained carbohydrates categorized as fermentable oligo-, di-, mono-saccharides and polyols (FODMAPS) (Moat, 2008). These components go through the tricarboxylic acid cycle and result in energy production for the body. Also approximately 1.6 g of protein is contained in the beetroot, whose function is production of energy, enzymes and protein based structures.

Beetroots have high level of fibres, essential vitamins and minerals. The specific elements from vitamins and minerals are folate B9 that is significant for cellular function and normal tissue growth. It is highly beneficial for pregnant women (Ferguson *et al*., 2015). Manganese serves as an essential trace element that is a good anti-oxidant (Institute of Medicine , US, 2001). Potassium functions as a reducing agent for blood pressure levels as well as has a positive impact on cardiovascular health (Carter, 2018). Iron is also another essential mineral that is required for transportation of oxygen in the red blood cells. Vitamin C serves as an antioxidant that is needed for immune function as well as skin health (Moat, 2008). These numerous health factors depict the importance of beetroot consumption in Kenya. Therefore, this study aims to analyse beetroots for essential elements content from different geographical regions in Kenya.

1.2 Statement of the Problem

Malnutrition is a major challenge in developing countries like Kenya. Consequently, there is need for crops with high nutritional content to be incorporated in the diet. In general, common sources of minerals and essential elements are vegetables such as kale, spinach, garlic, cabbage, green beans, carrots, whole wheat and lettuce among many; contain manganese, calcium, iron, potassium and vitamins.

Many households in Kenya lack enough nutritious food due to poverty, therefore there is need to provide an affordable source of nutrition to the population. Available literature shows that nutritious elements are present in beetroots. However, in Kenya, beetroot is not a popular vegetable and yet it has the potential to be a good natural source of minerals and essential elements. While it is possible that the beetroot taste and its deep colour can lead to a bias against it, the major problem is that many people are not aware of its potential nutritional value.

It is important to have information on the nutritional value of beetroot bulbs in Kenya since there are minimal studies conducted on them. Therefore, it is crucial to carry out a study on beetroot bulbs from different geographical regions in Kenya to assess their nutritional level on essential elements.

1.3 Objectives

1.3.1 General Objective

Assess the nutritional content of beetroots bulbs grown in selected different geographical regions of Kenya

1.3.2 Specific Objectives

1) To evaluate the mineral nutritional content; potassium (K), calcium (Ca), Iron (Fe), manganese (Mn) and zinc (Zn) in beetroot varieties;
2) To determine essential mineral variation in beetroot bulbs (Beta *Vulgaris*) from Karatina, Gilgil and Naivasha farms and Joska-Machakos and Kisumu markets;

1.4 Justification and limitation of the study

Beetroot consumption is slowly gaining popularity in Kenya, both as a cash crop and for medicinal purposes. However, its use for nutritional and health benefits to support farming is not well documented, because there are few studies that exist locally.

In doing this research, I have specifically focused on one beetroot bulb variety (*Crimson globe*) and analysed for the following essential elements; potassium, calcium, manganese, iron and zinc using Energy Dispersive X-Ray fluorescence. The leaves and stems of the beetroot plant were not studied, as well as the soil on which the bulbs grew on.

This study supports the Government's efforts of improving health care and in the fight for "*hidden hunger-malnutrition*" in the country. Kenya's Vision 2030 aims at transforming the country into a middle-income economy, which will be achieved by having a healthy nation.

CHAPTER TWO: LITERATURE REVIEW

2.1 Historical background of Beetroots Farming and Uses

The taproot part of the beet plant is known as table beet, red or golden beet or commonly as beetroot (VFN, 2017). Archaeological proofs indicate that human beings have used beetroots in ancient times. The use of beets dates back in the third millennium BC as evidenced by Neolithic site of Aartswoud in the Netherlands and in the Saqqara pyramid at Thebes, Egypt (Newton, 2005). Assyrian texts indicate that beetroots may have been growing in the Hanging Gardens of Babylon in 800 BC. However, the existence of the garden is unclear. Further research indicates that Mesopotamians knew about beetroots around 300 BC where they ate the leaves of the plant only, and offered the roots to Apollo, the sun god, in the temple of Delphi. Moreover, Mesopotamians believed the value of beetroots to be equivalent to silver in weight (VFN, 2017).

The Talmud recommended eating beetroots to prolong life. The medicine men also used its leaves for wounds dressing. Romans used raw beetroots for curing illnesses such as fever. They also used it as aperients while the Roman connoisseur, Apicius, used it as food. In his book, 'The Art of Cooking', Apicius used it in various recipes; to make broths, salads with mustard, oil and vinegar (VFN, 2017).

Most of these various uses, were for the old species of beetroots; these were long and thin, resembling a carrot. Modern beetroot types appeared in the sixteenth and seventeenth century in Europe. However, it took many centuries for Central and Eastern Europe to incorporate beetroots in their cuisines. For instance, in Victorian times, beetroot was used as a sweetener in foods such as desserts (VFN, 2017). It also gave colour to various colourless food diets. Historically, natural preparation and conservation of vegetables was possible in the age of industrialization. During this time, beetroot became more available to people. However, after the Second World War, beetroots were pickled to enable longer storage duration. This was due to the shortage of food, which coincidentally made the earlier version of beetroot to disappear. Currently, the common type of beetroot is round and deep red. Other kinds are yellow, white and even white-red as shown in Figure 2.1 (Boudry *et al*., 1993).

Figure 2.1:Types of beetroots (Boudry *et al*., 1993)

Borscht is a beetroot special soup that has its origin in Ukraine. It is a common delicacy in Eastern and Central Europe. One can make several kinds of borscht; beef and pork borscht. For a variant result of the soup, potatoes and carrots are used. Moreover, to use it as a refreshing option, sour cream and milk, yoghurt, buttermilk, cucumber or radishes are used (VFN, 2017).

In Poland, there is a mixture of beetroot and horseradish known as ćwikła, which is used in salads and sandwiches. It can also be used as a side dish with potatoes and meat. In United States, the Pennsylvania Dutch makes "pickled beet egg”. In this recipe, the beetroots are pickled and the liquid is drawn from the root. The beetroot juice is used to marinate hard boil eggs until they become red. Moreover, wine can also be made from beetroots (VFN, 2017).

In Kenya, beetroot is among the major horticultural export products as of 1990 to the European Union. The total horticultural export in 1990 was 125.1 metric tons valued USD 133.4 million (Nyangweso & Odhiambo, 2004). The major beetroot growing counties in Kenya are Kiambu and Nakuru whose production account for approximately 88 percent of the total production of 216 metric tonnes with an approximate value of Kshs. 13 million, as of 2014. Beetroots are most commonly used in Kenya to make juice and sometimes mixed in salads (Mutua, 2016).

2.2 Beetroot Varieties and Planting

There are noticeable differences that exist in the various types of beetroots. These can be summarized in terms of the following ; time of cropping (early, mid or late), texture and taste, colour, shape, and resistance to diseases and growing conditions (Delahaut & Newenhouse, 2013).

Some of the earliest maturing bulbs types include Detroit Dark Red which matures in 58 days and grows to 2.5 to 3 inches in diameter. This type is grown for its leaves and roots consumption, in a wide range of soil and temperature conditions. Other variants include Early Wonder and Crimson King that mature in 52 days. Sweetheart, Sangria and Ruby Queen matures in 58 days, 56 days and in 60 days, respectively. Lastly, Lutz Green Leaf matures in 70 days. It is purplish-red with green tasty tops grown in winter (Delahaut & Newenhouse, 2013).

There are special beetroot types that are grown for a variety of reasons. Cylindria matures in 60 days and is grown for its long, cylindrical shape, which is good as it results in equal slice size. Touchstone Gold is a new variety with small yellow roots that retain their colour once cooked. Green Top Bunching matures after 65 days and has bright red roots with superior tops for greens. Golden matures after 55 days and are carrot-coloured but taste the same as red beetroot and the leaves for this type are sweet. Di Chioggia matures in 50 days and is an Italian heirloom known for its striped red and white interior, sweet, mild taste and early maturation (Gasztonyi et al., 2001).

Currently,beetroot farming in Kenya is gaining popularity among Western Kenya farmers, replacing the traditional cereal planting because of its high returns but at a slow pace. In Kenya, the main producing counties are Nakuru and Kiambu, that contribute about eighty-eight (88) percent of the total production of 216 metric tonnes with a total income value of Kshs. 13 million, as of 2014 . Other counties that also grow beetroots are Nandi, Turkana, Elgeyo Marakwet and Trans Nzoia, bringing the total area under beetroot cultivation to be thirty-two (32) hectares (Mutua, 2016).

Beetroot takes 50-70 days to mature and it can be planted during any period of the year as long as there is plenty of water to irrigate the plant. Additionally, harvesting of beetroot starts when it attains a diameter of ten (10) cm, this is after a period of three months (Mbithi, 2016). Beetroots can be planted any time of the year as long as they are watered regularly. The seeds are planted about 5cm (2in) apart in rows, leaving about 25cm (10in) between rows. The seeds should be approximately 2cm (¾in) deep and watered regularly. The bed can be applied with manure or fertilizers to increase the growth and yield of the beetroot bulbs. Germination is expected within a week or two after planting, after which a fungicide should be sprayed to prevent pests from attacking the plant and moulding of the seedling. Harvesting begins in three months after planting to get medium sized bulbs, but if large bulbs are required, the harvesting should be done on the fourth month (Mbithi, 2016).

An article by The Standard newspaper of the 7th of December 2008 cites a farmer in Western Kenya, Mamboleo farm, Kisumu, who reported that on an acre of land, he planted 400 g of beetroot seeds and harvested 60,000 bulbs. The total cost of production the farmer incurred was Kshs. 20,000- 30,000 for planting, fertilizer, weeding and ploughing, and he reported to have made a handsome return. The current market price per bulb retails between Kshs. 10 - 20. The farmers are starting to learn and understand the economic value of the plant (Otieno, 2008).

2.3 Essential Elements and Nutritional Requirements

Essential elements are nutrients required by the human body, and are elements such as K, Na, Ca, Mg, Zn, Cu and Mn. The elements are important in the body system to assist in various metabolic activities and physiological processes (Prasad, 2004). They are essential to the body; to help in preventing numerous disorders such as reduced immunity, anaemia, poor wound healing among others (Goldhaber, 2003). Essential elements considered in this study and their importance in the human body are briefly discussed in the following subsections.

2.3.1 Potassium

Potassium has many benefits to the human body. It reduces instances of stroke, lowers blood pressure, lowers muscle mass and reduces formation of kidney stones (Carter, 2018). It also helps reduce the mortality rate among children and adults by 20 percent. Moreover, potassium is useful in controlling the electrical activity of the heart and the neurons, while also regulating fluid balance. Lack of potassium has been associated with cardiovascular and renal diseases, and hypertension (Rodan, 2017). The World Health Organization (2015) recommends daily potassium intake of at least 3.5 g/day, while the Institute of Medicine recommends an intake of at least 4.5 g/day for children aged 9–13 years, and 4.7 g/day for older children and adults (Electrolytes & Water, 2005)

In the study of *Betalainic and nutritional profiles of pigment-enriched red beet root (Beta vulgaris L.) dried extract*s (Nemzer et al., 2011) used Inductively Coupled Plasma Atomic Emission Spectroscopy for elemental determinations and found that the average concentration of potassium was 13730 $mgkg^{-1}$.

The concentration of potassium from the study of *Nutritional Value and Economic Feasibility of Red Beetroot from different production systems* was reported to have an average of 33,766 mg kg^{-1} (Straus et al., 2012).

In the determination of mineral constituents, phyto-chemicals and antioxidant qualities of Cleome *gynandra*, Brassica *oleracea* and Beta *vulgaris*, Moyo et al., (2017) reported the content of potassium to be significantly higher in Cleome *gynandra* compared to both Beta *vulgaris* and Brassica *oleracea*.

2.3.2 Calcium

Calcium has many health benefits such as helping to build and maintain strong healthy bones. Children are encouraged to take diets with high calcium nutrients to facilitate their growth, and adults over 50 years need calcium to strengthen their bones. In general, individuals require calcium to help increase functioning of the heart, muscles and nerves. Calcium is beneficial in the reduction of life-threatening diseases such as cancer, high blood pressure and diabetes (Medline Plus, 2012).

Campbell (2000), carried out a study on reference sufficiency ranges in various vegetables that included carrots, broccoli, muskmelon, spinach and tomatoes. Table 2.1 shows the data for calcium concentrations in the various vegetables.

Table 2.1 Calcium concentration in different Vegetables (Campbell, 2000)

Vegetable type	Ca concentration (% wt.) for dry weight
Carrot	2-3.5
Broccoli	1.2-2.5
Muskmelon	3-5
Spinach	1-1.5
Tomatoes	1-3

Fahad et al., (2015) compared the accumulation of calcium in different vegetables where he reported the following; lettuce 95213 ± 101 $mgKg^{-1}$, spinach 86606 ± 102 $mgKg^{-1}$, hyacinth bean 84615 ± 79 $mgKg^{-1}$ and cauliflower 80743 ± 87 $mgKg^{-1}$.

Calcium is the most plentiful mineral found in the human body. The teeth and bones contain the most calcium. Nerve cells, body tissues, blood, and other body fluids contain the rest of the calcium. Calcium helps the body with building strong bones and teeth, blood clotting, sending and receiving nerve signals, squeezing and relaxing muscles, releasing hormones and other chemicals and keeping a normal heartbeat (Medline Plus, 2012).

The intake of calcium depends on the age of an individual and gender. For example, among men and females aged above 19 years, the daily intake is 1000 mg while it is 1200mg for females over 51 years of age (Bauer, 2013).

2.3.3 Manganese

Manganese is an essential nutrient to the body for all ages. It helps support bone health and prevents diseases such as osteoporosis. The nutrients are absorbed by the bones together with calcium and facilitate a healthy bone development. It is also an antioxidant with an enzymatic function. Manganese is essential for cognitive development in young adults and children and in prevention of diseases such as diabetes and arthritis. Manganese helps women reduce their premenstrual symptoms and helps in weight loss. The daily recommended intake for manganese for adults is 2-5 mg/day and 2-3 mg/day for children (Cappuyns et al., 2000; WHO, 2005).

A comparative study of reference sufficiency ranges in various vegetables that included carrots, broccoli, Muskmelon, spinach and tomatoes is shown in Table 2.2 for Manganese.

Table 2.2 Manganese concentration in different Vegetables (Campbell, 2000)

Vegetable type	Mn concentration (mg Kg^{-1})
Carrot	30-60
Broccoli	25-150
Muskmelon	20-100
Spinach	25-200
Tomatoes	25-200

Moyo et al. (2017), compared the concentration of Mn in Cleome *gynandra* (spider plant) and Beta *vulgaris* L (leaf vegetable known as chard or spinach beet) and found that Beta *vulgaris* had 1.5 times higher concentration of manganese than Cleome *gynandra.*

Table 2.3 shows the concentration of manganese in various vegetables (Odhav et al., 2007).

Table 2.3 Concentration of manganese in some sampled vegetables (Odhav et al., 2007)

Vegetable Samples	Concentration in leaves (mg/100g)
Amaranthus *dubius*	82
Amaranthus *hybridus*	24
Amaranthus *spinosus*	3
Asystasia *gangetica*	18
Cleome *monophylla*	10
Oxygonum *sinuatum*	4

Ekholm et al (2007) carried a study in Finland to analyse changes in the mineral and trace element contents of cereals, fruits and vegetables. It was reported that the concentration of manganese in different root vegetables were: potato 0.7 mg/100 g, organic carrot 1.2 mg/100 g, beetroot 3 mg/100 g, parsnip 1 mg/100 g, celery root 1.5 mg/100g, turnip 1mg/100 g, swede 1.1 mg/100 g, radish 1 mg/100 g and Jerusalem artichoke 0.2 mg/100g.

Manganese plays a very important role in the human body's physiological processes such as help to regulate blood sugar level, proper function of the thyroid gland and sex hormones, metabolism of fats and carbohydrates and also as a powerful antioxidant (Shan et al., 2016). However, an overdose of manganese can lead to symptoms of Parkinson's disease, nerve damage and lung embolism. On the other hand, effects of manganese deficiency include skin problems, skeletal disorder, glucose intolerance and neurological symptoms (Crossgrove & Zheng, 2004).

2.3.4 Iron

Iron in diets is available in two forms, heme and non-heme. Some of the sources of the heme form are found in red meat, fish and poultry. On the other hand, non-heme form is found in fruits and vegetables, cereals and legumes. The bioavailability of the latter is lower but is in a greater quantity than the heme-iron. Therefore, non-heme iron is more beneficial and is found in beetroots (Moat, 2008).

Iron is useful in metabolic processes such as oxygen transportation, deoxyribonucleic acid (DNA) synthesis and electron transport. Iron ensures that enough red blood cells are transported throughout the body. Among women, iron is beneficial during pregnancy and menstrual periods. Lack of it results in deficiency, which may be fatal to a pregnant woman or any adult as it inhibits right amount of oxygen to be transported. Beetroots contain sufficient iron that increases its absorption and use in the body (Abbaspour et al., 2014).

In a study by Tidemann-Andersen et al. (2011), on iron and zinc content of selected foods in the diet, they reported a mean of 50 $mgKg^{-1}$ in cassava, 140 $mgKg^{-1}$ in Irish potatoes and 56 $mgKg^{-1}$ in sweet potatoes. In a survey of plant iron content, (Ancuceanu et al., 2015) determined the mean Fe content for roots to be 225 $mgKg^{-1}$, 74.3 $mgKg^{-1}$ for barks, 195.4 $mgKg^{-1}$ for fruits and 333 $mgKg^{-1}$ for seeds

Petry et al., (2015) analysed beans from Cali, Colombia for Iron content, where a mean concentration of 71 µg/g - 280 µg/g was reported. Ekholm et al. (2007) carried a study in Finland to analyse changes in the mineral and trace element contents of cereals, fruits and vegetables. He reported the concentrations of iron in different root vegetables as; potato 3.4 mg/100 g, organic carrot 3.4mg/100 g, beetroot 2.9 mg/100 g, parsnip 3 mg/100 g, celery root 5.5 mg/100g, turnip 4 mg/100 g, swede 3.4 mg/100 g, radish 6.5 mg/100 g and Jerusalem artichoke 1.8 mg/100g.

The dietary requirement of iron varies with age and sex. Boys and girls need the same amount of iron 10 mg/ day from ages 4 to 8, and 8 mg/ day from ages 9 to 13. Women need more iron from ages 19 to 50 because they lose blood each month during their periods, that is why they need to get 18 mg of iron per day, while men of the same age can get 8 mg/day (Beck et al., 2014).

2.3.5 Zinc

Zinc has many health benefits such as treating diarrhoea, regulating the immune function and helps in learning and memory (cognitive function). It also helps treat common colds and assist in wound healing. The nutrient is part of components in platelets that helps in treating wounds and foster quick healing. The daily dietary requirement allowance for zinc is 14 mg/day for adults and 10 mg/day for children above one year (Wessels *et al*., 2017; WHO, 2005).

In the evaluation of black tea, it was reported that the zinc concentration had a range of 0.2-1.5 mg Kg^{-1}(Brzezicha-Cirocka *et al*., 2017). Basha et al. (2014), conducted a study on vegetables and fruits cultivated around the surroundings of Tummalapalle uranium mining site and reported a zinc concentration of 4.1±0.8 mg Kg^{-1} for tomatoes, 8.5±6.5 mg $Kg^{-1}$for green chilli, 5.2±0.8 mg $Kg^{-1}$for bitter gourd, 4.5±2.9 mg $Kg^{-1}$for bananas, 7.3±5.1 mg $Kg^{-1}$for papaya and 9.2 ± 4.2 mg $Kg^{-1}$for melons.

Odhav *et al*. (2007) determined zinc levels in some sampled vegetables and the concentrations are shown in Table 2.4.

Table 2.4: Concentration of zinc in some sampled vegetables (Odhav *et al*., 2007)

Vegetable Samples	Concentration in leaves (mg/100g)
Amaranthus *dubius*	56
Amaranthus *hybridus*	18
Amaranthus *spinosus*	15
Asystasia *gangetica*	7
Cleome *monophylla*	5
Oxygonum *sinuatum*	7

Ekholm et al. (2007) carried a study in Finland to analyse changes in the mineral and trace element contents of cereals, fruits and vegetables. They reported the concentration of zinc in different root vegetables to be: potato 1 mg/100 g, organic carrot 2.1 mg/100 g, beetroot 1.6 mg/100 g, parsnip 2.1 mg/100 g, celery root 3.7 mg/100g, turnip 2.4 mg/100 g, swede 1.9 mg/100 g, radish 3.1 mg/100 g and Jerusalem artichoke 1.4 mg/100g.

Zinc is an essential trace element because very small amounts of zinc are necessary for human health. It is used for treatment and prevention of stunted growth and acute diarrhoea in children. It is used for boosting the immune system, treating common cold, recurrent ear infections and preventing lower respiratory infections (Wessels *et al*., 2017). However, high levels of zinc promote development of kidney stones, which lead to stomach aches, vomiting, headaches and loss of appetite. While low levels of zinc are associated with slow healing of wounds, repressed growth, loss of appetite and hair loss (Beaver *et al*., 2017).

2.4 Health Benefits of Fruits and Vegetables

Research indicates that increased consumption of fruits and vegetables help to reduce lifestyle related diseases and illnesses; obesity, heart related diseases and diabetes. Moreover, it is beneficial in improving skin and hair tone in people. It also helps to increase energy levels among its users and helps in weight reduction (LD M, 2017). More health benefits of consuming fruits and vegetables are discussed briefly below:

Heart health and blood pressure: a proven effect of consuming vegetables is visible in its reduction of blood pressure. In a study done in 2008, ingesting 500 millilitres of beetroot juice among health volunteers lowered blood pressure. Scientists attributed the reduction possibly due to the high levels of nitrates in beetroots. The nitrates were likely to be the cause of reduction in blood pressure and in the treatment of cardiovascular conditions. A similar research conducted in 2010 confirmed the assertions (Cacciapuoti, 2012).

Dementia: oxygenation to the brain is likely to be improved by consuming beetroot juice, according to researchers at Wake Forest University. The oxygenation is likely to slow the progression of dementia among older adults. According to Daniel Kim-Shapiro, director of Wake Forest's Translational Science Centre, blood flow to certain areas of the brain decrease with age and leads to a decline in cognition and possible dementia (LD M, 2017). Drinking beetroot juice can help improve blood flow and oxygenation with the high intake of nitrates.

Diabetes: beetroots are known to contain an antioxidant known as alpha-lipoid acid. The antioxidant may help lower glucose levels and increase insulin sensitivity (LD M, 2017). It can also prevent stress-induced changes that affect many patients with diabetes.

Further research on the antioxidant indicates a decline in symptoms of peripheral and autonomic neuropathies among diabetic people (LD M, 2017). However, a different research suggests that benefits of the antioxidant may be restricted with the administration of the acid. The authors concluded: "It is unclear if the significant improvements seen after 3 to 5 weeks of oral administration at a dosage of more than 600 milligrams a day are clinically relevant."

Digestion and regularity: beetroot is good in digestion due to its high fibre content. It helps minimize constipation and promotes a healthy digestive system.

Inflammation: beetroot contains a vital nutrient referred to as choline. Choline helps in muscle movement, sleep, memory and learning. It assists in maintaining the structure of cellular membranes. It also assists in absorption of fat and helps to reduce chronic inflammation (LD M, 2017).

Exercise and athletic performance: The high nitrate content in beetroot juice helps to supplement oxygenation during exercise hence resulting to high tolerance in long-term endurance exercise. In a research, scientists concluded that beetroot juice improved performance by 2.8 percent, or 11 seconds, in a 4-km bicycle time trial and by 2.7 percent, or 45 seconds, in a 16.1-kilometer time trial (LD M, 2017).

2.4.1 pH Classification of Fruits and Vegetables

Fruits and vegetables are classified as either acidic or basic. To determine if a food is acidic or alkaline, it is required to first heat the food until it only remains ashes- a process that mimics the digestive process in the body. Water is then added to the ashed sample to make a saturated solution which is used to measure the pH of the food (Virginia, 1985).

Lemon which tastes sour and tests highly acidic, is classified as basic because it has a high content of sodium, potassium, calcium and magnesium. The ashes of samples, which contain high levels of these elements give an alkaline solution when mixed with water, hence, are classified as basic. Also, foods that contain elements like chlorine, phosphorus or sulphur are acidic. Such foods include spinach and carrots (Virginia, 1985).

2.5 Principles of EDXRF Method of Analysis

X-ray fluorescence method of elemental analyses is based on the principle that individual atoms, when excited by an external energy source, emit X-ray photons of a characteristic energy or wavelength.

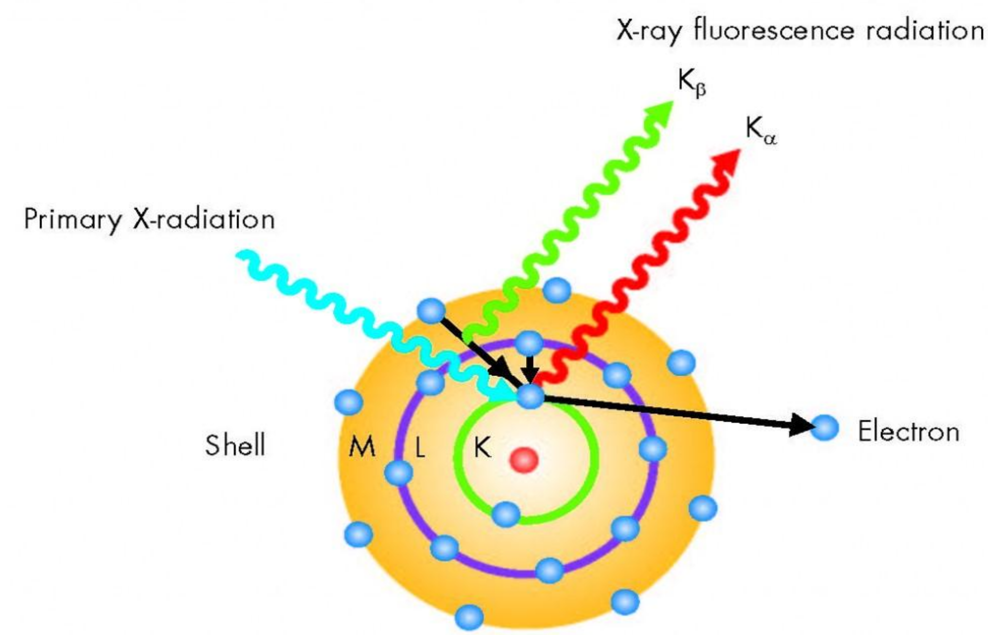

Figure 2.2: X-ray Fluorescence Radiation (Verma, 2007)

Most atoms have electron orbitals (K shell, L shell, M shell) and therefore a number of fluorescent transitions are possible. Interaction of X-rays with an atom result in a hole forming in the K shell, for example, which is then filled by an electron from the L shell or from the M shell (Verma, 2007). In either case, these are termed as K transitions. K_α radiation is emitted when an L shell electron fills the vacant space in the K shell, and K_β radiation is emitted when an M shell electron fills the vacant space in the K shell.

Alternatively, a hole could be formed in the L shell, subsequently filled by an electron from the M shell (L transition). Thus, for a single element, several fluorescence peaks are possible, and typically these will all be present in the energy spectrum. They form a characteristic fingerprint for a specific element (Verma, 2007).

In principle, a typical X-ray fluorescence spectrometer consists of a source of X-rays which is used to irradiate the sample, the sample and detector of the emitted fluorescent X-rays (Guthrie, 2017).

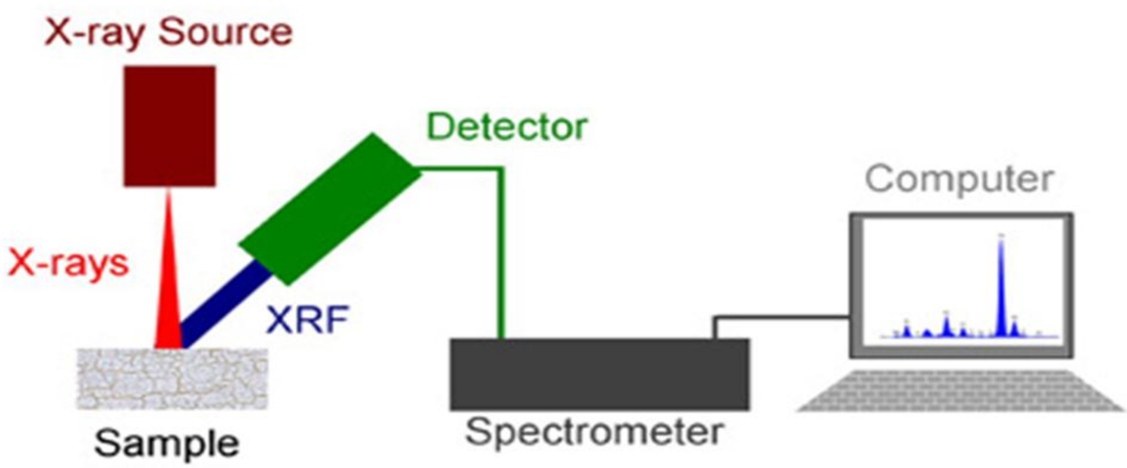

Figure 2.3: XRF set up (Guthrie, 2017)

The absorption of X-rays by a particular material varies according to energy of the incident X-rays. High energy photons are absorbed less than low energy X-ray. To eject an electron from one of the orbitals, the X-ray photon energy must exceed the binding energy of that electron. Therefore, not all the incident X-rays result in fluorescence (Dolenko & Latośińska, 2017).

The identification of an element is determined by energy of the X-ray photon emitted by that particular element. The number of photons emitted gives the amount of that analyte in the sample. The analysis is rapid and usually requires minimal sample preparation (Verma, 2007).

The range of detectable elements depends on the instrument configuration and set up, but typically covers all elements from sodium (Na) to uranium (U). Limits of detection depend upon the specific element and the sample matrix. Heavier elements will have lower detection limits (Verma, 2007).

Some of the emission energies of identifiable elements are indicated in Table 2.5;

Table 2.5: Emission energies of selected elements (Davis *et al*., 2011)

Element	K_α (KeV)	K_β (KeV)
Potassium	3.3	3.6
Calcium	3.7	4.0
Manganese	5.9	6.5
Iron	6.4	7.1
Zinc	8.6	9.6

In practice, EDXRF has been used in various research studies, for example: Stihi et al., (2014) carried out a study on mineral content (K, Ca, Fe, Mn, Cu and Zn) of native vegetables from Romania using EDXRF as shown in Table 2.6.

Table 2.6: Mean concentrations of elements in leafy vegetables (%/ ppm ± s.d.) (Stihi *et al*., 2014) (n=5), d.w

Sample types	K %	Ca %	Mn	Fe	Cu	Zn
Atriplex *hortensis* (orache)	4.75±1.6	2.05±0.2	81.5±4.6	944.2.±12	15.2±4.6	84.5±6.8
Rumex *patientia* (patience)	4.55±2.3	1.72±0.5	72.04±10.2	952.05±17.25	19.06±7.9	80.12±9.6
Lactuca *sativa* (salad)	2.9±0.6	1.2±0.4	89.2±5.5	1002.5±14	25.5±1.7	45.5±3.6
Spinacea *oleracea* (spinach)	4.20±1.2	2.15±0.8	75±1.2	945.5±14.2	17.4±4.6	82.4±15.2

In a study of characterizing aerosol particles at an industrial park in Nairobi, EDXRF was used to analyse black carbon and trace elements (Si, S, Cl, K, Ca, Ti among others) to find out the source of air pollution. It was found that, industries, vehicles and biomass burning were the main sources of the black carbon, K and S in the fine fraction of the samples and Si, Ca and Fe in the coarse fraction, (Gatari et al., 2009).

Muohi et al., (2003) carried out a study in Makupa and Port- Reitz using EDXRF and AAS to assess Cu, Zn, Pd and Cd accumulation. Industrial activities and nearby municipal dumpsite were assumed to be the source of the high element concentration.

In a paper on heavy metal profiling of gold ores and sediments associated with gold mining at Migori gold belt, EDXRF was used to analyse for heavy metal (Ti, Ca, Zn, As, Au, Pd and Cd). The assessment of occupational exposure of the miners and investigation of environmental impact of toxic heavy metals found that As, Pd, Ti and Zn levels were above the WHO recommended levels of 50 mg/kg (Odumo *et al*., 2011).

For the assessment of air pollution level in Nairobi, Ca, Fe, Mn, Cu, Zn and Br were analysed using EDXRF. Vehicular emission was found to be the main source of pollution. The levels were found to be higher than the WHO limit of 150 $\mu g/m^3$ (Maina, 2004).

A study was carried out to analyse Kenyan rock samples for trace, minor and major elements. EDXRF was used for the analysis of K, Ca, Ti, Cr, Mn, Fe and Zn among many more *(Angeyo et al*., 1998).

2.6 Data Analysis

2.6.1 Students t test – distribution: Comparison of two data set

The t distribution is used to compare the experimental values with the certified values to determine the applicability, repeatability, reproducibility and accuracy of the analytical method. In general, samples are analysed in replicates.

t is calculated using the equation:

$$t_{cal} = \frac{\bar{X}}{s/\sqrt{(n)}} \quad \text{.. Eq 2.1}$$

Where $\bar{X}$ - mean of the analysed reference value

s- Standard deviation of the analysed reference values

n- Number of measurements

t_{tab} values can be found from the t-test distribution table at various confidence intervals for degrees of freedom. In the t-test, if $t_{calc} < t_{tab}$, there is no significant difference between the experimental value and the certified values (Bluman, 2009).

Altundag & Tuzen, (2011), carried out a study on comparison of dry, wet and microwave digestion methods for the multi-element determination in some dried fruit samples from Turkey. They used student's t-test to estimate the significance of the data values they obtained. The comparison of dry, wet and microwave digestion methods showed no statistically significant difference at $p < 0.05$.

Mohammed & Khamis, (2012), did an assessment on heavy metal contamination in vegetables; Amaranth and cabbage consumed in Zanzibar. There was a significantly higher concentration of Zn, Fe, Cr and Mn in Amaranth than in cabbage, and there was a significantly higher concentration of Cd, Ni and Pb in cabbage than in Amaranth at $p<0.05$.

Otaka et al. (2014), determined trace elements in soybeans using EDXRF. Student t-test was used to show significant differences between soybean from Japan and those from abroad. The results showed that there were significant differences in Mg, P, Cl, K, Mn, Cu, Br and Ba with 95% confidence level, suggesting that those elements could be used for discrimination of geographical origins.

Cherop, (2011), carried out a study on selected essential elements in traditional vegetables, medicinal plants, fruits and conventionally grown vegetables in Koibatek, Kenya. T-test was used to compare AAS and EDXRF mean levels of Zn, Fe, Mn, Cu and Cr in vegetables at $p<0.05$. There is a significant difference in levels obtained by both methods for all the elements except Zn.

2.6.2 Analysis of Variance (ANOVA)

ANOVA is used for testing significant differences between more than two means. It is based on the fact that variance can be partitioned (Bluman, 2009). The variance is calculated using the equation:

$$F_{cal} = \frac{s_B^2}{s_W^2} \quad \text{……………………………………………………………… Eq 2.2}$$

Where $s_B^2 = \frac{\sum n_i \{\bar{X}_i - \bar{X}_{GM}\}^2}{k-1}$

$$\bar{X}_{GM} = \frac{\sum \bar{X}_i}{N}$$

$$s_W^2 = \frac{\sum (n_i - 1) s_i{}^2}{\sum n_i - 1}$$

s_B^2- Between group variance

s_W^2- Within group variance

GM- Grand mean

k- Number of groups

N- Total number of samples analysed

f_{tab} values can be found from the f-test distribution tables at various confidence intervals for degrees of freedom of the denominator and numerator. If $f_{cal} < f_{tab}$, there is no significant difference at a given probability, for example at 95% probability (Bluman, 2009).

Kaniu et al. (2012) did a study on direct rapid analysis of trace bio-available soil macronutrients. There was no statistical difference at 95% confidence level when he compared predicted macronutrients with reference standards using a one-way ANOVA test.

Amin et al. (2013) used ANOVA to report that heavy metal accumulation was significantly greater in vegetables irrigated with waste water of Baghdada Mardan and Sheikh Maltoon Town Mardan, as compared to the vegetables irrigated with tube well water. The trend in heavy metals in vegetable samples was in the order of Mn> Cu> Fe> Zn> Cr>Pb> Ni> Co.

Kaymak et al. (2010) carried out a study on elemental analysis of four different radish cultivars (Raphanus *sativus* l.). There was a statistically significant difference in the four different types of radish cultivars at $\alpha=0.01$ in both years of the experiment done in 2004 and 2005.

Mutua, (2014),determined lead, cadmium and zinc speciation in garage soils level. ANOVA was used to find out whether there was significant difference in the means of heavy metals at different sub levels of each garage site samples. It was found that the means of the three sub levels at two garage sites; Kaunda and Mwangi were significantly different from each other. Meanwhile, the mean concentrations of the three levels at Industrial Area garage site was not significantly different.

2.7 Summary of Literature Review

The history of beetroots usage dates back in the third millennium BC. Different varieties of beetroots and their specific growing conditions are discussed. Essential elements considered in this study; K, Ca, Fe, Mn, Zn, and their role in the human health are also discussed. The health benefits of fruits and vegetables consumption are also discussed.

The principles of EDXRF analysis are explained and several studies that have used EDXRF for their analyses have been mentioned. T- test and ANOVA are some of the statistical tools used for data analysis.

To date, there are limited studies conducted in Kenya on essential nutritional elements in vegetables. In particular, this study seeks to profile essential elements in beetroots.

CHAPTER THREE: MATERIALS AND METHODS.

3.0 Introduction

Beetroot samples used in this study were randomly sampled from farms in Karatina, Gilgil and Naivasha, and open air markets in Kisumu and Joska. In general, these farms are small-scale farms, where the main food crops grown include; maize, beetroots, cabbage, spinach, onions, kale, carrots, potatoes, beans and bananas. The farming is mainly rain dependent, but some farmers also irrigate when necessary and all the farmers use manure and fertilizers to improve on the crop yield.

3.1 Description of the Study Areas

Gilgil and Naivasha are located in Nakuru County, approximately 80-125 Km to the north of Nairobi. Nakuru has an area of about 7,500 km^2 and is home to approximately two million people as per the 2009 national census in Kenya (KNBS, 2012). The county is cosmopolitan with Kalenjin accounting for the biggest population. The county has a mean annual rainfall of 700-1200 mm, with the two rainy seasons experienced in mid-March and May (long rains) and October and December (short rains). The temperatures in the county have a high of 25 °C and a low of 10°C. The county has well drained, red volcanic soils (Andosols) with a pH range of 5.8-6.8 (Gachene C K & Kimaru G, 2003; KNBS, 2012; Ogeto et.al, 2013).

Karatina is located 55 Km northeast of Nairobi. It has a population of approximately 6,852, with Kikuyu being the predominant ethnicity (KNBS, 2012). The area has a mean annual rainfall of 1432 mm, with two rainy seasons experienced in April and May (long rains) and October and November (short rains). Karatina experiences a high of 26°C and a low of 8 °C in temperature, the soil found is highly weathered, deep red clay soil (Nitisols) (Gachene C K & Kimaru G, 2003; Kihonge, 2014; Speck, 1982).

Kisumu is a Kenyan port city on Lake Victoria and is approximately 360 Km from Nairobi. It has a population of about 968, 909 as of the 2009 census and the vast majority of people belong to the Luo ethnic community (KNBS, 2012). The county's climate is modified by the presence of Lake Victoria, with the annual relief rainfall ranging between 1200 mm and 1300 mm and is normally accompanied by thunderstorms. The County is warm throughout the year with a high of 35 °C and a low of 20 °C and has black cotton soils (Gachene C K & Kimaru G, 2003; Karanja, 2010).

Joska is a town located in Machakos County, about 66 Km to the east of Nairobi. Machakos has a population of approximately one million people and the predominant ethnical community is Kamba. The county has an annual rainfall of 958 mm, low fertile loam soil and a high in temperature of 28 °C and a low of 11 °C (KNBS, 2012). Figure 3.1 shows the different geographical sampling points.

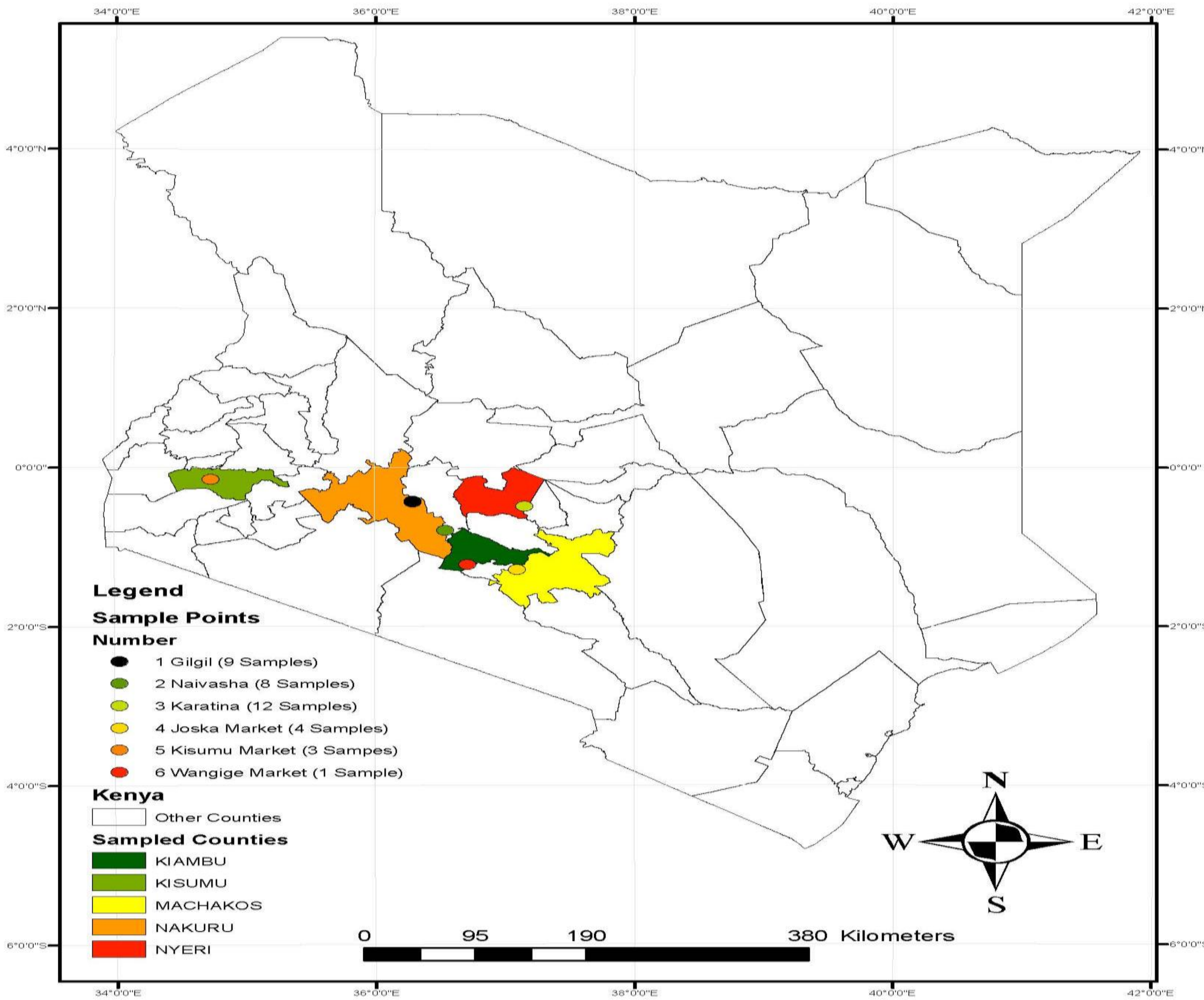

Figure 3.1 Sampling points

3.2 Sampling Design

The three farms in; Karatina, Gilgil and Naivasha were chosen because they are located in the major beetroot growing areas in Kenya, according to information obtained from a survey of market places in Nairobi. Random sampling design with offset grid sampling pattern was used to sample beetroots bulbs as shown in Figure 3.2.

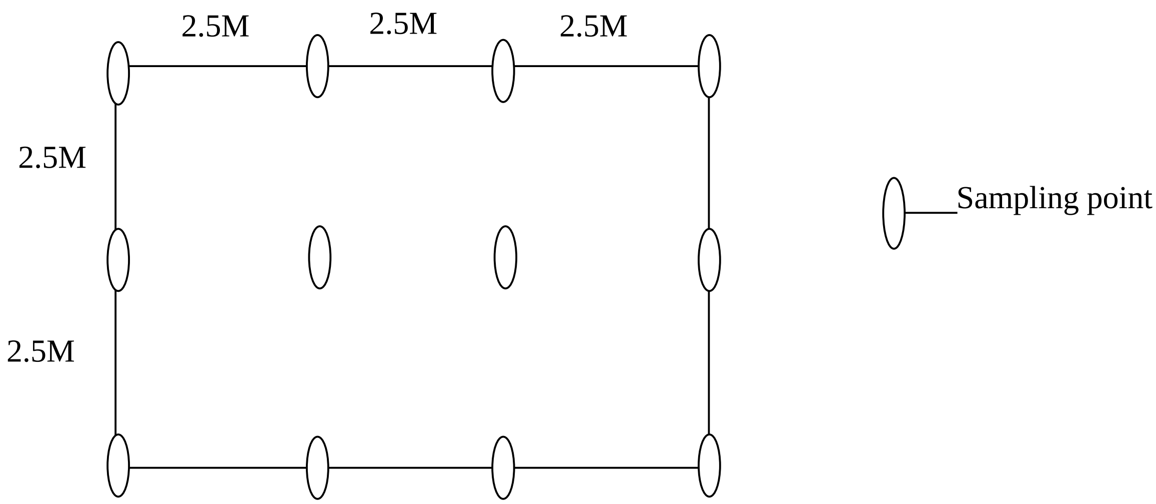

Figure 3.2 Sampling pattern

From each sampling point, a beetroot sample, either one bulb or two bulbs depending on the size were collected and placed in a brown paper bag. The soil attached to the bulb was not removed because the soil helps the bulb remain fresh for long, then labelled as follows: K1-S (00°29.142), E (037°09.219): to represent sample1 from Karatina at GPS location S (00°29.142), E (037°09.219). In general, twelve (12) samples were obtained from Karatina (K1S (00°29.142), E (037°09.219) - K12 S (00°29.145), E (037°09.218)). Garmin eTrex 10 GPS device was used to get the coordinates of sampled locations.

Additional fresh and large beetroots bulbs samples were purchased from two markets: two (2) from Kisumu and four (4) from Joska-Machakos. For comparison purposes, samples of carrots (Daucus *carota*) bulbs, potato (Solanum *tuberosum*) bulbs and Moringa *oleifera* leaves were also sampled for analyses for essential elements content.

There was no sample preservation and pre-treatment prior to laboratory sample preparations for elemental analyses. A total of thirty-four (34) samples were collected for analyses of essential element content in this study- Appendix 1.

3.3 Sample Preparation

The sample preparation was done in two-steps: one for EDXRF analysis and the other for pH measurement.

3.3.1 Sample Preparation for EDXRF Analyses

Initially, all the samples were washed in double distilled water to remove soil and any foreign particles. The samples were then sliced into thin slices then dried in the oven at 60^{o}C for 56 hours. This temperature and time was arrived at after carrying out preliminary tests, where bulbs were first put in the oven at 30^{o}C for 24 hours but complete drying was not achieved. Then the beetroots were further dried at 60^{o}C for 24 hours, which also resulted in incomplete drying.

Finally, the 60^{o}C was retained, but the drying time increased until such a time a constant weight was recorded, which was after 56 hours. After drying, the samples were ground into fine powder using a grinder then sieved with a 75-micron sieve.

For each ground sample, a mass of between 0.5-0.6 grams was accurately weighed for making pellets for EDXRF analyses. The powder was placed between two stainless steel dies enclosed in a cylindrical stainless steel chamber then pressed using the hydraulic press at 10 kPa to make thin pellet that were 25 mm in diameter. Each pellet was accurately weighed, labelled and stored in a Petri dish ready for analysis. For each sample, three pellets were prepared for EDXRF analysis (Woods & Collins, 2004).

Figure 3.3 – 3.5 shows a photographs of beetroot powder after preparation, the hydraulic pellet press used and a beetroot pellet sample for EDXRF analysis.

Figure 3.3: Beetroot powder

Figure 3.4: Hydraulic pellet press

Figure 3.5: Beetroot pellet

3.3.2 Sample Preparation for pH measurements

1.0 g of beetroot powder was transferred into a 250 ml beaker and 100 ml of distilled water was added then stirred thoroughly to make a saturated solution. The mixture was let to settle for fifteen minutes (15) then filtered using a filter paper, Whatman grade 1: 125 mm and the filtrate stored in a plastic sample bottle container ready for pH measurement (Agrawal *et al*., 2015).

3.4 Sample Analysis

3.4.1 pH Determination

An Orion star AZ14 from Thermo Scientific PH/ISE meter was used to determine the pH of the samples solution. Prior to measurements, the pH meter was calibrated using three buffer solutions; pH 4, 7 and 9.2. 20.0 ml of the beetroot solution was pipetted into a sample holder then the pH meter electrode was dipped to determine the pH (Agrawal *et al.*, 2015).

3.4.2 Elemental Content Determination by Energy Dispersive X-Ray Fluorescence

3.4.2.1 Instrumentation for Energy Dispersive X-Ray Fluorescence

In this work, the following EDXRF spectrometers were used for determination of elemental content:

1) Rigaku NEX CG bench top EDXRF at the Department of Physics, College of Biological and Physical Sciences, University of Nairobi- Figure 3.6
2) EXP-1 XRF Experimenter's Kit, Mini-X at the Institute of Nuclear Science and Technology, University of Nairobi- Figure 3.7

The Rigaku EDXRF spectrometer NEX CG used has the following specifications as shown in Table 3.1:

Table 3.1: Specifications of Rigaku NEX CG EDXRF (Moriyama, 2013)

X-Ray tube	Pd target, air cooled
Tube power	50W: 50 Kv-2mA (max)
Secondary targets	5 targets (max); RX9, Cu, Mo, Si and Al
Detector	High performance SDD (Silicon Drift Detector)
Atmosphere	Vacuum, Air, He

Figure 3.6: Bench top Rigaku EDXRF for sample analysis

The energy dispersive X- ray fluorescence spectrometer NEX CG with polarized optics enables data measurements with low background and high peak- to- background ratio (P/B ratio). Thus, trace metals of hazardous elements contained in environmental samples can be analysed with high sensitivity. Moreover, the scattering fundamental parameter method corrects for the influence from non-measurable components in samples such as coal fly ash, soils and biological samples by using Compton and Thomson scattering intensities from a Mo secondary target (Moriyama, 2013)

The NEX CG is equipped with RX9, Cu, Mo, Si and Al secondary targets, and the measurable elements range from Na- U. A specialized light element optimization (LEO) secondary target is also available for improved analysis accuracies of Na and Mg. A high performance SDD (Silicon Drift Detector), which requires liquid nitrogen, is utilized to realize high analytical precision and accuracy (Moriyama, 2013).

Secondary targets and the polarized optics provide a high P/B ratio spectrum compared to direct excitation optics in a wide energy range. Polarized optics gives greatly improved performances compared to the direct excitation optics for trace element analysis (Yoshiyuki *et al*., 2005).

NEX CG has the advantage of causing no radiation damage to the samples because of very low X-ray radiation to sample dose due to indirect radiation in the secondary target optics (Moriyama, 2013).

The mass, diameter, thickness and position of the pellet were keyed into the computer then the X-rays turned on and the chamber evacuated because the analysis is done in vacuum. Each pellet took an average of 4 minutes to analyse given that each secondary target took 50 seconds to irradiate on the pellet and there were 5 targets in the Rigaku EDXRF.

The data obtained was displayed on the computer screen as concentration for each element in parts per million and the spectrums in energy (keV) and Intensity (counts per second). The data was stored in a compact disk.

The AMPTEK EXP-1 EDXRF Spectrometer used is available at the Institute of Nuclear Science and Technology consists of an XR100 SiLi detector system, the PX4, the Mini-X, baseplate and associated sample analysis software.

Specifically, the X100 constitutes the X-ray Si (Li) detector and its preamplifier. The PX4 constitutes three main parts: shaping amplifier, multichannel analyser and power supply. The Mini-X is an X-ray tube system that constitutes a 40 kV/100 μA power supply, X-ray Mo tube, USB provision for communication between computer and electronics. The base plate holds the sample, detector, and radiation source to a well-known fixed geometry. The source and detector are positioned at an angle of 45^{o} relative to each other for scatter and incident angles of 67.5^{o}. The distance between the detector and the sample is 1.6 cm. The software used in this system constitutes the Analog to digital multichannel analyser (ADMCA) tube control and data acquisition software and XRS-FP spectrum analysis software (Lyman et al., 2012).

Figure 3.7 shows the AMPTEK EXP-1 EDXRF Spectrometer system used in this study and is available at the Institute of Nuclear Science and Technology, Nairobi University.

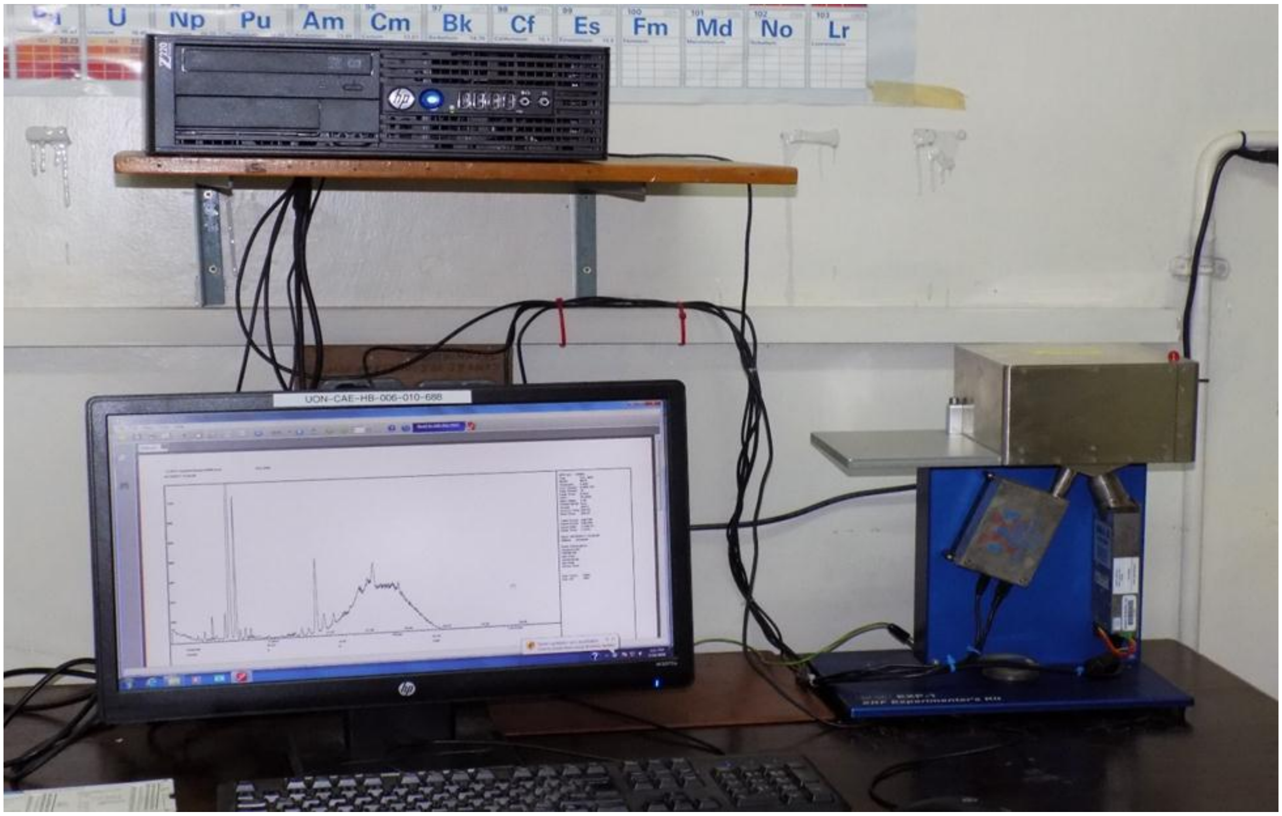

Figure 3.7: AMPTEK EXP-1 EDXRF for sample analysis

3.5 Quality Control

For quality control of measurements, two certified reference materials (CRM), Bowen kale and Soils 5, all from the IAEA were analysed by the two EDXRF instruments and the results compared to the certified reference values. All the samples were also analysed in triplicates.

3.6 Statistical Analysis

The data obtained for the mean essential elements concentration (mg/Kg) of the sampled beetroots was analysed using ANOVA and t-test. Then the spectrum was plotted using Micro soft Excel.

CHAPTER FOUR: RESULTS AND DISCUSSION

4.0 Introduction

Essential elements play an important role in human nutrition and health. In this chapter, the analytical results of the beetroot bulb samples, concentrations of K, Ca, Mn, Fe and Zn and pH are presented and discussed. Comparison of data from two EDXRF instruments are also presented and discussed as well as the validation of EDXRF method. The presentation is done using tables, spectra and graphs.

4.1 Sample Excitation Conditions with Secondary Targets: Rigaku NEX CG

The five targets utilized in this analysis are RX9, Cu, Al, Mo and Si target (Moriyama, 2013). They provide a high peak to background ratio spectrum compared to direct excitation optics in wide energy range of the x-rays.

All the samples were analysed using the five secondary targets. The spectrum for a typical beetroot sample is shown in Figures 4.1– 4.5.

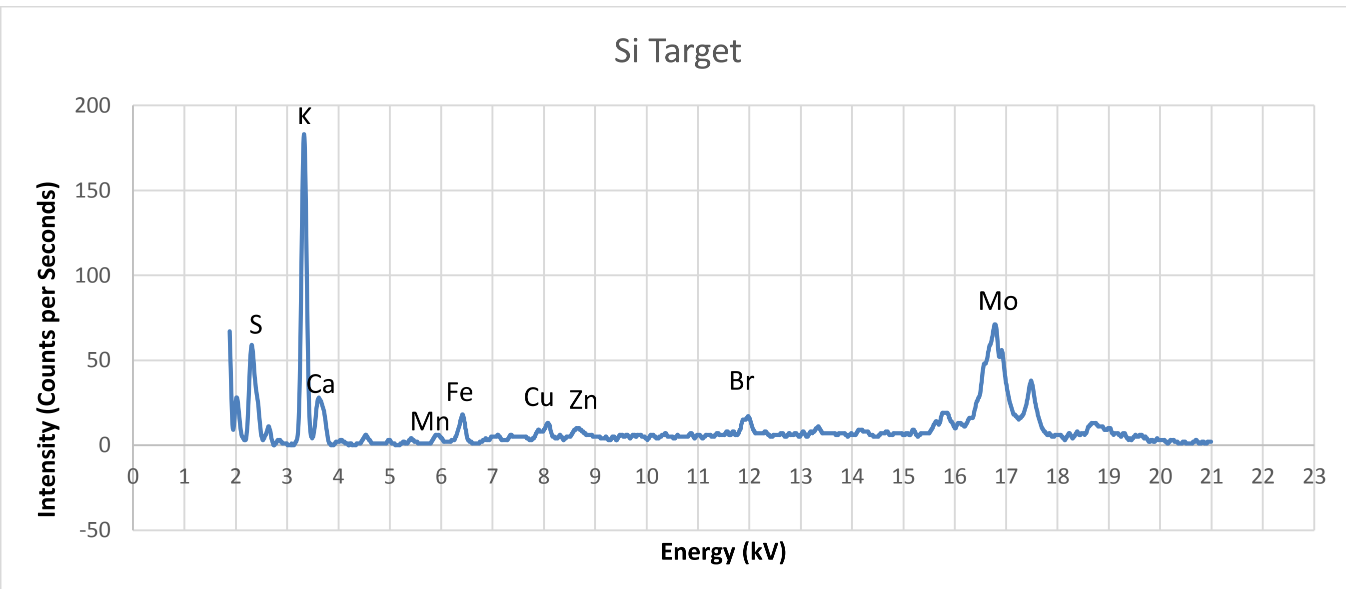

Figure 4.1: Beetroot Spectrum using Si target

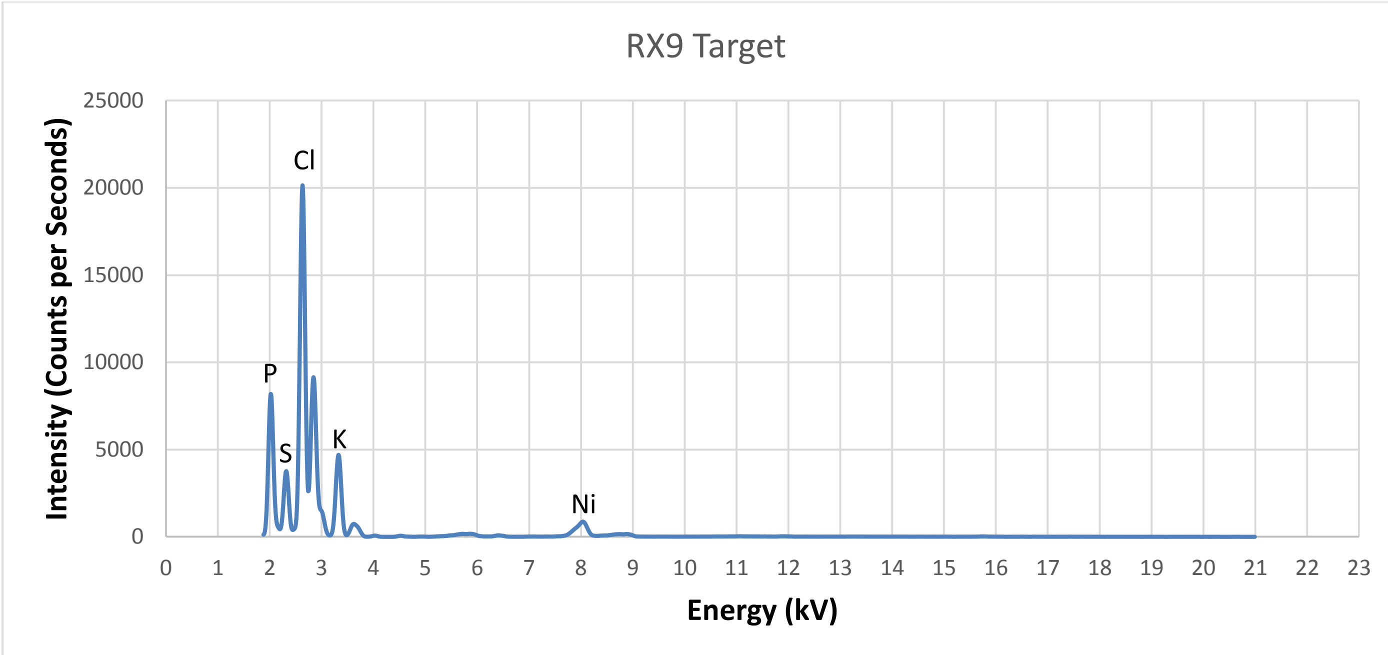

Figure 4.2: RX9 Target

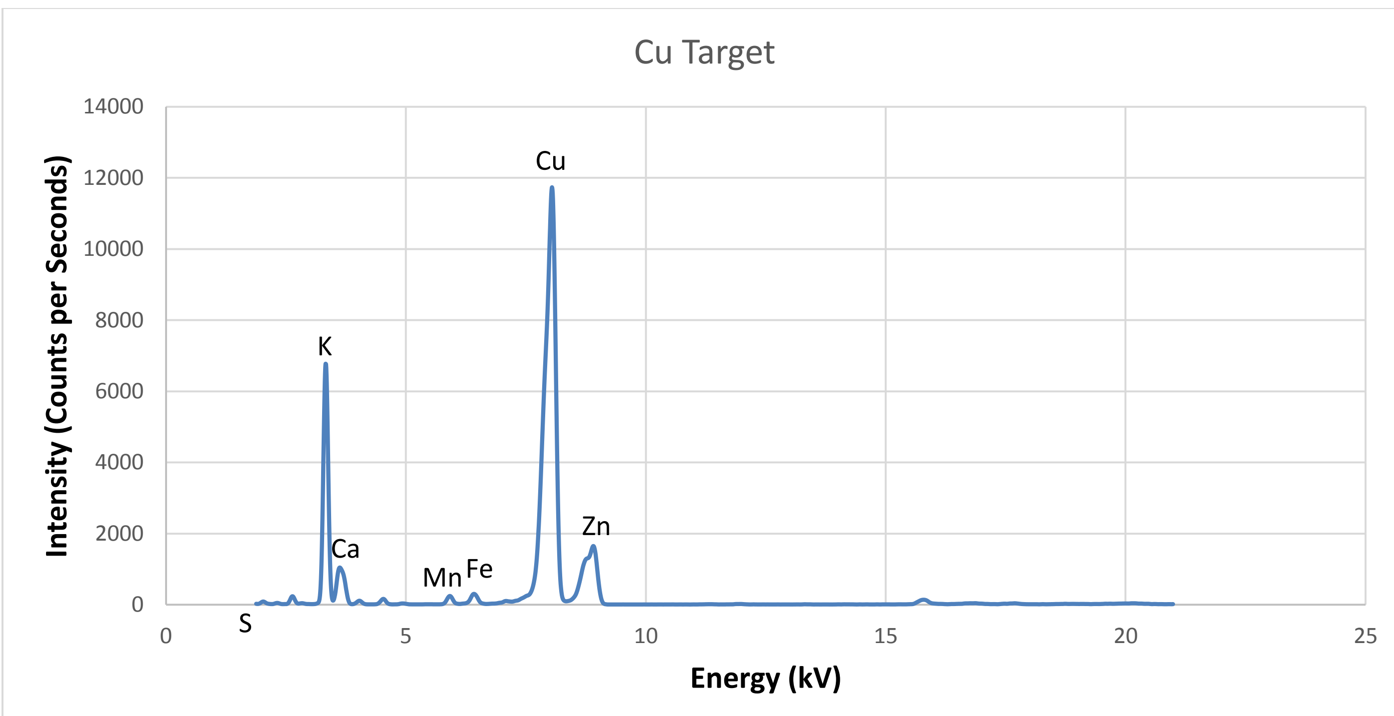

Figure 4.3: Cu target

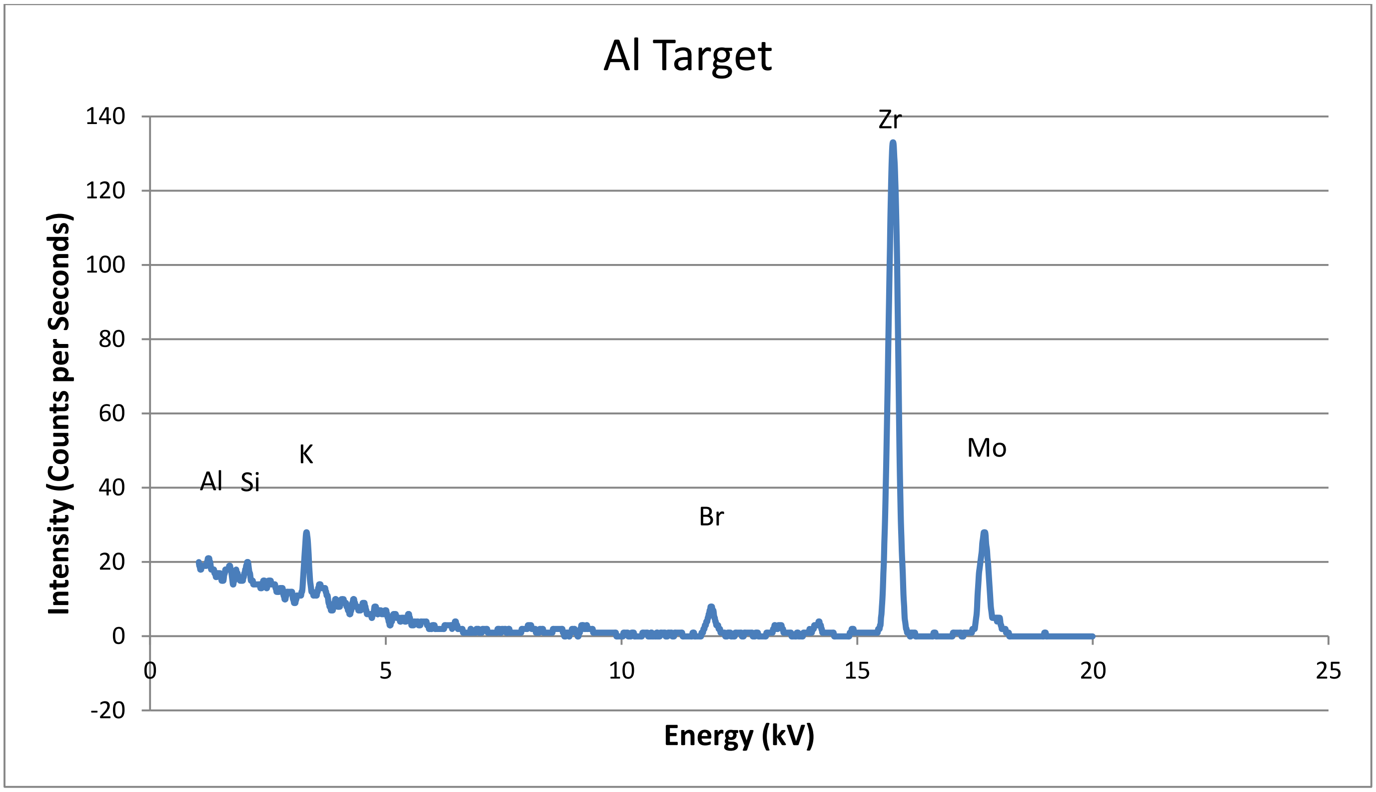

Figure 4.4: Al target

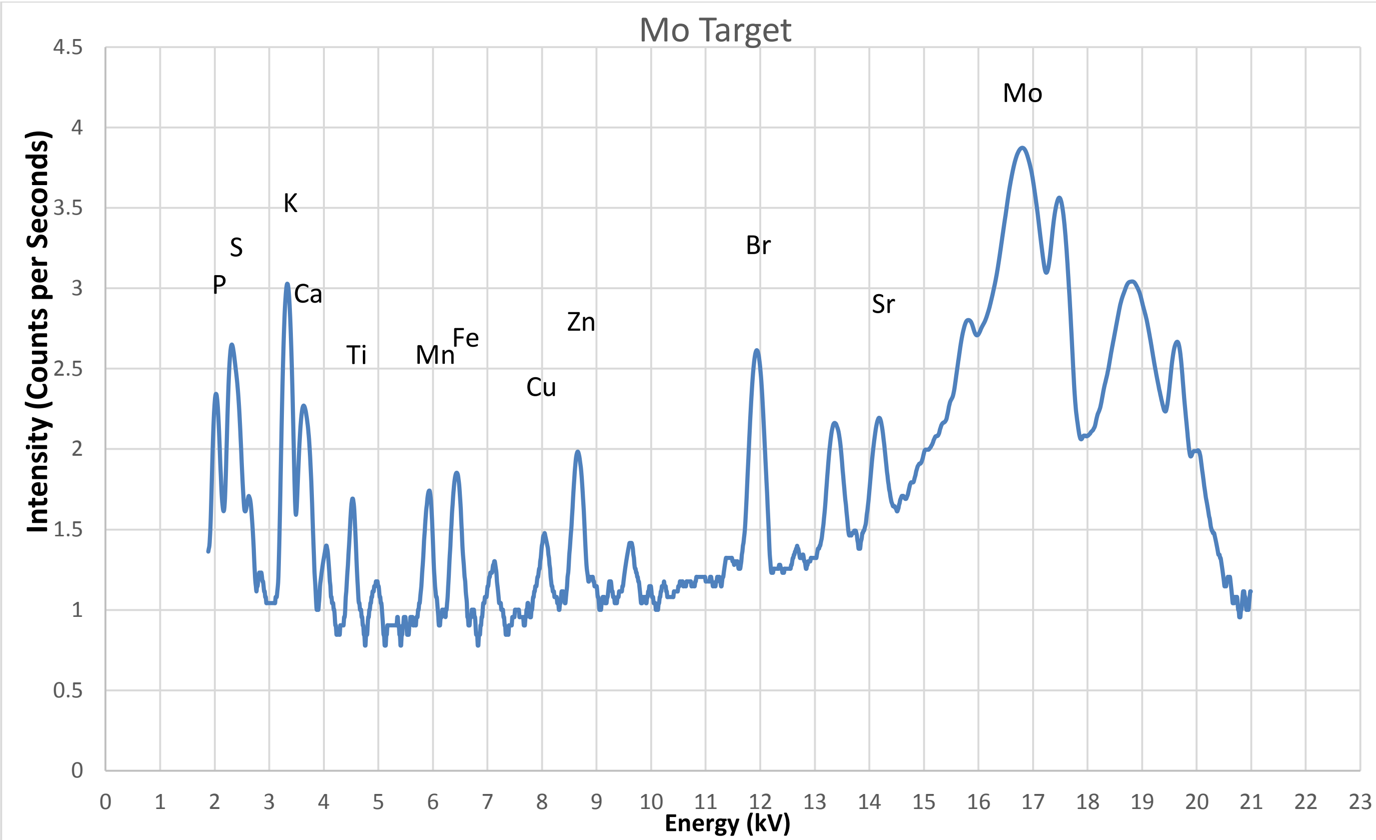

Figure 4.5: Mo target

For the Si target, potassium had the highest intensity; Rx9 target had Cl with the highest peak, while Cu and Zr were the highest intensities in the Cu and Al targets respectively. For the Mo target, most elements had relatively high peaks. The five targets were used interchangeably for optimum excitation conditions for the element of interest.

4.2 Comparison of results of two EDXRF Instruments: Rigaku and AMPTEK EXP-1

Table 4.1 shows the results of measurements of IAEA Bowen Kale Standard Reference Material using two EDXRF instruments for comparison, for the selected nutritional elements; potassium, calcium, manganese, iron and zinc.

Table 4.1: Elemental concentration (mg/Kg) of Bowen kale by two EDXRF instruments, n=3

Elements	Rigaku NEX CG	EXP-1 XRF	t-test values
K	24000 ±138	22000 ± 112	2.5
Ca	42000 ± 160	41000 ± 154	0.5
Mn	23.0 ± 3.0	15.0± 3.0	3.3
Fe	128 ± 5	120 ± 19	0.7
Zn	32.9 ± 1.4	30.0 ± 4.0	1.2

The t-test distribution table gives t-tabulated (t_{tab}) (α=0.05, 2) = 4.3 whereas the results obtained for t-calculated (t_{calc}) are less than 4.3. This showed that there was no significant difference in the results obtained from the two EDXRF instruments measurements of the sample. In this study, measurements were done using the Rigaku NEX CG, because multiple fifteen (15) samples were analysed simultaneously. This saves time on sample analyses to determine concentration levels.

4.3 EDXRF Method Validation for Accuracy

Table 4.2 shows the results of EDXRF analysis of certified reference Materials; Bowen Kale and Soil-5

Table 4.2: Experimental values of certified reference materials (mg Kg^{-1}; n=3)-

	Bowen kale			Soil-5 (IAEA- Soil-5)		
Element	Experimental values	Certified values	Rel. dev %	Experimental values	Certified values	Rel. dev %
Mn	23±3	15.0±1.7	55 %	1100±200	850±37	29%
Fe	130±5	115 ±15	7 %	51000±1100	45000±1700	14%
Zn	33.0±1.4	32.0±2.8	1.8 %	490±4	370±84	32%
Ca	42000±160	41000±2200	1.6 %	29000±1300	22000±1600	33%
K	24000±140	24000±1500	- 0.7%	23700±1200	19000±1000	27%

In general, the relative standard deviations (RSD) of the experimental values of the certified reference material were below 10% for all the elements in Bowen Kale except for Mn. For Soil-5, all the values are between 14% and 30 %. However, the experimental concentrations were within the standard reference certified values.

Cherop, (2011), reported similar findings for Bowen Kale using EDXRF spectrometer available at the Institute of Nuclear Science and Technology where the experimental values were within the range of certified values, thus showing that the method used was accurate, reliable and valid. He reported the concentrations of Mn, Fe and Zn as 15.2 ±1.2 $mgKg^{-1}$, 117±3$mgKg^{-1}$ and 31.2 ±2.1 $mgKg^{-1}$ respectively, which were within the certified range.

4.4 Essential Element Concentration in Beetroot Samples

Appendix 2-6 shows the tables of results of elemental concentration levels in beetroots from different geographical regions sampled for this study. Figure 4.6 shows the variation of elemental composition in beetroot samples from the various geographical regions.

There is a uniform trend in the concentrations of K, Ca and Fe in all the sampling sites, where K > Ca > Fe. However, for Mn and Zn, the trend of distribution in samples from Karatina, Naivasha and Kisumu is similar but significantly differs from those in Gilgil and Joska. The lowest concentration of Zn (15.5±1.2 $mgKg^{-1}$-34.1±3.3 $mgKg^{-1}$) was recorded in samples from Gilgil. This may be due to differences in soil types on which these beetroots are grown.

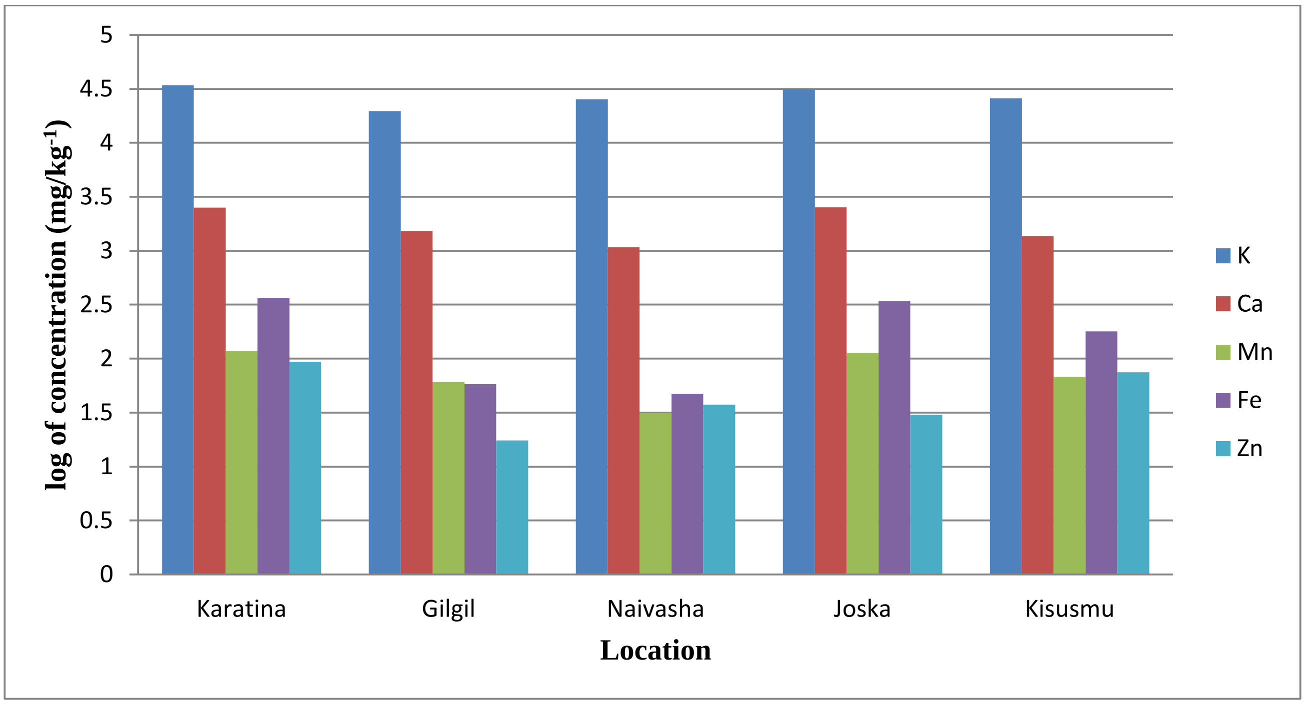

Figure 4.6: Trend in concentration of elements in beetroot from all the sites

In general, all the elements have high concentrations in Karatina samples in comparison to the other regions. This can be attributed to the fact that, Karatina is found in the Kenyan highlands where the soils are deep, have high content of clay, porous and have excellent capacity to hold moisture compared to the other regions. Karatina experiences high annual rainfall approximately 1432 mm thus suitable for planting throughout the year (Muchena & Gachene, 1988).

4.4.1 Potassium

Table 4.3 shows a summary of the results of ANOVA analysis of beetroot samples from the five sampling regions.

Karatina was found to have the highest concentration of K in all the samples with a mean of 34000 $mgkg^{-1}$. The results from ANOVA for all the five sampled regions show that there is no significant difference in the means since F-calculated $F_{(calc)}=2.5$ <F-tabulated $F_{(tab)}= 2.7$.

In general, the potassium mean concentration varies from 20000- 34000 $mgkg^{-1}$.

Table 4.3: ANOVA analysis for K in beetroot samples N=34, K=5

Statistical parameters	**Gilgil (n=9)**	**Karatina (=12)**	**Kisumu(n=2)**	**Naivasha(n=7)**	**Joska(n=4)**
Mean(ẋ)	20000	34000	26000	25000	31000
Variance(s^2)	56000000	47000000	10000000	31000000	170000000
d.f.N	4				
d.f.D	29				
$F_{(tab)}(\alpha=95\%)$	2.7				
grand mean	28000				
s^2B	290000000				
s^2W	120000000				
$F_{(calc)}$	2.5				

In the study of Betalainic and nutritional profiles of pigment-enriched red beet root (Beta vulgaris L.) dried extracts (Nemzer et al., 2011) using Inductively Coupled Plasma Atomic Emission Spectroscopy, it was reported that the potassium had an average concentration of 13730 $mgkg^{-1}$which was less than potassium reported in this study.

The concentration of potassium from the study of Nutritional value and economic feasibility of red beetroot (Beta *vulgaris* L. ssp. *vulgaris* Rote Kugel) from different production systems (Straus et al., 2012), was reported to have an average of 33,766 mg kg^{-1}, which is within the range of what was found in this study. Beetroot contains sufficient potassium levels to contribute to daily dietary recommended intakes as a supplement.

4.4.2 Calcium

The results obtained from the analysis of ANOVA on beetroot samples from different regions for the calcium content are presented in table 4.4. Karatina was found to have the highest concentration of Ca in all the samples with a mean of 2800 $mgkg^{-1}$. The results show that there is a slight significant difference in the means since $F_{(calc)}$6.55> $F_{(tab)}$ 2.7.

Table 4.4: ANOVA analysis for Ca in beetroot samples N=34, K=5

Statistical parameters	**Gilgil (n=9)**	**Karatina(n=12)**	**Kisumu(n=2)**	**Naivasha(n=7)**	**Joska(n=4)**
Mean(X)	1900	2800	1400	1200	2700
Variance(s^2)	620000	500000	100000	390000	820000
d.f.N	4				
d.f.D	29				
$F_{(tab)}$(α=95%)	2.7				
grand mean	2100				
s^2B	3500000				
s^2W	530000				
$F_{(calc)}$	6.5				

Campbell (2000), carried out a study of trace elements sufficiency ranges in various vegetables that included carrots, broccoli, Muskmelon, spinach and tomatoes where the percentage concentrations were 2-3%, 1-2%, 3-5%, 1-2% and 1-3% respectively. The Ca levels from this study were lower than those reported by Campbell.

Fahad et al., (2015) compared the accumulation of calcium in different vegetables where he reported the following; lettuce 95213 ± 101 $mgKg^{-1}$, spinach 86606 ± 102 $mgKg^{-1}$, hyacinth bean 84615 ± 79 $mgKg^{-1}$ and cauliflower 80743 ± 87 $mgKg^{-1}$. The values for the concentration of Ca in the different vegetables were all higher than those reported for concentration of Ca in beetroot bulbs in this study.

4.4.3 Manganese

Manganese is one of the essential nutrients in the human body, with some of the key functions including regulation of blood sugar levels, assisting in proper functioning of the thyroid glands, formation of connective tissue, bone formation and absorption of calcium (Kazi et al., 2008)

Table 4.5 presents results obtained for the analysis of ANOVA on beetroot samples from five different regions for the analysis of Mn. Karatina was found to have the highest concentration of Mn in all the samples with a mean of 130 $mgkg^{-1}$ The results show that there is a slight difference in the means since $F_{(calc)}$ (5.88) >$F_{(tab)}$ (2.7)

Table 4.5: ANOVA analysis for Mn in beetroot samples N=34, K=5

Statistical parameters	**Gilgil (n=9)**	**Karatina(n=12)**	**Kisumu(n=2)**	**Naivasha(n=7)**	**Joska(n=4)**
Mean(X)	78	130	68	36	120
Variance(s^2)	1400	3200	8500	480	770
d.f.N	4				
d.f.D	29				
$F_{(tab)}$(α=95%)	2.7				
grand mean	91				
s^2B	11000				
s^2W	1800				
$F_{(calc)}$	6				

In Campbell's work, (2000), the Mn levels were 30-60 mgkg^{-1}, 25-150 mgkg^{-1}, 20-100 mgkg^{-1}, 25-200 mgkg^{-1}and 25-200 mgkg^{-1} for carrots, broccoli, Muskmelon, spinach and tomatoes respectively. The Mn levels from this study were within the range reported by Compbell for different vegetables.

Moyo et al., (2017), compared the concentration of Mn in Cleome *gynandra* (spider plant) and Beta *vulgaris* L (leaf vegetable known as chard or spinach beet) and found that Beta *vulgaris* (57 μg/g d.w) had 1.5 higher concentration of manganese than Cleome *gynandra* (38 μg/g d.w) The Mn levels reported in this study are higher than those reported by Moyo et al.

Odhav et al. (2007) determined manganese levels in some sampled vegetables, Amaranthus *dubius*, Amaranthus *hybridus*, Amaranthus *spinosus*, Asystasia *gangetica*, Cleome *monophylla* and Oxygonum *sinuatum*. It was reported that the concentrations of Mn were ranging from 4 mg/100g- 82 mg/100g. These concentrations were within the range of manganese concentrations reported in this study.

Manganese concentrations reported in this study were within the range of those reported by Ekholm et al (2007) in a study carried out in Finland to analyse changes in the mineral and trace element contents of cereals, fruits and vegetables. It was reported that the concentration of manganese in different root vegetables as; potato 0.7 mg/100 g, organic carrot 1.2 mg/100 g, beetroot 3 mg/100 g, parsnip 1 mg/100 g, celery root 1.5 mg/100 g, turnip 1mg/100 g, swede 1.1 mg/100 g, radish 1 mg/100 g and Jerusalem artichoke 0.2 mg/100 g.

The daily recommended intake for manganese is 2-5 mg/day and 2-3 mg/day for adults and children respectively (WHO, 2005), the Mn content in beetroots is 68- 126 mg kg^{-1}, hence the plant can be used as a manganese supplement.

4.4.4 Iron

The results obtained for the analysis of ANOVA on beetroot samples from different regions for the Fe content are presented in table 4.6. The results show that there is a significant difference in the means since $F_{(calc)}=14.22>F_{(tab)}=2.7$. Iron was found to be most abundant in samples from Karatina with a mean concentration of 420 $mgkg^{-1}$. The high iron concentration could be attributed to high background concentration of iron since the soil is reddish brown clay. A study by Galgallo, (2015), reported high Fe concentrations in a model farm in Central Kenya with mean Fe concentration of 5.6 ±4%. Since Karatina is in Central Kenya, the high Fe concentration found in beetroots can be attributed to its growing area.

Table 4.6: ANOVA analysis for Fe in beetroot samples N=34, K=5

Statistical parameters	**Gilgil (n=9)**	**Karatina(n=12)**	**Kisumu(n=2)**	**Naivasha(n=7)**	**Joska (n=4)**
Mean(X)	78	420	180	53	370
Variance(s^2)	1700	33000	14000	760	27000
d.f.N	4				
d.f.D	29				
$F_{(tab)}$(α=95%)	2.7				
grand mean	230				
s^2B	230000				
s^2W	16000				
$F_{(calc)}$	14				

The iron content in beetroot samples in this study are higher than those reported in a study by Tidemann-Andersen et al. (2011) , on iron and zinc content of selected foods in the diet. They reported a mean of 50 $mgKg^{-1}$ in cassava, 140 $mgKg^{-1}$ in Irish potatoes and 56 $mgKg^{-1}$ in sweet potatoes.

In a survey of plant iron content, Ancuceanu et al. (2015) determined the mean Fe content for roots to be 225 $mgKg^{-1}$, 74.3 $mgKg^{-1}$ for barks, 195.4 $mgKg^{-1}$ for fruits and 333 $mgKg^{-1}$ for seeds, all lower than most of the beetroot values reported in this current study.

Petry et al. (2015), analysed beans from Cali, Colombia for Fe content, where a mean concentration of 71 µg/g - 280 µg/g was reported and the values are lower than those reported for beetroot levels in this study.

Ekholm et al. (2007), carried a study in Finland to analyse changes in the mineral and trace element contents of cereals, fruits and vegetables. They reported the concentrations of iron in different root vegetables as; potato 3.4 mg/100 g, organic carrot 3.4mg/100 g, beetroot 2.9 mg/100 g, parsnip 3 mg/100 g, celery root 5.5 mg/100 g, turnip 4 mg/100 g, Swede 3.4 mg/100 g, radish 6.5 mg/100 g and Jerusalem artichoke 1.8 mg/100 g. These concentrations are lower than beetroot Fe concentrations in this study.

Iron is an essential nutrient in the body and plays an important role in transporting oxygen and carbon dioxide in the blood, preventing and treating anaemia, reducing heart failure and improving thinking and memory (Watson, 2011). The dietary requirement of Fe varies with age and sex. Boys and girls need the same amount of iron10 mg/ day from ages 4 to 8, and 8 mg/ day from ages 9 to 13. Women need more iron from ages 19 to 50 because they lose blood each month during their periods, that is why they need to get 18 mg of iron per day, while men of the same age can get 8 mg/day (Beck et al., 2014). Since beetroots have a mean range of iron concentration of 53-416 $mgKg^{-1}$ they can be a good source of iron to meet the recommended intake.

4.4.5 Zinc

Table 4.7 presents the results obtained for the analysis of ANOVA on beetroot samples from different regions for the zinc content. Karatina was found to have the highest concentration of Zn in all the samples with a mean of 120 $mgkg^{-1}$. The results show that there is no significant difference in the means since $F_{(calc)}$=1.13< than $F_{(tab)}$ =2.7.

Table 4.7: ANOVA analysis for Zn in beetroot samples N=34, K=5

Statistical parameters	Gilgil (n=9)	Karatina(n=12)	Kisumu(n=2)	Naivasha(n=7)	Joska(n=4)
Mean(X)	23	120	74	43	32
Variance(s^2)	38	32000	2900	4400	110
d.f.N	4				
d.f.D	29				
$F_{(tab)}$(α=95%)	2.7				
grand mean	64				
s^2B	14000				
s^2W	12000				
$F_{(calc)}$	1.10				

In the evaluation of black tea Brzezicha-Cirocka et al. (2017) reported the Zn concentration range as 0.2-1.5 mg kg^{-1} which are low compared to levels of beetroot reported in this study. Basha et al. (2014), conducted a study on vegetables and fruits cultivated around the surroundings of Tummalapalle Uranium mining site. They reported a zinc concentration of 4.1±0.8 mg Kg^{-1} for tomatoes, 8.5±6.5 mg $Kg^{-1}$for green chilli, 5.2±0.8 mg Kg^{-1} for bitter gourd, 4.5±2.9 mg $Kg^{-1}$for bananas, 7.3±5.1 mg Kg^{-1} for papaya and 9.2 ± 4.2 mg $Kg^{-1}$for melons. These concentrations are low compared to those for beetroot samples in this study.

Odhav et al. (2007) determined zinc levels in Amaranthus *dubius*, Amaranthus *hybridus*, Amaranthus *spinosus*, Asystasia *gangetica*, Cleome *monophylla* and Oxygonum *sinuatum*, and reported the concentrations of Zn as ranging from 5 mg/100g- 56 mg/100g. These concentrations are higher than the Zn concentration levels in beetroot reported in this study.

Ekholm et al. (2007) carried a study in Finland to analyse changes in the mineral and trace element contents of cereals, fruits and vegetables. They reported the concentration of zinc in different root vegetables as; potato 1 mg/100 g, organic carrot 2.1 mg/100 g, beetroot 1.6 mg/100 g, parsnip 2.1 mg/100 g, celery root 3.7 mg/100g, turnip 2.4 mg/100 g, swede 1.9 mg/100 g, radish 3.1 mg/100 g and Jerusalem artichoke 1.4 mg/100g. These concentrations are lower than the Zn concentrations reported in this study.

The daily dietary requirement for zinc is 4.2 – 14 mg/day (WHO, 2005), the zinc content in beetroot is 22 – 115 $mgKg^{-1}$, thus it can be concluded that beetroot is a very good source of zinc and can be used as a supplement.

4.4.6 Comparison of essential elements variation in Vegetables

For comparison purposes, potatoes bulbs, Moringa leaves and carrots bulbs were analysed and their results compared to beetroot as shown in Table 4.8:

Table 4.8: Elemental concentration of selected vegetables in mg Kg^{-1} n=3

Type	Mn	Zn	Fe	Ca	K
Beetroot	91.0 ± 15.0	64.0 ± 8.3	230 ± 27	2100 ± 250	32000 ± 2600
Potato	15.0 ± 2.4	130 ±10	53.0 ± 3.0	420 ± 38	28000 ± 2500
Moringa	89.0 ± 7.2	24.0 ± 2.1	370 ± 27	19000 ± 1100	24000 ± 2140
Carrot	29.0 ± 5.2	31.0 ± 2.7	140 ± 13	6800 ± 440	44000 ± 3500

From the results in Table 4.8, the concentrations in beetroots are high for all the elements except calcium where Moringa has eight times higher concentration and carrots has three times higher calcium concentration than beetroot. In addition, the concentration of iron for Moringa is higher than that of beetroot. Thus, we can conclude that beetroot is a good source of K, Ca, Mn, Fe and Zn.

4.5 Beetroot pH

The beetroot solution pH was determined to average at 8.9 ± 0.2. This value is within the range of the recommended pH value of a general diet. It is recommended to consume basic foods, which are of pH 8 and above because they prolong life and stave off diseases (Schwalfenberg, 2012). Beetroot can also be used as an acid-base indicator. This can be explained by beetroots containing pigments of red and yellow known as betacyanins and betaxanthins, respectively (Herbach et al., 2004).

CHAPTER FIVE: CONCLUSION AND RECOMMENDATIONS

5.1 Conclusion

Cases of malnutrition and nutritional related diseases have been on the rise in Kenya, thus bringing forth the need for research to be carried out on assessing the nutritional value of different vegetables, fruits and food crops in general that constitute a diet.

This study determined the levels and variations of essential elements in beetroots sampled from Karatina, Gilgil, Naivasha, Joska-Machakos and Kisumu. The elemental concentrations were measured using Energy Dispersive X-ray Fluorescence spectrometer.

In the study, the pH of beetroot was found to be 8.87 which is basic. The results of the concentration of the essential elements had a range of: 11000 mg kg^{-1} to 50000 mg kg^{-1} for potassium, 94.0 mg kg^{-1} to 4500 mg kg^{-1} for calcium, 36.0 mg kg^{-1} to 230 mg kg^{-1} for manganese, 12.0 mg kg^{-1} to 770 mg kg^{-1} for iron and 14.0 mg kg^{-1} to 680 mg kg^{-1} for zinc. In general, the trend in the concentration of the essential elements was K >Ca > Fe >Mn > Zn. Karatina samples registered the highest concentrations for all the elements of interest in the study

Beetroot was found to contain high levels of K, Ca, Mn, Fe and Zn. The vegetable was found to be a good source of essential elements.

5.2 Recommendations

1) Since beetroots have sufficient levels of essential elements they should be incorporated in our diets.
2) More research should be conducted on the whole plant to include the essential elemental concentration of the beetroot leaves and stem.
3) Soil samples where the beetroot is harvested should also be assessed for any correlation of the essential elements in the soil to those elements found in the plant.
4) Research to determine the optimum growing conditions and age of harvesting the beetroot bulbs in terms of bio-accumulation of essential elements.

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APPENDICES

Appendix 1: Global Positions of the Sampling Sites

Sample code	**Location**	**Elevation (m)**	**X**	**Y**
K1	Karatina	1746	37° 09.217 E	0° 29.142 S
K2	Karatina	1746	37° 09.217 E	0° 29.146 S
K3	Karatina	1743	37° 09.228 E	0° 29.143 S
K4	Karatina	1743	37° 09.231 E	0° 29.146 S
K5	Karatina	1743	37° 09.229 E	0° 29.154 S
K6	Karatina	1745	37° 09.225 E	0° 29.154 S
K7	Karatina	1744	37° 09.231 E	0° 29.152 S
K8	Karatina	1745	37° 09.233 E	0° 29.152 S
K9	Karatina	1745	37° 09.225 E	0° 29.154 S
K10	Karatina	1746	37° 09.218 E	0° 29.145 S
K11	Karatina	1747	37° 09.217 E	0° 29.148 S
K12	Karatina	1742	37° 09.219 E	0° 29.143 S
G1	Gilgil	2036	36° 17.557 E	0° 25.974 S
G2	Gilgil	2037	36° 17.556 E	0° 25.975 S
G3	Gilgil	2036	36° 17.555 E	0° 25.973 S
G4	Gilgil	2037	36° 17.552 E	0° 25.975 S
G5	Gilgil	2035	36° 17.561 E	0° 25.970 S
G6	Gilgil	2034	36° 17.558 E	0° 25.967 S
G7	Gilgil	2034	36° 17.557 E	0° 25.684 S
G8	Gilgil	2036	36° 17.551 E	0° 25.974 S
G9	Gilgil	2038	36° 17.554 E	0° 25.971 S
N1	Naivasha	2477	36° 32.432 E	0° 47.260 S
N2	Naivasha	2474	36° 32.429 E	0° 47.264 S
N3	Naivasha	2475	36° 32.436 E	0° 47.264 S
N4	Naivasha	2482	36° 32.431 E	0° 47.275 S
N5	Naivasha	2483	36° 32.428 E	0° 47.294 S
N6	Naivasha	2485	36° 32.438 E	0° 47.296 S
N7	Naivasha	2483	36° 32.438 E	0° 47.286 S
J1	Joska	1490	37° 05.668 E	01° 17.035 S
J2	Joska	1484	37° 05.666 E	01° 17.033 S
J3	Joska	1486	37° 05.664 E	01° 17.035 S
J4	Joska	1491	37° 05.667 E	01° 17.034 S
Kisumu1	Kisumu	1131	34° 45.093E	0° 06.276 S
Kisumu2	Kisumu	1132	35° 45.081E	0° 06.274 S

Appendix 2: Essential Element Concentration in Karatina (mg kg^{-1}), n=3

Sample code	Mn	Fe	Zn	Ca	K
K1	64.1 ± 2.5	350 ± 26	680 ± 84	2500 ± 210	38000 ± 2400
K2	59.0 ± 5.1	710 ± 33	29.0 ± 2.4	2600 ± 230	33000 ± 2300
K3	84.0 ± 6.0	350 ± 20	39.8 ± 3.2	2900 ± 197	49000 ± 3500
K4	83.0 ± 4.0	260 ±25	43.0 ± 3.0	2000 ± 175	32000 ± 2900
K5	60.0 ± 4.0	350 ± 20	72.0 ± 6.0	2500 ± 192	33000 ± 2500
K6	170 ± 16	630 ± 48	80.0 ± 6.0	3600 ± 340	32000 ± 2500
K7	150 ± 14	410 ± 36	73.0 ± 5.0	3000 ± 347	27000 ± 2420
K8	230 ± 11	770 ± 53	100 ± 12	4500 ± 370	44000 ± 3780
K9	170 ± 11	250 ± 22	90.0 ± 10.0	3000 ± 160	36000 ± 2200
K10	110 ± 17	290 ± 21	53.0 ± 4.0	2000 ± 140	28000 ± 2100
K11	170 ± 13	290 ± 20	65.0 ± 5.0	2300 ± 158	28000 ± 2640
K12	160 ± 19	350 ± 47	65.0 ± 7.0	2400 ± 220	28000 ± 2400
AVERAGE	**130**	**420**	**120**	**2800**	**34000**
MIN	**59.0 ± 5.1**	**250 ± 22**	**29 ± 2.4**	**2000 ± 175**	**27000 ± 2420**
MAX	**230 ± 11**	**770 ± 53**	**100 ± 12**	**4500 ± 370**	**49000 ± 3500**

Appendix 3: Essential Element Concentration in Gilgil (mgkg^{-1}), n=3

Sample code	K	Ca	Mn	Fe	Zn
G1	13000 ± 1800	920 ± 83	36.0 ± 5.0	35.0 ± 3.5	16.0 ± 1.1
G2	23000 ± 2300	3200 ± 320	160 ± 14	150 ± 18	29.0 ± 2.3
G3	14000 ± 1900	1100 ± 151	50.0 ± 3.5	110 ± 1.2	19.0± 1.2
G4	33000 ± 2300	2700 ± 160	110 ± 13	110 ± 12	34.0 ± 3.3
G5	23000 ± 2800	2400 ± 250	85.0 ± 7.2	45.0 ± 4.5	18.0 ± 1.2
G6	10000 ± 1600	1500 ± 160	58.0 ± 5.1	39.0 ± 3.6	16.0 ± 1.1
G7	27000 ± 2100	1900 ± 140	50.0 ± 4.7	110 ± 18	24.0 ± 2.4
G8	19000 ± 1100	2300 ± 280	84.0 ± 7.5	65.0 ± 4.6	26.0 ± 2.4
G9	16000 ± 1790	1200 ± 164	72.0 ± 7.2	45.0 ± 4.1	20.0 ± 2.1
AVERAGE	**20000**	**2300**	**66**	**120**	**21**
MIN	**10000 ± 1600**	**920 ± 83**	**36.0 ± 5.0**	**35.0 ± 3.5**	**16.0 ± 1.1**
MAX	**33000 ± 2300**	**3200 ± 320**	**160 ± 14**	**150 ± 18**	**34.0 ± 3.3**

Appendix 4: Essential Element Concentration in Naivasha ($mgkg^{-1}$), n=3

Sample code	**K**	**Ca**	**Mn**	**Fe**	**Zn**
N1	18000 ± 1800	570 ± 43	15.0 ± 1.5	30.0 ± 2.5	24.0 ± 2.1
N2	61000 ± 5300	2400 ± 320	80.0 ± 7.4	100 ± 18	84.0 ± 7.3
N3	34000 ± 2900	1500 ± 151	42.0 ± 3.9	56.0 ± 5.2	58.0 ± 4.2
N4	10000 ± 1300	500 ± 46	18.0 ± 1.3	24.0 ± 2.1	38.0 ± 3.3
N5	25000 ± 2800	1300 ± 150	24.0 ± 1.2	50.0 ± 4.5	36.0 ± 2.2
N6	15000 ± 1600	1300 ± 160	33.0 ± 3.1	76.0 ± 6.6	31.0 ± 3.1
N7	14000 ± 1000	1000 ± 160	41.0 ± 3.7	36.0 ± 3.4	32.0 ± 2.4
AVERAGE	**25000**	**1200**	**36**	**53**	**43**
MIN	**10000 ± 1300**	**500 ± 151**	**15.0 ± 1.5**	**24.0 ± 2.1**	**24.0 ± 2.1**
MAX	**61000 ± 5300**	**2400 ± 320**	**80.0 ± 7.4**	**100 ± 18**	**84.0 ± 7.3**

Appendix 5: Essential Element Concentration in Joska ($mgkg^{-1}$), n=3

Sample code	**K**	**Ca**	**Mn**	**Fe**	**Zn**
Stall 1	33000 ± 3210	3100 ± 250	160 ± 11	310 ± 31	37.0± 3.2
Stall 2	30000 ± 2400	2100 ± 170	110 ± 13	360 ± 31	24.0 ± 2.3
Stall 3	15000 ± 1230	1800 ± 130	98.0 ± 8.3	220 ± 23	23.0 ± 1.6
Stall 4	47000 ± 4500	3800 ± 350	120 ± 13	600 ± 54	44.0 ± 4.2
Average	**310000**	**2700**	**120**	**370**	**32**
MIN	**15000 ± 1230**	**1800 ± 130**	**98.0 ± 8.3**	**220 ± 23**	**23.0 ± 1.6**
MAX	**47000 ± 4500**	**3800 ± 350**	**160 ± 11**	**600 ± 54**	**44.0 ± 4.2**

Appendix 6: Essential Element Concentration in Kisumu ($mgkg^{-1}$), n=3

Sample code	K	Ca	Mn	Fe	Zn
Kisumu 1	24000 ± 1800	1100 ± 78	47.0 ± 4.1	95.0 ± 8.5	110 ± 13
Kisumu 2	28000 ±2600	1600 ± 150	89.0 ±7.5	260 ± 24	36.0 ± 2.7
Average	**26000**	**1400 ± 123**	**68**	**180**	**74**
MIN	**24000 ± 1800**	**1100 ± 78**	**47.0 ± 4.1**	**95.0 ± 8.5**	**36.0 ± 2.7**
MAX	**28000 ± 2600**	**1600 ± 150**	**89.0 ± 7.5**	**260 ± 24**	**110 ± 13**