SOIL-PLANT COMPOSITION IN RELATION TO MACRO AND TRACE ELEMENT REQUIREMENTS FOR GRAZING CATTLE IN UASIN GISHU DISTRICT

BY

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DECLARATION

I declare that the work presented in this thesis is my original work and has never been presented for a degree in any other University. This thesis has been submitted with approval of the University Supervisors:

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DEDICATION

I dedicate this thesis to my wife, Margaret and children, Osteen and Anjela.

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ABBREVIATIONS

AAS..... Atomic Absorption Spectrophotometer

ATP..... Adenosine Triphosphate

CMPT.....Compton Metabolic Profile Test

EDTA .....Ethylene Diamine Tetraacetic Acid

DIW.....De-ionised Water

DM.....Dry Matter

DNA .....Deoxy nucleic Acid

GDP.....Gross Domestic Product

GLM.....Generalized Linear Model

GPS .....Global Mapping System

GSH-Px ...Glutathione Peroxidase

KARI .....Kenya Agricultural Research Institute

MMA .....Methyl Malonic Acid

NMD .....Nutritional Mascular Dystrophy

RNA .....Ribonucleic Acid

TCA .....Trichloroacetic Acid

ABSTRACT

This study was carried in the six divisions of Uasin Gishu district in which twenty-eight (28) soils, twenty-eight (28) forage and forty-two (42) serum samples were collected at different sites. The study was necessary since the region had grazing cattle experiencing reduced growth, anaemia, reduced conception rates and fertility, which are symptoms of mineral deficiencies. Sampling sites were classified in terms of the forage species most favoured for grazing by the available cattle and age and breed of the grazing cattle. The purpose of the study was to determine the status of both macro- and trace elements in forages and compare them with those in grazing cattle and the soils on which forage grow, so as to identify those that might be limiting animal health and nutrition within the district. Soils were analyzed for extractable macro minerals; sodium (Na), potassium (K), calcium (Ca), magnesium (Mg) and trace elements; iron (Fe), manganese (Mn), copper (Cu), and zinc (Zn). The forage samples were assayed for the same elements plus phosphorus (P) as total concentration on dry matter (DM) basis while blood serum was analyzed for the same forage elements plus molybdenum (Mo). Atomic absorption spectrophotometer (AAS) and UV/Visible spectrophotometer (UV/V) were used to analyze metal elements and nonmetals respectively. Forage analysis revealed wide variations in mineral element concentrations. The mean values were as follows: Na-1.00 ± 0.30); K-11.8 ± 5.00; Ca-0.57 ± 0.19); Mg-1.35 ± 0.72; P-6.34 ± 3.22 in g/kg DM; Fe-56± 0.53; Mn-105 ± 0.58); Cu-5.32 ± 2.84 and Zn-19.50 ± 8.26 in mg/kg DM. The results revealed acute deficiencies of some mineral elements in forages. The percentage of forage samples with deficient elements was: 93(Na), 89(Mg), 93(Cu) and 39(Zn). Grazing cattle and soils revealed similar deficiencies in some minerals. The percentage of soil samples with deficient elements was: 14(Cu) and 4(Zn). The percentage of calves' serum samples with deficient elements was: 50(Mg) and 61(Cu) while the percentage of cows' serum deficient in the same minerals was 21(Mg) and 42(Cu). In other cases, cattle revealed further deficiencies in some minerals, which were quite different from those detected in forages. Soil pH and mineral interrelationships within samples were factors influencing mineral concentrations in the region. The soil pH range 5.1-7.2 was optimal for iron and copper concentrations. Correlation analysis of the results indicated that all the elements except calcium and copper revealed a small but positive relationship between soil and forage mineral concentrations. Rhodes/Kikuyu grass mixture gave the highest mineral concentration whose difference was significant ($P<0.05$) as compared to other grass species (Rhodes (*Chloris gayana*), Kikuyu (*Pennisetum Clandestinum*) and Natural grass). Friesian calves and lactating cows had a better mineral content ($P<0.05$) than either Ayrshire or Ayrshire/Friesian crossbreed. Recommendations on supplementation to animals have been suggested to help identify and alleviate any constraints on animal health and production.

CHAPTER ONE

INTRODUCTION

1.1 BACKGROUND

Livestock industry is one of the major contributors to the country's GDP and provides one third of the total available food in the country (NDP, 2000). Cattle in particular are reared for milk and meat production. Grazing cattle in the tropics and especially in Uasin Gishu district usually depend wholly on a variety of plant species for their nutrient supply. Any insufficient supply of minerals by the forages result in mineral imbalance, which limit animal production. However, with better nutrition management, improved production can be realized (MFP, 2001; Rojas *et al.*, 1993). One area of concern in animal grazing countrywide is to provide a balance in minerals requirements.

Grazing cattle derive their minerals from a variety of plant species. Plants in turn derive their mineral elements from the soil in which they grow and the amount is related to the underlying rock from which soils are formed (Jumba *et al.*, 1995a, b). This makes plants to be important immediate reservoirs through which minerals from soils reach animals. Studies have revealed that grazing cattle in the tropics do not usually receive mineral supplements except common salt and thus depend wholly on the forages to meet their nutritional mineral requirements (Jumba, 1989; Oduor, 2002). Apart from other dietary factors such as protein, fibre, energy (carbohydrates and fats), and vitamins, mineral deficiency may also limit animal production (McDowell *et al.*, 1993). The effects of these mineral imbalances in the tropics are manifested in such cases as low fertility, bone abnormalities, nutritional muscular dystrophy (NDP, 2000), retarded growth and

maturity, hair disorders, and low meat and milk production (Faria *et al.*, 1981). Increased milk yields by 7-260 % and meat production by 6-400 %, improved calving percentage of 20-100 % and growth rates by 10-25 % have been reported from mineral supplementation of cattle diets (McDowell and Conrad, 1977). This shows that monitoring growth and health responses after administration of certain supplements to the affected animal can help diagnose and determine mineral deficiencies.

Studies in grazing areas of Mt. Elgon region of western Kenya have shown wide variations in soils and forage mineral composition (Jumba, 1989; Oduor, 2002) as compared to similar surveys in other tropical countries such as Dominican Republic (Jerez *et al.,* 1984), Columbia (Pastrana *et al.*, 1991b), Guatemala (Valdez *et al.*, 1988), North Florida (Cuesta *et al.,* 1993) and South Eastern Venezuela (Rojas *et al.*, 1993). Soil-plant mineral interactions have been used to provide information on mineral interrelationships, which are then used to predict mineral levels in animals (Jumba *et al.*, 1995a, b; Siva, 1996). However, other studies have revealed that the status, age and breed of the animal influence mineral status in animals, hence broadening the scope of study from soil-plant to soil-plant-animal interactions (Oduor, 2002). Samples such as animal blood, hair, liver or bone specimens have been used to determine mineral status in animals (Kariuki, 2000; Oduor, 2002). Such a soil-plant-animal investigation system may reduce supplementation schemes to fewer and more specific minerals thus eliminating problems associated with direct mineral supplementation in animals in which certain minerals are in excess.

Uasin Gishu district is one of the potential cattle production regions in Kenya. Situated in Rift Valley, it borders Trans Nzoia and Mt. Elgon districts. It is a region that used to produce high quantities of meat and milk and related products, a trend that is declining (MFP, 2001). It comprises of six divisions, each recording at least one or more of cattle related complications such as poor conception rates, retained placenta, poor coat condition, skin disorders and retarded growth (MALD, 2003). Studies in Rift Valley have shown certain mineral deficiency problems in some areas (Siva, 1996). In the neighbouring Trans Nzoia and Mt. Elgon districts, studies have revealed severe nutritional problems of Ca, P, S and Cu in the forages and grazing cattle (Jumba, 1989; Oduor, 2002). The problems have partly been attributed to herbage species differences as the principal cause of variations in mineral uptake and retention. The studies revealed deficiencies of some minerals in soils and forages, some of which correlated well in both young growing calves and lactating cows. The studies noted that seasonal changes might have an impact on mineral status with severity of the impact varying with moisture availability and altitude. It is anticipated that Uasin Gishu district may also be experiencing the same nutritional deficiencies detected in Trans Nzoia and Mt. Elgon districts. Improvement of milk and meat production can be realized if mineral deficiency problems in the district are determined and alleviated.

1.2 STATEMENT OF THE PROBLEM

An imbalanced supply of minerals to livestock can lead to problems in grazing livestock. Investigations have revealed that Uasin Gishu district has farmers engaged in cattle production, which suffers setbacks as poor conception rates, retained placenta, mastitis

incidence, neonatal weakness, poor coat condition, colour and skin disorders. These are all related to mineral deficiency.

1.3 HYPOTHESIS

Grazing cattle in Uasin Gishu district are experiencing deficiencies in some mineral elements.

1.4 OBJECTIVES

1.4.1 General objective

To determine the mineral deficiencies experienced by grazing cattle in Uasin Gishu district.

1.4.2 Specific objectives

1. To determine the mineral element status of soils, forages and cattle from Uasin Gishu district.
2. To determine the factors that might be influencing mineral availability to grazing cattle in the district.

1.5 JUSTIFICATION AND SCOPE OF THE STUDY

Minerals play an important role in the bodies of animals. Macro elements are necessary for acid base balance (Na, K), transmission of nerve impulses (Ca), enzyme activity (Mg) and buffering of blood in the animal body (McDowell and Conrad, 1989). Trace elements on the other hand are important components of enzymes such as catalase (Fe),

ceruloplasmin (Cu), arginase (Mn and Zn), glutamate mutase (Co) and glutathione peroxidase (Se). Deficiency of these minerals in animal feed can result in low animal production. Therefore continued monitoring of minerals is necessary to ensure proper nutrition for the animals.

The study focused on the macro elements; Na, K, Ca, Mg, P, as well as the trace elements; Fe, Mn, Cu and Zn in soils, pastures and animal blood. In addition, pasture was analyzed for phosphorus while animal serum was analyzed for phosphorus and molybdenum. The mineral elements were selected based on the symptoms reported in grazing cattle from Uasin Gishu region, which are related to their deficiencies (MALD, 2003). Such deficiency symptoms were anaemia (Fe), reduced conception rates (Mn, Cu, Zn), reduced milk production (Na), nervous disorders (K, Mg), skeletal and dental abnormalities (Ca, P).

1.6 LIMITATIONS OF THE STUDY

The analysis of metal elements was only done using atomic absorption spectrophotometer that was available. Selenium analysis was not done due to unavailability of fluorospectrophotometer.

The study area was divided into three regions: the northern, the central and the southern region (Oduor, 2002). The northern region borders Trans Nzoia, which reported Cu, Zn, Fe, P, Mg and Ca deficiencies in cattle (Oduor, 2002). It consists of Soy and Moiben divisions. The central region, which is around Eldoret town, covered mainly Kapsaret division while the southern region covered Ainabkoi division.

The findings of the study are presented in five chapters. Chapter One gives a general introduction, the objectives of the study, justification for choice of study area and the scope. Chapter Two looks at the importance of minerals in livestock production and the assessment of their deficiencies in grazing cattle. Chapter Three focuses on the materials and methods used to obtain the results in Chapter Four while Chapter Five gives the conclusions and possible recommendations for the alleviation of these problems in the study area.

CHAPTER TWO

LITERATURE REVIEW

2.1 CLASSIFICATION OF MINERAL ELEMENTS

Mineral deficiencies and imbalances in soils and forages, apart from protein and energy availability, have been found to be responsible for the low production and reproduction in grazing cattle in developing countries (McDowell *et al.*, 1983). An essential mineral is one that is present in all healthy tissues of living things in constant levels and whose deficiency consistently leads to an impairment of a function from optimal to sub-optimal (Mertz, 1974). There are fifteen (15) mineral elements essential to ruminants' nutrition. These include seven (7) macro elements; Na, K, Ca, P, Mg, Cl and S. Macro elements are those elements present in the body in large quantities and whose daily amount exceeds 100 mg/kg. Trace elements, on the other hand, are those elements required in very small (μg/g) amounts for optimum body function but are equally important in animal nutrition, and whose deficiency can lead to lowered productivity and clinical disease (Cortzias, 1967; Mertz, 1974). These elements are found in the body in concentrations below 50 μg/g dry matter (McDowell and Conrad, 1989). There are eight (8) trace elements: Fe, Zn, Mn, Mo, Cu, Co, Se, and I.

2.2 FUNCTIONS OF MINERALS IN THE ANIMAL BODY

Though most regions of the world have had incidences of mineral deficiencies and toxicities, tropical regions are mostly affected since grazing livestock depend wholly on forage for their entire mineral requirements (McDowell and Conrad, 1989). A study in the tropics of Africa has reported deficiencies of Co, Cu, Se and Zn in 34 countries

(McDowell *et al.*, 1984). Problems of diverse nature therefore result from an imbalanced supply of minerals to the animals.

2.2.1 Macro elements

a) Calcium and Phosphorus

Phosphorus (P) and Calcium (Ca) exist in combined chemical form similar to that of the mineral hydroxyapatite $3Ca_3(PO_4)_2.Ca(OH)_2$ in the ration of 1:2.2. Calcium is the most abundant in animal body with the skeleton and teeth containing 90 % calcium. It provides strength and density to bones thus preventing easy fracture and deformation of the bones (Read *et al.*, 1986). In times of insufficient supply of calcium, grazing cattle react by redistributing skeletal calcium reserves by a modeling process, which involves resorption of bone from mineralized sites and its deposition at points of growth (Suttle, 1983). Calcium also plays a key role in a number of enzyme systems such as transmission of nerve impulses, contractile properties of muscles and coagulation/ clotting of blood plasma, rhythmic heart action, enzyme activation and permeability of membranes.

Dietary calcium concentrations below 2 g/kg dry matter (DM) are likely to cause deficiency disorders (NRC, 1989). Some of the Ca deficiency signs include skeletal and dental abnormalities like lameness and depressed growth, enlargement of joints, stiffness, misshapen bones, which lead to rickets in young calves and osteomalacia in mature cattle. Acute Ca deficiency (milk fever) occurs in parturition in dairy cows because of sudden increase in Ca requirement for milk production (Suttle, 1983).

Phosphorus is present in the animal body with bone containing 75 % phosphorus. Similar to calcium, classic signs of phosphorus deficiency are revealed in the skeleton (Little, 1984; Read *et al.*, 1986). Phosphorus occurs in body as phosphoprotein, nucleic acid and phospholids. Some of its key roles include cellulose digestion, utilization of energy from feeds (ATP), buffering of blood, protein metabolism and bone formation. The consequence of P deficiency in skeleton is similar to that of Ca. However severe P deficiency in addition can cause loss of appetite and depraved appetite, which is manifested in chewing bones of dead animals and debris, a condition similar to "pica" disease. Extreme P deficiency may prolong infertility period to 2–3 years although if the cow produces a calf, the cow may stay for long before the next oestrus period when the required P levels have been retained (Butler and Jones, 1973).

b) Sodium and Potassium

There is evidence that sodium (Na), other than chlorine (Cl) in sodium chloride is the chief limiting factor in salt deficient diets for cows (McDowell and Conrad, 1989). It is a major cation of the ionic balance between K, Na, Ca and Mg, which is present in blood plasma and other extra-cellular body fluids. Together with Cl, Na plays a role as electrolyte in body fluids in processes such as transmission of nerve impulses, absorption and transport of sugars and amino acids from the digestive tract and across the cell membranes, acid-base balance and osmotic regulation of body fluids (Mills, 1983).

Most cases of Na deficiency amongst grazing cattle in the tropics are due to high Na losses through sweat, which lead to body dehydration. The initial sign for this is craving for salt, which is demonstrated by licking of wood, soil and drinking a lot of water. However, loss of appetite, decreased growth, unthriftiness, reduced milk production and loss of weight arises on prolonged deficiency, which might lead to death of the animal. Feacal sodium concentration has been suggested as a practical method of detecting its deficiency (Khalili, 1991).

As a principal cation of intracellular fluids, potassium (K) is required for a variety of body functions such as osmotic regulations, acid-base equilibrium, water balance, nerve and muscle excitability and in carbohydrates metabolism in animal body. Animal potassium requirement increases in cases of stress, which increases urinary loss of K. Though young forage usually contain adequate K levels, cases of prolonged dry season can lead to reduction of potassium in mature forages (McDowell *et al.*, 1993). Potassium deficiency is manifested in slow growth, reduced feed and water intake, muscular weakness, emaciation, stiffness, nervous disorders and intracellular acidosis (McDowell *et al.*, 1993). Studies have shown that dietary K intake is the best indicator for determination of potassium status in animals (McDowell *et al.*, 1984; McDowell, 1985).

c) Magnesium

Magnesium (Mg) is closely associated with Ca and P with 70 % of total Mg being found in animal skeleton and the rest being distributed in soft tissues and fluids (McDowell and Conrad, 1989). It is the commonest metal ion in enzyme activators especially kinase,

phosphate transferases, decarboxylase and acyl transferases. Magnesium deficiency leads to hypomagnesaemia and the associated tetany; a disorder encountered when calves and cows are reared on milk for a long time without access to other foods. Reduction of Mg content in cerebrospinal fluid (<1.6 mg/100ml) is an indication of Mg deficiency (McDowell and Conrad, 1989). The typical symptoms of tetany are nervousness, tremors, twitching of facial muscles, staggering gait and convulsions. Incidences of non-clinical hypomagnesaemia are greater than clinical tetany and are significant in lowered production, including decreased milk and beef production. Hypomagnesaemic tetany has been prevalent in decreased availability of Mg in forages, which is caused by the antagonizing effects of potassium (Grunes and Mayland, 1975). Susceptibility to grass tetany is higher in old ruminants since the ability to mobilize skeletal Mg decrease with increasing age (Chico *et al.*, 1973). Most mineral supplementations of Mg contain inadequate Mg quantities that can protect against tetany in both susceptible and non-susceptible periods (McDowell *et al.*, 1984). However, a mineral mixture with 25 % MgO (or 14 % Mg) can be effective in managing tetany in beef cattle (Cunha *et al.*, 1973).

2.2.2 Trace elements

a) Iron and Manganese

Iron (Fe) is an essential component of several complexes including haemoglobin and myoglobin (Table 2.1), which are important for oxygen transport in blood and muscles (Hunt and Goff, 1990).

Table 2.1.Metabolic functions of some enzymes and metalloproteins associated with the agriculturally important elements

Element	Enzyme	Function
Fe	Catalase, Cytochromes Haemoglobin, Myoglobin Haemosiderin Succinate dehydrogenase Transferrin Xanthine oxidase	Protect the cells against H_2O_2 Electron transfer Oxygen transfer Storage Carbohydrate oxidation Storage Purine metabolism
Cu	Ceruloplasmin Cytochrome oxidase Cytocuprein Haemocuprein Lysyl oxidase Mitochondrocuprein Super oxide dimustase Tyrosine-3-monooxygenase	Fe utilization and storage Terminal oxidation Superoxide dismutase Storage Collagen cross-linking Storage Dismutation of superoxide free radical Hair and skin pigmentation
Mn	Arginase Deoxyribonuclease Glycolsyl transferase Oxaloacetate carboxylase Pyruvate carboxylase Superoxide dismutase	Urea formation DNA formation Glycoprotein synthesis Oxaloacetate formation Pyruvate formation Dismutation of superoxide free radical
Zn	Alcohol dehydrogenase Alkaline phosphotase Arginase Carbonic anhydrase Carboxypeptidase Collagenase Cytocuprein DNA and RNA polymerase Superoxide dismutase	Alcohol metabolism Phosphate ester hydrolysis Urea formation CO_2 formation, Protein metabolism, Collagen formation Superoxide dismutation DNA and RNA formation Dismutationof superoxide free radical
Co	Glutamate mutase Methyl malonylCoA mutase Methyl transferase	Oxygen transfer Succinate formation Methyl transfer
Mo	Aldehyde oxidase Sulphide oxidase Xanthanine oxidase	Aldehyde oxidation Sulphate oxidation Purine metabolism
Se	Glutathione peroxidase (GSH-Px)	Peroxide removal
I	Thyroxine	Growth control

(Source: Suttle, 1983)

It is also a major component of several body enzymes such as catalase, cytochromes and succinate dehydrogenase, whose functions are: to protect cells against damage by hydrogen peroxide (H_2O_2), electron transfer and carbohydrate oxidation, respectively (Suttle, 1983). The nutritional iron requirement for grazing animals is usually met at the concentration range from 50-100 ppm dry weight (DW) while that for weaned dairy cattle typically range from 13–43 ppm (NRC, 2001).

Iron deficiency in cattle is usually rare unless when there are incidences of blood loss. Young calves in particular can be victims especially if they are fed on milk only, which contains no supplemental iron for an extended period of time (NRC, 2001). Deficiency of iron in grazing cattle is manifested in listlessness and fatigue, palpitation on exertion, angular somatitis and sore tongue (Table 2.2), which lead to anaemia (Underwood, 1981; Suttle, 1988). Iron supplementation is usually not necessary in ruminant diets due to the high iron content in many feedstuffs and soil contamination of many feedstuffs that are ingested by cattle. Elements such as Cd, Co, Cu, Mn, P and Zn reduce absorption and utilization of iron by cattle (Puls, 1994). Excess iron increases the risk of infection since it enhances bacterial growth (Dallman, 1990).

Manganese is a component of the several enzymes such as arginase, deoxyribonuclease, glycosyl transferase, superoxide dismutase whose functions are urea formation, DNA formations, glycoprotein synthesis and dismutation of superoxide free radicals (Table 2.1).

Table 2.2. Impact of trace mineral deficiency on ruminant health and reproduction

Element	Animal species	Symptoms
Fe	Cattle and Sheep	Listlessness and fatigue, palpitation on exertion, angular somatitis and sore tongue, anaemia.
Mn	Cattle and sheep	Suppression of oestrus, cystic ovaries and reduced conception rates. Loss of appetite, loss of weight in calves, retarded growth.
Cu	Cattle and sheep	Depressed growth, hair and bone disorders, Anaemia, stiff gait, fibrosis of myocardium, diarrhoea depressed growth, listlessness and weakness, bone disorders, scouring, dehydration, delayed or depressed oestrus
Zn	Cattle and sheep	Reduced conception rates, increased retained placenta, inhibition of sperm maturation, sterility, Anorexia, retarded growth, bone disorders, decreased wound healing, inflammation of mucous membrane of nose and mouth.
Se	Cattle and sheep Calves	Sub-clinical growth loss, unthriftiness, impaired reproductive function, Arched back
I	Cattle and sheep	Retarded growth, depressed metabolic rate, hair and wool loss, reproductive failure, birth of weak, dead or hairless young ones, abortion and stillbirth, irregular or suppressed oestrus.
Co	Calves and lambs	Anorexia, loss of weight, listlessness, anaemia, weak lambs and calves at birth

(Source: Underwood, 1977, 1981; Howell, 1983; Suttle, 1988; Puls, 1994)

These enzymes play a role in metabolism of carbohydrates, fats, proteins and nucleic acids (Puls, 1994). Apart from being an essential part in the normal brain function, Mn also helps in collagen formation, bone growth and synthesis of fats (Keen and Zidenberg-Cherr, 1990). Manganese also plays a role in reproduction (Suttle, 1983). It is necessary for cholesterol synthesis, which in turn is required for synthesis of the steroids, oestrogen, progesterone and testosterone. Insufficient steroid production results in decreased circulating concentrations of these reproductive hormones resulting in abnormal sperm in males and irregular oestrus cycles in females (Brown and Casillas, 1986). The corpus luteum has high manganese content and responds to manganese supplementation during times of insufficient dietary supply. Also, vaginal manganese concentrations are higher in cycling than in an oestrous ruminant (Miller *et al.*, 1988).

A deficiency in Mn is associated with suppression of oestrus, cystic ovaries and reduced conception rates (Puls, 1994; Corrah, 1996). Mn deficiency is also manifested in loss of appetite, loss of weight in calves and retarded growth in cattle and sheep (Underwood, 1977). The manganese requirement of dairy cattle typically ranges between 13 and 22 ppm depending upon stage of the life cycle and dry matter intake (NRC, 2001). Feeding manganese at increased levels has been able to minimize these problems (Corrah, 1996). Dietary intake of calcium and potassium increase manganese requirements due to increased feacal losses (Miller *et al.*, 1988). Iron, magnesium, phosphorus and cobalt also reduce the availability of manganese (Puls, 1994).

b) Copper and Molybdenum

Copper (Cu) is an important physiological component in a variety of enzymes. Typical examples of these enzymes are cytochrome oxidase, ceruloplasmin, lysyl oxidase, and tyrosine-3-monooxygenase (Suttle, 1983). An imbalance in Cu levels results in a wide

range of consequences on the activity of these enzymes. These involve depressed activity of cytochrome oxidase and tyrosine-3-monooxygenase that would reduce activities dependent upon ATP and synthesis of noradrenaline or pigment melanin. Reduced superoxide dismutase activity will affect cell antioxidant systems with Fe metabolism being altered by loss of activity of ceruloplasmin.

Copper deficiency in grazing livestock is experienced worldwide including the tropics. Although gross signs of Cu deficiency are rare when concentrates are fed, worldwide reports are concerned with a "conditioned Cu deficiency" where normal Cu levels of 6–16 mg/kg DM are inadequate because forage constituents such as Mo and S, block the Cu utilization (Jumba, 1989). Copper deficiency occurs also as a result of decreased production of antibody producing cells since levels of T and B cells, neutrophilis and macrophages are usually affected. Copper deficiency in forages occurs when Cu levels are below 5 mg/kg DM or when Mo levels exceeds 3 mg/kg DM. Normal Cu and low Mo, with high levels of protein results from increased sulphide amounts produced in the rumen from fresh pasture, thus resulting in unavailable Cu sulphide formation (Dick *et al.*, 1975). Both Cu deficiency and Mo toxicity signs are similar and are corrected by provision of extra Cu in animal's diet. The clinical signs of Cu deficiency are scouring, rough and bleached hair, slow growth and loss of body weight, pale eyes and mouth membranes (Mills, 1983). The primary cause of Cu deficiency in some areas has not been fully identified. Soil-plant studies on Cu have attempted to investigate its deficiency and the effect of interfering elements especially Cu-Mo-S (Jumba *et al.*, 1995b). A recent soil-plant-animal study on mineral interactions revealed that higher amounts of either Mo

and/or S relative to Cu usually antagonize copper thereby lowering its level (Oduor, 2002).

c) Zinc

Zinc is an essential component of over 200 enzyme systems, which are responsible for carbohydrate metabolism, protein synthesis, nucleic acid metabolism, epithelial tissue integrity, cell repair and division, and vitamin A and E transport and utilization (Table 2.1). In addition, it plays a major role in the immune system and certain reproductive hormones. Zinc is also known to be essential for proper sexual maturity, reproductive capacity, and more specifically, onset of oestrus (Cousins and Hempe, 1990). Zinc has a critical role in the repair and maintenance of the uterine lining following parturition, speeding return to normal reproductive function and oestrus (Green *et al.*, 1998). Zinc is found in enzymic structures of RNA, DNA and Ribosomes. The recommended dietary content of Zn for dairy cattle is between 18 and 73 ppm depending upon stage of the life cycle and dry matter intake (NRC, 2001).

Zn deficiencies are rare in the grazing animal but could occur in presence of high concentrations of interfering elements such as Ca, Cu, Fe and Cd (Puls, 1994). Zinc deficiency in grazing cattle is manifested by reduced feed intake, reduced growth rate and feed efficiency, skin disorder, hair loss and inflammation of nose and mouth (Table 2.2). Low Zn levels are also manifested in reduced reproductive activity and development in both males and females. In bulls for example, zinc deficiency results in poor semen quality and reduced testicular size and libido (Arora, 1988). Zinc has also been shown to

increase plasma beta-carotene levels, which is directly correlated with improved conception rates and embryonic development (Mass, 1987; TB B-9202-A, 1992.). Improved zinc status also enhances fertility by reducing lameness, resulting into cows showing more willingness to heat, improved mobility and performance in bulls (Suttle, 1988). Inadequate Zn supplementation results in mild to severe claw (hoof) disorders, including weak claws that are more susceptible to inter-digital and digital dermatitis and foot rot (Puls, 1994).

d) Selenium

Selenium is recognized as an essential element that defends the body against oxidative stress (Chesters and Arthur, 1988). The functional role of selenium (Se) in animals as part of the enzyme glutathione peroxidase (GSH-Px) is in peroxide removal (Table 2.1). The activity of this enzyme responds rapidly to alterations in dietary Se intake and provides the best link between the intake of trace elements and their functional adequacy (Hafeman *et al.*, 1971). The selenium requirement of dairy cattle is 0.3 ppm (NRC, 2001).

The symptoms of Se deficiency vary in severity from general unthriftiness, scouring and infertility in mature animals to severe white muscle disease (or nutritional muscular dystrophy, NMD), which is more prevalent in growing lambs and calves (Judson and Obst, 1975). The occurrence of Se deficiency is somewhat complicated by the vitamin E status that share the role of protecting tissues from oxidative stress (ARC, 1980; Chesters and Arthur, 1988). Selenium deficiency in mature cattle has been associated with retained

placentas, infertility (McPherson *et al.*, 1988) and increased susceptibility to mastitis (Smith *et al.*, 1984). Marginally, selenium deficient animals usually abort, or give birth to weak calves (Table 2.2).

Cadmium, copper, calcium, mercury, lead, zinc and sulphur can induce a selenium deficiency. Dietary Ca levels greater than 0.8 mg/kg for instance, can reduce selenium absorption (Puls, 1994). Research indicates that selenium supplementation reduces the incidence of retained placentas, cystic ovaries, mastitis and metritis (Puls, 1994). In addition, cattle that maintain adequate blood selenium levels have reduced incidence of abortions, stillbirths and peri-parturient recumbence (Miller *et al.*, 1988; Puls, 1994). Compromised selenium status has also been associated with poor uterine involution and weak or silent heats. In males, selenium supplementation has been shown to increase semen quality (Puls, 1994).

e) Cobalt

Cobalt (Co) exists in body as a component of vitamin B_{12} and is found in several enzymes like glutamate mutase, methyl malonylCoA mutase and methyl transferase whose functions are oxygen transfer, succinate formation and methyl transfer (Table 2.1). In particular, the conversion of propionate via L-methylmalonic acid (MMA) to succinate is an energy-leasing reaction catalyzed by methyl malonyl-CoA mutase (Jumba, 1989). Cobalt also assists in the formation of proteins from amino acids and the metabolism of fats and carbohydrates (McDowell *et al.*, 1993). Cobalt deficiency is restricted to grazing ruminants having no access to mineral concentrates. The clinical signs of Co deficiency

include poor appetite, loss of vigour, muscle wasting in cattle and sheep, and anorexia, loss of weight, anaemia (Table 2.2). However, Co deficient ruminants respond quickly to Co supplementation.

2.3 TRACE ELEMENT TOXICITY

A trace element is said to be toxic when it is either ingested (or inhaled) at sufficiently high levels for a prolonged period or is available in diets at high concentrations. The excess of any element leads to failure of homeostatic mechanism, body processes that lead to occurrence of clinical disorders. Toxicity may be chronic or acute. In chronic toxicity there is frequently no correlation between the signs of toxicity and dietary concentrations of the element. The time course is dependent on many factors, including the species and breed of the animal, the storage capacity in the animal's body for the element, the physiological and elemental status of the animal prior to exposure (Suttle, 1983) and the intensity of the toxicity. Addition of Mo and sulphur to diets rich in copper can reduce absorption and accumulation of Cu and hence prevent toxicity (Campell and Mills, 1979; Woolliams *et al.*, 1982; Allen *et al.*, 1983). Cases of trace element toxicities have been reported for Cu, Zn, Mo, and Se and As (Howell, 1983). Zinc toxicity leads to depraved appetite, decreased weight gains, decreased feed efficiency, and anorexia (Table 2.3). In pregnant animals, selenium toxicity will lead to abortions, stillborns and weak and lethargic calves as selenium accumulates in the fetus at the expense of the cow.

Table 2.3 Symptoms of toxic intakes of certain trace elements

EELEMENT	SYMPTOMS
Copper	**Chronic**: Loss of appetite, thirst, apathy, haemolytic icterus, haemoglobinuri jaundice, and death from hepatic coma. **Acute**: Abdominal pain, Diarrhoea
Molybdenum	Growth retardation, loss of weight, diarrhoea (cattle), wool defects, joi abnormalities, connective tissue changes and reproductive problems
Zinc	Parakeratosis, salivation, anorexia, diarrhoea, dehydration, profour weakness.
Selenium	**Cattle**: Dullness and lack of vitality, loss of appetite, coat roughness, sorene and sloughness of hooves, stiffness and lameness, blindness, abdominal pai diarhoea, teeth grinding, paralysis, anaemia, death.
Arsenic/Lead	Anaemia, diarhoea, constipation, death

(Sources: Underwood, 1977, 1981; Howell, 1983)

The occurrence of copper toxicity can arise from high intake of other elements like Fe, Zn and Cd and grazing of plants containing hepatotoxic alkaloids (Underwood, 1977; Bremner, 1979). Soils and forage selenium levels of 30-3000 mg/kg and 5-500 mg/kg DM have revealed cases of toxicity in grazing animals (Howell, 1983).

2.4 FACTORS INFLUENCING DIETARY MINERAL REQUIREMENTS

2.4.1 Stage of development of animal

Mineral requirements depend mainly on the level of production and physiological state of the animal. A pregnant beef cow in her first lactation would need higher mineral requirements than a mature dry cow (NRC, 1984). Also, lactating cows will require higher levels of calcium and phosphorus than non-lactating cows. For example, 0.17-1.53 % Ca and 0.17-0.59 % P are adequate for growing development of steers and heifers while 0.43-0.77 % Ca and 0.25-0.48 % are adequate for lactating dairy cows (NRC, 1984). The Ca: P ratio is important since a ratio of 1:1 or 2:1 is good for growth and bone formation

2.4.2 Mineral interrelationships

The bioavailability of certain element in a compound or supplement is of major concern during selection of a feed. The chemical form and mineral interrelationships determine specific mineral requirements for animals (Underwood, 1981). Iron for example is mostly available as ferrous sulphate than as ferric oxide. The components of the diet taken have a role on amount of mineral available in that diet. Copper availability varies closely with the amount of molybdenum present. An increase in the amount of molybdenum will require a similar increase in copper levels since Cu-Mo interaction may arise. The amount of organic component in diet affects the amount of the element constituting it (McDowell *et al.*, 1993). High amounts of selenium will depend on the relative amount

of vitamin E in the feed. Other mineral interrelationships include: Ca-P, Fe-P, Al-P, Ca-Zn, Cu-Mo, Fe-Cu, Se-As-S and K-Na-Mg.

2.4.3 Age of animal

Dietary requirements may decline with age since the major requirements for growth often remain constant for a given live weight gain (McDowell and Conrad, 1989). Young calves will require more minerals for growth than mature cows since they are more efficient in mineral metabolization than mature cows. Mineral availability will also depend on other sources of minerals available to animals since most calves depend on milk while mature cows derive their minerals from forages and probably ingested soils.

2.4.4 Breed differences and adaptations

Mineral requirement differ depending on the breed of animal (Field, 1984). Indigenous breeds are slow growing and late maturing while exotic matures faster with high productivity returns. This is due to marked variations within breeds in terms of efficiency of absorption of minerals from the diet. Local breeds will adapt more easily than exotic breeds in a given region since local breeds usually have high degree of adaptability than exotic ones. Any sudden change like grading up will require higher maintenance and production requirements in terms of nutrients. Efficiency in absorption of specific mineral in diet depends on genetic differences of breeds leading to higher mineral requirement for particular breed than the other. Friesian, Ayshire and Friesian/Ayshire breeds require more minerals than indigenous breeds.

2.4.5 Forages

Forage species differ genetically in their ability to absorb mineral elements from soil. This is due to their ability to adapt to soil conditions of moisture, pH, chemical condition and drainage (Loneragan, 1975). Herbs and legumes usually are richer in a number of minerals than grasses (McDowell *et al.*, 1993). Most grazing ruminants in the tropics depend almost exclusively on forages for their requirements. However, these tropical forages do not completely satisfy all mineral requirements (McDowell *et al.*, 1983). Reduced forage intake due to low protein content (<7.0 %) and increased degree of lignifications may therefore be responsible for decrease in mineral amounts consumed. Since tropical forages may contain lower concentration of minerals during the dry season, it may be assumed that ruminants mostly suffer mineral insufficiencies during this time. However some studies have indicated some specific mineral deficiencies, which are prevalent during wet season (McDowell, 1985).

2.4.6 Water

Drinking water may be a significant source of minerals. However, less than 20 % of the trace element requirements are provided in water especially for Fe, Zn, Cu and Se. Water may occasionally contain toxic concentrations of some trace elements, which is manifested in fluorisis (McDowell, 1992). Wells and springs have been found to contain high concentrations of As, Li, Sr, B, Se and Pb, which seldom bear any relationship to their concentration in soils. Water naturally has high Mo and/or S concentrations, which can induce Cu deficiencies in grazing animals (McDowell, 1992).

2.4.7 Soil

Grazing livestock usually ingest large quantities of soil that is found attached on the grass. The ingested soil can be either beneficial in improving performance through availability of essential trace elements like Co and Se or injurious through excess uptake of toxic substances such as pesticides, cadmium and lead (McDonald and Sutter, 1986). However, deliberate soil consumption (geophagia) is classified as a form of pica (a condition of animals chewing and eating materials not considered as natural feedstuffs) which is due to insufficiency of some nutrients in diet (McDowell, 1985).

2.5 ASSESSMENT OF MINERAL IMBALANCES IN GRAZING LIVESTOCK

The methods of assessing the incident and impact of mineral imbalances in grazing livestock are classified into three major ways: (i) use of clinical symptoms (ii) using soil, plant and animal biomedical data in relation to the set standard requirements and (iii) using responsive conditions in the absence of clinical signs (Langlands, 1987; Suttle, 1988) each of which is discussed below.

2.5.1 Clinical symptoms

Table 2.2 and Table 2.3 give deficiency and toxicity symptoms of trace elements. These symptoms reveal that same deficiency symptoms arise when the dietary intake of an element is in excess. Studies have reported corresponding clinical effects due to excessive and insufficient intakes of some trace elements (Philipo, 1983). Excess or insufficient intake of Cu for example can lead to enhancement in symptoms such as bone

defect, hair/wool changes, scouring, anaemia while also depressing animals' reproduction (Philipo, 1983). This means that symptoms overlap and may not offer the best diagnostic tool in the assessment of mineral imbalances. Occurrence of anaemia therefore gives little clue as per the element involved since it is a characteristic of Fe, Cu and Co deficiencies and of Zn, Mo and S toxicities. Similarly, the same deficiency does not always give the same symptom. This is because the activity of some enzymes will decline rapidly while that of others will be strongly conserved. Mills *et al.*, (1976), found a differential decline in the activity of independent enzymes when they experimentally induced Cu deficiency in calves. This shows that clinical and biochemical consequences of a trace element deficiency vary widely between different species of animals and within the same species at different stages of growth and environmental conditions. The failure to conclusively assess incidence and impact of mineral imbalances based on diagnostic records of clinical symptoms has led to use of alternative procedures as below.

2.5.2. Geo-chemical and regional reconnaissance techniques

This is an indirect method of sampling, which indicates an average composition in soils of a relatively large area with minimum number of samples. This may be due to capability of soil having profound variations both vertically and laterally. It involves basically four methods. Firstly, geological maps provide some information on gross distribution of elements in the parental materials, which form soils. However, many rock types show considerable chemical variations and hence detailed chemical data are rarely available for mapping areas of significant size. Mineral deficiencies or toxicities in grazing livestock on the other hand can be predicted systematically by mapping or

regional reconnaissance especially in the tropics. Mapping techniques based on forage and soil analyses have been done for Ca and P in Brazil and Se in Venezuela (McDowell *et al.*, 1984). Deficiencies of Co and/ or Cu in grazing cattle, depicted by their low concentration in liver, have been associated with deficient levels in Brazilian soil (McDowell *et al.*, 1984). Secondly, remote sensing by aircraft or spacecraft, which is based on colour or reflectivity variation induced in plants by variation in soil chemistry, can also be used (Appleton, 1994). However, this method has the disadvantage of being limited to areas nurturing a very restricted variety of plants (natural or cultivated). Also, not all plants show significant colour or reflecting responses to chemical areas of the soil which retard their growth or plants/animals introduced to the area.

Thirdly, airborne scintillometer surveys based on radiation from uranium (U) and potassium (K) has been demonstrated as a reconnaissance technique for mapping soils in USA. This method is potential for rapid assessment for homogeneity and patterns of soil types in survey areas although it does not assess the abundance of elements other than U and K (Appleton, 1994). Lastly, stream sediment geochemistry offers the widest scope for agricultural reconnaissance. Low-density sampling of stream sediments makes it a widely practiced and successful method of prospecting for cancelled mineral deposits. This method is based on the premise that a sediment sample has chemistry, which is approximated to the average mineral composition of soils within catchment area upstream especially for Cu deficiency and Mo poisoning (Thornton *et al.*, 1972).

2.5.3 Soil analysis

A soil usually has a mineral composition similar to that of the parent bedrock from which it is formed. Since forage, which is fed on by animals, derive minerals from various soil types; a mineral imbalance in animal can be pegged on mineral levels in the soils. Soil data was used to depict the Se deficient areas of New Zealand although further evidence showed that climate and effect of weathering process were responsible for soil type and its mineral availability (Langlands *et al.*, 1981). This information has been used to assess livestock mineral deficiencies in tropical and subtropical regions of Brazil (Conrad *et al.*, 1980) and USA (Kiatoko *et al.*, 1982).

Several factors that normally reflect soil mineral composition include:

i) Weathering process. Any parent rock material has mineral content which reflects the elemental distribution that occurred during formation of earth's crust (Jumba, *et al.*, 1995a, b). The ability of trace elements to enter crystal lattices or spatial networks of different minerals as they are crystallized is related to chemical properties of elements such as charge, ionic radius and electronegativity. Physical, chemical and biological weathering of any parent material leads to the formation of soil in an integrated and continuous process. Soil formation is influenced by such effects as climate, relief and how long these factors may have interacted (West, 1981). More recent pedogenic processes like leaching, gleying, surface organic matter accumulation and soil properties such as reaction pH and redox potential further influence both total amount and forms of trace elements in the soil (Berrow and Mitchell, 1980).

ii) Assessment of soil mineral status. Examining the "total " or "extractable" elemental concentration during soil analysis may indicate need to apply deficient elements at establishment or sowing time (Jumba, *et al.*, 1995a, b). Most assessments are based on available elements obtained by extraction of soil with chemical reagents. However, extractable soil elements may correlate poorly with plant uptake due to wide variations in soil properties including pH (Loneragan, 1975), organic matter uptake by plant species (Fleming, 1973), action of microbes (Bromfield, 1978) and effects of inter-element interactions due to fertilization (Reuter, 1975a, b).

Since plants are the final arbiters of mineral amounts "available" in soils, measuring plant composition and relating this to the elemental content in soil can assess their mineral availability. The choice of extractant used depends on both the element being measured and the nature of soil. The critical soil concentration below which deficiency is expected is estimated by using either regression analysis or by separating deficient and non-deficient soils into classes on basis of plant analysis which however leaves "grey" area between these extremes (Reuter, 1975a).

2.5.4 Forage analysis

Forage analysis has been used successfully in assessing the mineral status of pastures in relation to disorders in grazing ruminants on a specific or regional basis and for estimating the mineral supply in conserved feeds (Hogan *et al.*, 1971; Jumba, *et al.*, 1995 a, b). The data obtained is usually compared to critical dietary concentrations/levels or recommended standards below or above which imbalances in livestock can be expected

(McDowell *et al.*, 1982; NRC, 2001). Concentration of minerals depends on interaction of different factors such as soil, plant species, stage of maturity, pasture management and climate. Large variations in mineral content of different plant species growing on same soil have been reported (McDowell *et al.*, 1982). Herbs and legumes for example are rich in a number of mineral elements than grasses (Fleming, 1973). This shows that mineral status depends on botanical composition and species analysis. Stage of growth of plant can also influence mineral content in herbages. The decline in mineral content of plants as senescence sets in is due to reduced translocation to the root system (Tergus and Blue, 1971). Phosphorous deficiency for example is more common than Ca deficiency in grazing animals because P content of forage is more sensitive to senescence, falling rapidly than forage Ca with plant age. This means paddock roughage can be a major constituent of Ca for animals receiving cereals as supplement in times of drought. Organic nutrients present in pasture may also influence the mineral content since they are interrelated. Both Se and vitamin E protect cell membranes from damage by peroxide removal. Dietary vitamin E as provided by a number of tocopherols and tocotrienols moderates other active radicals in some manifestations of Se insufficiency. Vitamin E concentrations also vary widely within plants and decline as pastures mature, senesce or are stored as conserved feeds. Cattle grazing green pastures throughout the year are likely to be Se than Vitamin E deficient (Langlands, 1987). The use of forage element analysis in assessing mineral adequacy for grazing livestock has its own disadvantages such as: -

i) Uncertainty of samples representing what livestock consumes. Livestock usually tend to select particular species (usually with high protein and highly digestible) during periods of pasture abundance (Egan, 1975).

ii) Variations in the availability of forage elements due to interactions between dietary components. Manifestation of Cu deficiency (Table 2.2) for example can be brought about by Mo toxicity (Table 2.3) which is complicated by high levels of dietary S through formation of insoluble copper thiomolybdates. Herbages with less than 3 mg Cu/kg DM and no Mo indicate " a simple Cu deficiency " while those with 5 mg Mo/kg and 9 mg Cu/kg DM were described as "Mo in excess hence Cu inadequate" in cattle (Jumba, 1989).

iii) Possibility of soil contaminated forage samples. Research has shown that animals can ingest large quantities of soil while grazing pasture with soil from rain splash or wind blow (Metson *et al.*, 1979). This is a problem for Cu and Mo, which is found in plants and soils in some amounts but can be serious for elements, which are more abundant in soil like Fe, Al and sometimes Mn. A study revealed that for a plant Co concentration of 0.075 mg/kg DM, a 0.05% contamination by a soil containing 60 mg/kg as total Co would increase herbage Co status from deficiency level of <0.08 mg/kg DM to dietary adequacy of 0.105 mg/kg DM (Metson *et al.*, 1979).

2.5.5 Biochemical indices (markers) in animals

Animal tissue or blood analysis has been considered more reliable than visual symptoms approach (Suttle, 1986, 1988). Tissue samples may be collected from the main site of mineral storage e.g. liver or from blood or fluids like saliva/milk (Table 2.4). These may be analyzed for the mineral itself, a metabolite, (e.g. methylmalonic acid (MMA) for Co), or biologically active forms such as enzymes, hormones and vitamins. Some analysis

reflect mineral intake (e.g. plasma Se), body reserves (e.g. liver Cu) while others indicate changes in normal metabolic processes (e.g. serum MMA as an index of Co deficiency).

Table 2.4. Best matrix for elemental analysis

Element	Tissue/fluid
Calcium	Bone
Phosphorus	Bone
Magnesium	Urine/bone/blood
Copper	Liver
Cobalt	Blood
Selenium	Whole blood/plasma
Sodium/Potassium	Saliva

(Source: Tartour, 1975)

Biochemical changes which can be monitored and those, which precede the appearance of clinical disorders usually, characterize the change from mineral depletion to deficiency. These changes have been attributed to four phases: depletion, deficiency, marginal dysfunction and disease.

Compton metabolic profile test (CMPT) is another approach in detecting mineral contribution from animal data to metabolic nutritional disorders. This attempts to identify abnormalities in the composition of blood. Samples are analyzed for packed cell volume, haemoglobin, glucose, serum urea, albumin, total proteins, Ca, Cu, Fe, K, Mg, and inorganic P. This test was developed for possibility of non-mineral causes for ill health (Payne *et al.*, 1970). It is however costly and restricted to a few of the elements covered below.

i) Calcium, Phosphorus and Magnesium

The concentrations of these elements are not always useful in determining deficiency. For example, plasma Ca which is regulated homeostatically, is sensitive to changes in albumin concentrations and declines only in cases of severe deficiency or suddenly at parturition. The normal values of Ca in plasma can range from about 80-120 mg/l. The buffer capacity of the skeleton is illustrated by the fact that a 400 kg cow contains 5.6 kg Ca and 3.2 kg P (NZSAP, 1983). As a component of bone (Section 2.2.1), Ca and P are preferentially reabsorbed from spongy bone of the vertebra with the ribs being more sensitive to changes in Ca and P status than the long bones. Though Ca and P status can be obtained better from the bone than plasma sample analysis, the bone composition is relatively constant during depletion and therefore Ca: P ratio remains at about 2:1 whether Ca or P is deficient. The main effect of depletion is to reduce total mineral content, which cannot be monitored by biopsy. Biopsy techniques for P studies have been extended from sheep (Little, 1984; Read *et al.*, 1986) to cattle in assessment of P reserves. These studies have shown that P concentration in total fresh ribs is not always a sensitive criterion of the body P reserve since other sites and criterion as biopsy sample of 12th rib, thickness of compact bone can be used. Bone biopsy is also not specific for detecting inadequate intakes of dietary P since cattle receiving adequate P diet but insufficient protein show similar demineralization (Siebert *et al.*, 1975). Therefore plasma inorganic P is used to assess the extent of P imbalance being most sensitive in young growing animals (Suttle, 1987b). Stored Mg in adult ruminant is about 0.3-0.5 g/kg with 70% in skeleton and 25% in the skeletal muscle (NZSAP, 1983). Depletion of Mg can be assessed by its levels in rib biopsy or post mortem samples. Decline in Mg

intake (availability) leads to fall in plasma Mg concentrations from a normal value of 18-30 mg/l. Urine is the principal means by which Mg absorbed in excess of animal's requirement is excreted (Read *et al.*, 1986). Plasma Mg is however the most widely used index for Mg status, as it is more reliable than either plasma Ca or P (Suttle, 1987a).

ii) Copper

Liver is the main site for body Cu and can be used as an index for assessing the Cu status of grazing animals (Tartour, 1975). However, the use of liver for diagnosing Cu imbalance has its own disadvantages; (a) the specimen is not easily obtainable with samples only being obtained by biopsy or by sacrificing the animal; (b) variability between animals in liver Cu concentration is high; (c) concentration can fall to very low levels before health is affected (Suttle, 1986). Depressed concentrations of plasma Cu or enzyme ceruloplasmin indicate depletion of Cu reserves although ceruloplasmin is less sensitive to contamination. Normal plasma Cu varies between 0.6 and 1.2 mg/l and values < 0.5 mg/l indicate a low Cu status (Suttle, 1986). Other tissues, which can be assigned, include hair in cattle (Suttle and McMurray, 1983) and wool in sheep (Woolliams *et al.*, 1983). Care and standardized procedures are needed to clean the fibre to remove contaminants but not the fibre constituents.

iii) Selenium and Vitamin E

Animal Se status is often assessed from the Se content of whole blood, plasma or erythrocytes, or glutathione peroxidase (GSH-Px) activity in whole blood or erythrocytes. Blood Se in cattle and sheep is located in erythrocytes and Se concentrations and GSH-Px

activities are closely correlated especially when Se concentrations are low (Suttle, 1986). GSH-Px enzymes offers advantage of being a biologically active form of Se whose activity can be determined spectrophotometrically or by a simple spot test. Also, preliminary digestion of the sample with perchloric acid is not required. However glutathione peroxidase GSH-Px enzyme assay procedures are difficult to standardize hence may lack comparability between laboratories (Langlands *et al.*, 1980). Results obtained can be reported in units such as per ml of haemoglobin, per ml of cells or per ml of whole blood at temperature range of 25–37^{O}C. Activities in plasma are low, unsuitable and sensitive to hemolytic process. Determination of blood Se concentration involves a lengthy fluorimetric assay or assay of volatile anhydrides by AAS method, neither of which is very easy to perform with consistent results. Since vitamin E determines how much Se is adequate, its response to Se supplements may be needed to effectively diagnose Se-responsive conditions. High vitamin E for example satisfies induced Se-responsive conditions. Attempts to solve Se deficiency problems by top-dressing pastures with sodium selenate appeared to be successful (Shepherd *et al.*, 1984). Due to several disadvantages encountered when using tissue/fluid samples, this study has countered these limitations by using animal blood.

2.5.6 Responsive conditions in the absence of clinical signs

Improved wool production and lamb growth rate were reported in large areas of Australia where increased amounts of Cu and Co were supplied through herbage or by mineral supplementation to animal with no clear deficiency signs recognized (Reuter, 1975b). Supplementation studies of specific trace elements in the field also have revealed

improved female reproductive performance. In pen studies for example, Zn requirement for testicular development was greater than for rapid body growth in young male sheep (Suttle, 1986). The surest diagnosis for all trace elements is often an improvement in growth or health in response to a specific supplement although adoption of preventive measures should be prompted by biochemical evidence of marginal deficiencies in animals rather than soils or pastures (Philipo, 1983; Suttle, 1986; McPherson, 1987). Problems associated with mineral supplementation particularly in tropical regions include: -

i) Insufficient chemical analyses and biological data to determine which mineral is required and in what quantities.

(ii) Lack of mineral consumption data needed for formulation on mineral supplements.

(iii) Inaccurate and/ or unreliable information on mineral ingredient labels.

(iv) Supplements that contain inadequate amounts or imbalances.

(v) Standardized mineral mixtures that are inflexible for diverse ecological regions e.g. supplements containing Se distributed in a set toxic region.

(vi) Farmers amending commercial mixtures recommended by the manufacturers (e.g. mineral mixtures diluted 10:1 and 1000:1 with additional salt).

(vii) Difficulties concerned with transportation, storage and cost of mineral supplements (McDowell and Conrad, 1977).

Chemical analysis of feeds should therefore be done to determine which minerals are required and their respective qualities. Schemes to check responses can therefore be

managed effectively after the limiting elements have been identified; hence the purpose of this study.

2.6. METHODS OF ANALYSIS

A variety of instrumentation methods are available for quantitative determination of macro and trace elements. The most common are atomic absorption spectrophotometer (Shimadzu Corporation, 1991) and ultra violet/visible spectrophotometer (Pye Unicam, 1985).

2.6.1 Atomic absorption spectrophotometry

Principle

AAS uses a certain kind of monochromatic lamp; the hollow cathode lamp, which is able to emit the spectral lines corresponding to the energy required for an electronic transition of an element from a ground state to an excited state. The sample solution is aspirated into the flame where it evaporates to give the dry salt and then the vapour. This vapour dissociates into atoms of the element that absorb resonance radiation from the lamp. Absorption is measured as the difference in the transmitted signal in the presence and absence of test element (Shimadzu Corporation, 1991).

CHAPTER THREE

MATERIALS AND METHODS

3.1 THE STUDY AREA

Uasin Gishu district (3,327 km^2) is one of the eighteen districts that make up Rift Valley province of Kenya. It is located between longitudes 34^0 50' and 35^0 37' East and latitudes 0^0 03' to 0^0 55' North of equator with Eldoret as its headquarters. It borders Trans Nzoia (North), Lugari (Northwest), Nandi (West), Koibatek (Southeast), Keiyo and Marakwet (Eastern) and Kericho (Southwest) districts. It has six divisions: Soy, Turbo, Kapseret, Kesses, Moiben and Ainabkoi (Fig.3.1). Moiben is the largest (778.2 km^2) while Kapsaret is the smallest (297 km^2).

It is a highland plateau with altitude falling gently from 2700 metres above sea level at Timboroa in the East to 1500 metres above sea level at Kipkaren in the west. It is a catchment zone for lake Victoria with rivers Sosian, Kipkaren, Kerita, Masaba and Sergoit providing water. The district receives an average annual rainfall of 900 to 1200mm between March and September with distinct peaks in May and August. The wettest areas are Ainabkoi, Kapsaret and Kesses while lower rains are received in Turbo, Moiben and Soy divisions. Ninety percent (90 %) of the land (2000 km^2) is high potential agricultural land while 1000 km^2 is of medium potential. The forest covers an area of 327 km^2 with nabkoi, kipkurere, lurenge, singalo and kapsaret forests dominating. The major soil types for the region are red loam, red clay, brown clay and brown loam clay soils. The major grass species are rhodes, kikuyu, nandi setaria, guatemala, coloured guinea,

sudan grass, columbus, star, and natural grass. The major grazing cattle breeds include friesian, ayrshire, guernsey and african zebu (MALD, 2003; MPND, 2002b).

The region has two settlement patterns with Turbo and Kapsaret being densely populated due to urbanization while Moiben is sparsely populated due to large farms.

3.2 SELECTION OF SAMPLING SITES

Field sampling was done at the start of the dry season (December, 2002) and before the onset of rains (April, 2003) to obtain samples for laboratory analysis.

The selection of farms and sampling sites were based on the following criteria:

(i) Land use

(ii) Pasture availability and management

(iii) Availability of grazing cattle especially friesian, ayrshire and friesian/ayrshire breeds

(iv) Animal counts and agronomy

(v) Availability of animal health records

3.3 THE SAMPLING SITES

Figure 3.1 shows the location of the sampled sites in the study area. Twenty-eight soil and forage samples and forty two blood samples from 18 young growing calves (2-14 months old) and 24 lactating cows (2-10 years old) were collected from six major farms in the district.

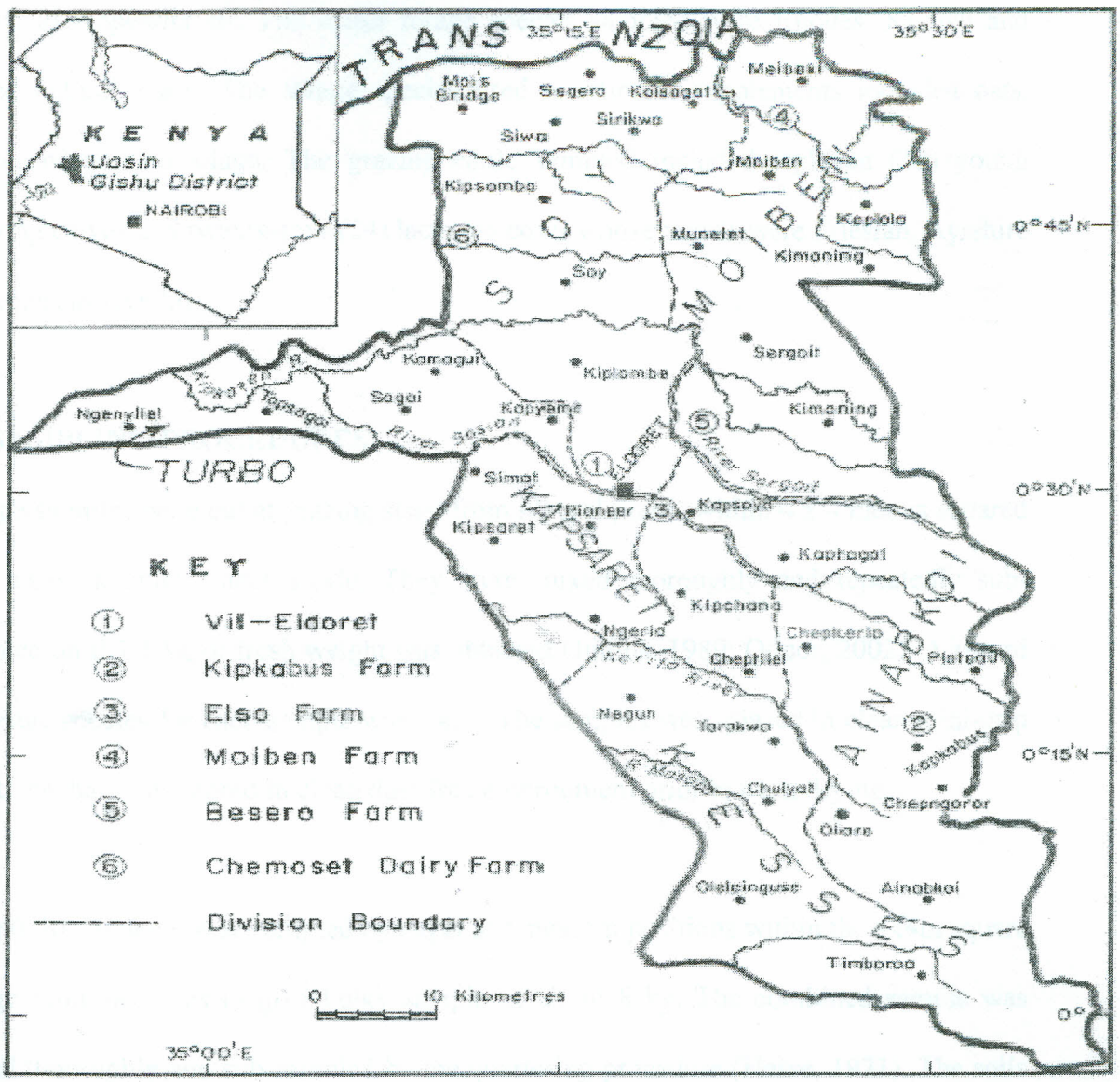

Fig. 3.1. Distribution of sampling sites

The sampled farms were numbered 1 to 6 and are situated in three different regions of the district. The central region around Eldoret had farms 1, 3 and 5, the northern region had farms 4 and 6 while the southern region had farm 2. The actual locations and altitude of the sampled sites obtained using Global Mapping System (GPS) Model Magellan Map 410 are presented in Appendix I. Soil and grass species locations are presented in Appendix II while serum identifications for young growing calves and lactating cows are shown in Appendix III. The major forage species sampled were Rhodes, Kikuyu and mixed natural grass. The forage species used as mineral supplements included oats, maize, wheat and silage. The grazing cattle sampled included eighteen (18) young growing calves and twenty-four (24) lactating cows whose breeds were Friesian, Ayrshire and Friesian/Ayrshire.

3.4 SAMPLING PROCEDURES

Forage samples were cut at grazing stage from several points within 4 x 4 metres squared sites using stainless steel sickle. They were mixed thoroughly and repeatedly sub-sampled until 1.5 kg of fresh weight was obtained (Jumba, 1989; Oduor, 2002). A record of forage species for each sample was taken. The samples were placed in clean mini-grip polythene bags and stored in clean dust-free environment prior to oven drying.

Soils 0-30 cm deep were sampled by auger at 5 random positions within the 4x4m square forage sampling sites to give bulky sample of about 8 kg. The combined sample was mixed thoroughly and sub-sampled by the quartering procedure (Hesse, 1971). The soils were spread uniformly over a sheet of polythene and divided into 4 equal portions

numbered 1-4. Portions 1 and 4 were discarded and the remaining 2 and 3 further spread and reduced to half by the same procedure. The process was repeated to retain a final sample of 1 kg. The samples were stored in clean sealed paper bags.

Blood from each animal was collected in clean vials using 15 gauge "California" bleeding needles by puncture of the jugular vein. The blood was transferred to heparinised vials to ensure clot-free preservation (Oduor, 2002). Blood samples from each farm were classified on the basis of age of animal (young growing calf or lactating cow) and breed (Friesian, Ayshire or Friesian/Ayshire). All the blood samples were stored in a refrigerator prior to serum separation.

3.5 SAMPLE TREATMENT

Soil samples were air-dried at room temperature (<26° C) for one week. Using porcelain pestle and mortar, they were ground and screened through 2 mm (80 mesh) nylon sieve. One portion was oven dried at 105° C for 48 hours and kept in polythene bags for mineral element analysis while the second portion was kept for pH determination.

Forage samples were dried in a forced drought oven at 70 ° C for 48 hours. They were ground in a Wiley laboratory mill and sieved through 0.5 mm stainless steel screen. The sub-sample was stored in polythene containers.

Blood samples were allowed to stand for about eight hours in a slanting position for serum to separate. The serum was transferred to clean cuvettes using Pasteur glass pipettes awaiting analysis (Fick *et al.*, 1979).

3.6 CHEMICALS AND GLASSWARE

Chemicals were of Analar grade obtained from Fisher Scientific. Water was distilled in an all Pyrex distiller and de-ionized by passing through a de-ionizer cartridge before storing.

All glassware were thoroughly washed with laboratory detergent, rinsed with dilute nitric acid solution and finally with de-ionized water. They were dried in an oven at 80^0C. Plastic containers were also thoroughly washed and stored in dust-free cupboard at room temperature.

3.7 INSTRUMENTATION TECHNIQUES

A ChemTech computerized Analytical AAS fitted with a monitor (CTA 2000 Model) was used with an appropriate air/acetylene flame burner. Auto calibration and direct read-out modes were used.

Colorimetric determinations were performed on a Pye Unicam spectrophotometer model SP150 using 10mm path length quartz cells with a background correction system.

Measurements of pH were done using Pye Unicam 292 MK2 Model pH meter with glass electrodes. Appropriate buffer solutions pH 4, pH 7 and pH 9 were used for calibrations of the meter. Each soil sample aliquots (2.5 g) was weighed and added to 10 ml of water. The mixture was shaken on a rotating shaker and allowed to stand overnight. The clear suspension was subjected to determination of soil pH.

3.8 EXPERIMENTAL PROCEDURES

3.8.1 Extraction procedures

Soils

Available sodium, potassium, calcium and magnesium were extracted by shaking 1g aliquots of soil end-over-end for one hour with 10 ml of 1M-ammonium acetate solution adjusted to pH 7 (Jumba, 1989). After settling, the suspension was filtered using Whatman filter paper No.40.

The trace elements Fe, Mn, Cu and Zn were extracted from 1g aliquots by shaking end-over-end for one hour with 10 ml of ammonium acetate-EDTA solution (a mixture of 0.5M ammonium acetate, 0.5M acetic acid and 0.02M ethylene diammine-tetraacetic acid diasodium salt). The extractant was prepared by dissolving 57.1 ml glacial acetic acid, 37.3 ml conc. ammonia solution and 7.44 g Na_2EDTA in distilled water and diluting the mixture to one litre. The pH of the mixture was adjusted to 4.6 with acetic acid or

ammonium hydroxide (Lakanen and Ervio, 1971). The suspension was filtered using Whatman No.42 filter paper and the filtrate retained for analysis.

Forages

Macro elements (Na, K, Ca, Mg and P) and trace elements (Fe, Mn, Cu and Zn) were extracted from forages by wet digestion using an acid mixture (Fick *et al.*, 1979).

To 1g of samples and 1ml distilled water blank, 10 ml of concentrated HNO_3 was added and left for overnight pre-digestion. The samples were heated at 90^0C for one hour. 0.5 ml of concentrated H_2SO_4 and 1 ml concentrated $HClO_4$ was added and heating continued for 30 minutes until white fumes of $HClO_4$ appeared consistently (reflux conditions). The digests were then extracted by boiling in 15 ml of 1M HCl for 5 minutes before diluting to 25 ml with the acid. Charring was prevented by dropwise addition of HNO_3. The cooled extracts were filtered using Whatman No.1 filter paper and stored in polythene bottles for analysis.

Serum

Serum aliquots (1 ml) were deproteinized by shaking with 9 ml of 10 % trichloroacetic acid (TCA) on a vortex mixer and letting it to stand for 10 minutes before centrifuging for 10 minutes at 2500 rpm. The supernatant was separated and stored for macro element (Na, K, Ca, Mg, P) analysis. The rest of serum samples were appropriately diluted and stored in a refrigerator for trace element (Fe, Mn, Cu, Zn, Mo) analysis (Fick *et al.*, 1979).

3.8.2 Analysis procedures

Metals

Sample solutions, calibrating standards and blanks were diluted further to give a suitable working concentration range of 0-10 mg/l for Na and K and 0-5 mg/l for Ca, Mg, Fe, Mn, Cu and Zn. Dilutions for soil extracts were made using the appropriate extracting solution while that of forage digests were made with 1M HCl. The metals were determined by AAS using an air/acetylene flame and manufacturer's recommended instrumental settings. For Ca and Mg analysis, standards and samples were enriched with La^{3+} at a concentration of 0.31 % to eliminate phosphate interference during analysis (Oduor, 2002).

Phosphorous

Phosphorus in the digests was determined by reacting it with excess molybdate ions in the presence of ammonium metavanadate in acid medium to form a yellow coloured complex whose intensity, attributed to substitution of oxyvanadium and oxymolybdenum radicals for oxygen of phosphate, was measured.

Sample solutions, calibrating standards and blanks were diluted further to give a suitable working concentration range of 0-20 mg/l P. To 5 ml of working standard and unknown extract containing upto 0.2 mg P, 0.5 ml of 60 % $HClO_4$ was added followed by 1 ml stock vanado-molybdate reagent (50:50 mixture of 0.25 % ammonium metavanadate in 33 % HNO_3 and 5 % w/v aqueous ammonium molybdate). The contents were mixed,

diluted to 10 ml with de-ionized water and let to stand for 10 minutes. The contents were further shaken on a vortex mixer before taking absorption measurements of the yellow complex at 470 nm.

Molybdenum

Molybdenum in serum was determined spectrophotometrically by a modification of the method of Bingley (1963). Each test sample (10 ml) had its pH adjusted to 1 with ammonium hydroxide or hydrochloric acid. To the 10 ml test sample solution, 0.25 ml of Fe^{3+} solution (10 % w/v ferric ammonium sulphate in 2 % sulphuric acid) was added and its volume adjusted to 25 ml with deionised water. A 0.25 ml aliquot of 50 % w/v potassium iodide was added and the mixture allowed to stand for 10 minutes with occasional swirling. A 10 % sodium thiosulphate solution was added dropwise followed by 0.25 ml of 50 % aqueous tartaric acid and 2 ml of 10 % thiourea. After mixing, dithiol reagent (2 ml of 2 % 4-methyl1, 2-dimercaptobenzene in 1 % sodium hydroxide) was added; the contents were mixed and allowed to stand for 30 minutes. The Mo complex was extracted by shaking with 5 ml of isoamylacetate (Boiling point range 136-142^{0}C) for 30 minutes. The organic layer was separated, centrifuged for 5 minutes before measuring the molecular absorption intensity of the green Mo-dithiol complex colorimetrically at 680 nm using the isoamylacetate in the reference channel. Calibrating standards containing 0-10 μg/ml Mo were made to 10 ml with deionized water (DIW) and treated in accordance with the sample procedure starting with the pH adjustment.

3.9 STATISTICAL ANALYSIS OF DATA

Data from soil, forage and serum were statistically analysed by two models: a mixed model for soil and forage data (Snedecor and Cochran, 1984) and a fixed model for serum (Montgomery, 1984). This was computed using the Generalized Linear Model (GLM) procedure of the Statistical Analysis System (SAS, 1985). Analysis from two means obtained was based on Duncan's multiple range tests to find significant differences between the means. Determination of significant differences entailed comparing means of relevant quantities using the BIVARIATE procedure of SAS. The analysis was carried out at the significance levels P=0.05 and P=0.01. In addition to the analysis of variance, spearman's rank correlation for stipulated variables were used to establish relationships between parameter values observed in the soil, forage and blood serum for various mineral elements.

CHAPTER FOUR
RESULTS AND DISCUSSION

4.1 INTRODUCTION

Mineral requirements for grazing livestock were determined from both field and laboratory experiments. Various authorities have suggested different mineral requirements for grazing livestock for different stages of growth; young growing calves, pregnant or lactating cows. The authorities considered in this work were Agricultural Research Council (ARC, 1980), National Research Council (NRC, 1985, 1989, 2001) and publications by workers like McDowell *et al.*, (1993) and McDowell, (1985) who have given critical levels below which mineral deficiency is anticipated. The concentrations of all parameters in soils and forages have been expressed in mg/kg and g/kg on dry matter (DM) basis while those parameters in serum are expressed in g/l for macro elements and µg/ml for trace elements. Conversion from one unit to another may be necessary for discussion purposes.

The parameters determined along with their symbols included macro elements: sodium (Na), potassium (K), calcium (Ca), magnesium (Mg), phosphorus (P) and trace elements: iron (Fe), manganese (Mn), copper (Cu), zinc (Zn) and molybdenum (Mo). This Chapter reports the mineral concentrations in soils, forages and serum in the three regions of Uasin Gishu district and by inference compare the overall means of these mineral concentrations with standard mineral requirements so as to identify those that might be limiting animals' health and production.

4.2 CONCENTRATIONS OF MINERALS IN SAMPLES

This section gives a discussion of the mineral concentrations from analyses of soil, forage and serum as shown in the sections below.

4.2.1 SOIL ANALYSIS

Macro and trace element analysis was done on the selected soils in this study. The results are discussed in the sections below.

4.2.1.1 Macro elements

Table 4.1 gives the mean concentrations and range of macro elements in soils analyzed in this study (Appendix V).

Table 4.1 The mean concentrations and range of macro elements in soils (mg/kg DM)

Para.	Mean±SD (n=28)	Range (n=28)	Central±SD (n=16)	North±SD (n=8)	South±SD (n=4)
Na	101.00 ± 32.00	58.13-155.00	88.00±22.30	109.00±36.52	135.00±27.39
K	709.00 ± 24.40	189.10-1135.00	709.00±20.77	756.00±38.68	614.00±181.04
Ca	720.00 ± 33.30	280.00-1840.00	667.00±40.83	805.00±105.07	765.00±148.85
Mg	330.00 ± 11.90	161.30-627.70	317.00±90.84	332.00±94.11	381.00±104.26

From Table 4.1, the concentration of sodium in soils had a range of 58.13-155.00 mg/kg with a mean of 101 mg/kg (Table 4.1). The concentration of potassium in soils was 709 mg/kg with a range of 189.10-1135.00 mg/kg. The soils revealed high concentrations of calcium ranging between 280-1840 mg/kg with a mean of 720 mg/kg. The mean concentrations of magnesium in soils ranged from 161.30-627.70 mg/kg with a mean

concentration of 330 mg/kg. Examination of results in Table 4.2 revealed that the concentrations of all the macro elements were above the recommended soil critical levels (Rhue and Kidder, 1983; Breland, 1976). In Table 4.2, the %sample deficiency refers to the number of soil samples that had macro element concentrations below recommended levels.

Table 4.2 Macro element concentrations (mg/kg DM) in soils in relation to recommended levels

Parameter	Mean±SD (n=28)	Range (n=28)	Critical levels	%Samples deficient
Na	101.00 ± 32.00	58.13-155.00	55	0
K	709.00 ± 24.40	189.10-1135.00	60	0
Ca	720.00 ± 33.30	280.00-1840.00	71[a]	0
Mg	330.00 ± 11.90	161.30-627.70	30	0

(Critical level source: Rhue and Kidder, 1983; [a] Breland, 1976)

On the basis of regions, the concentration of sodium was highest for southern region in soils (135 mg/kg) with the differences when compared with central (88.80 mg/kg) and northern (109 mg/kg) regions being significant ($P<0.05$). However, the concentration of potassium was higher in the northern region (756 mg/kg) than in the southern (614 mg/kg) and central (709 mg/kg) soils with the difference being also significant ($P<0.05$). The difference in concentration of calcium in soils between the highest in the north (805 mg/kg) and the lowest in central (667 mg/kg) was significant ($P<0.05$). The concentration of magnesium was higher in soils from the southern region (381 mg/kg) than from central (317 mg/kg) and northern (332 mg/kg) regions ($P<0.05$). All the regions had samples with macro element concentrations above the recommended critical levels. However the

southern region recorded the lowest levels of potassium while calcium and magnesium were lowest in central region.

These results are in agreement with similar studies in the tropics. The concentration of sodium in soil obtained in this study (101 mg/kg) was lower than 155 mg/kg reported for western Kenya (Oduor, 2002), but higher than 6.6 mg/kg of North Florida (Cuesta *et al.*, 1993) and 88 mg/kg of South Eastern Venezuela (Jerez *et al.*, 1984). The concentration of potassium in soil (709 mg/kg) was higher than 379 mg/kg of Mt. Elgon region in western Kenya (Oduor, 2002), 18.9 mg/kg of North Florida (Cuesta *et al.*, 1993) and 114 mg/kg of Dominican Republican (Jerez *et al.*, 1984). The calcium concentrations obtained in soil from this study (720 mg/kg) was also higher than 92.7 mg/kg of Mt. Elgon region in western Kenya (Oduor, 2002), 3.8 mg/kg of North Florida (Cuesta *et al.*, 1993) and 219 mg/kg of South Eastern Venezuela (Jerez *et al.*, 1984). The concentration of magnesium in soil of 330 mg/kg was higher than 37.2 mg/kg of Mt. Elgon region in western Kenya (Oduor, 2002), 2.28 mg/kg of North Florida (Cuesta *et al.*, 1993) and 38.9 mg/kg of Dominican Republican (Jerez *et al.*, 1984).

4.2.1.2 Trace elements

Table 4.3 gives the mean concentrations and range of trace elements in soils determined in this study (Appendix VI). From Table 4.3, the concentration of iron in soils was 551 mg/kg with a range of 228.30-1198.30 mg/kg. The concentrations of manganese in soils ranged between 239.70-1583.50 mg/kg with mean concentrations of 630 mg/kg.

Table 4.3 The mean concentrations and range of trace elements in soils (mg/kg DM)

Para	Mean±SD (n=28)	Range (n=28)	Central±SD (n=16)	North±SD (n=8)	South±SD (n=4)
Fe	551.00 ± 22.80	228.30-1198.30	598.00±115.46	526.00±103.80	412.00±107.29
Mn	630.00 ± 34.50	239.70-1583.50	703.00±180.37	536.00±139.27	525.00±109.65
Cu	3.30 ± 0.90	1.60-5.13	3.00±0.89	3.80±0.67	3.50±1.50
Zn	6.70 ± 0.40	1.97-22.06	8.00±3.52	5.50±2.43	4.10±1.90

The copper concentrations in soils were within a range of 1.60-5.13 mg/kg with a mean of 3.3 mg/kg. The soils had zinc concentration of 6.70 mg/kg with a range of 1.97-22.06 mg/kg.

The concentration of iron in soils was higher in central region (598 mg/kg) than in southern region (412 mg/kg) and northern region (526 mg/kg) with the difference being significant ($P<0.05$). The difference in concentration of manganese between the highest in central (703 mg/kg) and the lowest in the south (525 mg/kg) was also significant ($P<0.05$). The concentration of copper in soils was higher in the north (3.80 mg/kg) than in central (3.0 mg/kg) and in the south (3.5 mg/kg) with significant differences ($P<0.05$). The concentration of zinc was significantly higher in central region (8.00 mg/kg) than in the south (4.10 mg/kg) in soils ($P<0.05$). The southern region actually recorded very low concentrations of iron, manganese and zinc. Analysis of the data whose results are presented in Table 4.4 revealed some trace element deficiencies in soils.

Table 4.4 Trace element concentrations (mg/kg DM) in soils in relation to recommended levels

Parameter	Mean±SD (n=28)	Range (n=28)	Critical levels	%samples deficient
Fe	551.00 ± 22.80	228.30-1198.30	30	0
Mn	630.00 ± 34.50	239.70-1583.50	10 [a]	0
Cu	3.30 ± 0.90	1.60-5.13	2 [b]	14
Zn	6.70 ± 0.40	1.97-22.06	2 [b]	4

(Source: Critical levels, Bahia, 1978; [a] Mtimuni, 1982; [b] McDowell *et al.*, 1983)

All the soil samples had iron and manganese concentrations above the critical level (C L) of 30 mg/kg (Bahia, 1978) and 10 mg/kg DM (Mtimuni, 1982), respectively. Other studies have revealed similar high levels of iron and manganese in soils. The concentration of iron in soil of 551 mg/kg was significantly higher than 460 mg/kg of Mt. Elgon region in western Kenya (Oduor, 2002), 505 mg/kg of Columbia (Pastrana *et al.*, 1991b) and 50 mg/kg of South Eastern Venezuela (Rojas *et al.*, 1993). The soil manganese concentrations obtained (630 mg/kg) was also significantly higher than 300 mg/kg of Mt. Elgon region in western Kenya (Oduor, 2002), 49 mg/kg of Columbia (Pastrana *et al.*, 1991b) and 49.5 mg/kg of South Eastern Venezuela (Rojas *et al.*, 1993).

Copper and zinc concentrations in 14 % and 4 % of these samples, respectively were below critical levels of 2 mg/kg DM (McDowell *et al.*, 1983). These results are similar to those of Mt. Elgon region in western Kenya, which revealed 26 % and 66 % of soils being deficient in copper and zinc, respectively. This could also be true for other areas since copper levels in this study (3.30 mg/kg) were significantly higher than 0.7 mg/kg of Columbia (Pastrana *et al.*, 1991b) and 1.18 mg/kg of South Eastern Venezuela (Rojas *et*

al., 1993). The zinc concentration in soil (6.7 mg/kg) was also significantly higher than 2.9 mg/kg of Columbia (Pastrana *et al.*, 1991b) and 1.42 mg/kg of South Eastern Venezuela (Rojas *et al.*, 1993).

4.2.2 FORAGE ANALYSIS

Macro and trace element analysis was done on selected forage in this study. The results are discussed in the following sections.

4.2.2.1 Macro elements

Table 4.5 gives the mean concentrations of macro elements in forages assessed in this study (Appendix VII).

Table 4.5 The mean concentrations and range of macro elements (g/kg DM)

Parameter	Mean±SD (n=28)	Range (n=28)	Central ±SD (n=16)	North±SD (n=8)	South±SD (n=4)
Na	1.00 ± 0.30	0.55-2.12	0.92±0.17	0.94±0.22	1.47±0.60
K	11.80 ± 5.00	3.73-20.53	11.08±5.50	14.00±4.34	10.50±3.60
Ca	0.57 ± 0.19	0.15-1.17	0.56±0.19	0.54±0.08	0.72±0.29
Mg	1.35 ± 0.72	0.10-3.13	1.36±0.50	1.42±0.81	1.18±0.90
P	6.34 ± 3.22	1.83-14.41	6.69±3.02	6.69±2.07	4.23±1.51

The forages had sodium concentrations, which ranged from 0.55-2.12 g/kg with a mean concentration of 1.00 g/kg. The mean concentration of potassium in forages was 11.80 g/kg with ranges of 3.73-20.53 g/kg. Forages had calcium concentrations, which ranged from 0.15-1.17 g/kg with a mean concentration of 0.57 g/kg. The mean concentration of magnesium in forages was 1.35 g/kg with a range of 0.10-3.13 g/kg. Forages had

phosphorus concentrations, which ranged from 1.83-14.41 g/kg with a mean concentration of 6.34 g/kg (Table 4.5).

The forages from the southern region revealed significantly higher levels ($P<0.05$) of sodium (1.47 g/kg) than those from central (0.92 g/kg) and northern regions (0.94 g/kg). Concentration of potassium in forages was however higher in the north (14 g/kg) than in the south (10.50 g/kg) and in central (11.08 mg/kg) with the difference being significant ($P<0.05$). The difference in concentration of calcium in forages between the highest in the south (0.72 mg/kg) and the lowest in the north (0.54 mg/kg) was also significant ($P<0.05$). The concentration of magnesium was higher in the northern region (1.42 g/kg) than in central (1.36 g/kg) and in the south (1.18 g/kg) with significant differences ($P<0.05$). The concentration of phosphorus in forages was the same in the central and northern region (6.69 g/kg), which was higher than that for southern region (4.23 g/kg) with the difference being significant ($P<0.05$). Analysis of the data whose results are presented in Table 4.6 revealed that forages were critically deficient in mainly calcium and magnesium in relation to both calves' and cows' needs.

Table 4.6 The mean concentrations of macro elements in forages (g/kg DM) in relation to ruminants' needs

Parameter	Mean ±S.D	CL for Calves	CL for Cows	%Sample def. (Calves)	%Sample def. (Cows)
Na	1.00 ± 0.33	0.60-1.00	1.80	7	93
K	11.80 ± 5.00	5.00-7.00	9.00	7	36
Ca	0.57 ± 0.19	0.80-15.00	0.80-15.00	86	86
Mg	1.35 ± 0.71	0.50-3.00	2.10	86	89
P	6.34 ± 3.22	2.00-6.00	3.40	4	25

C L - source: calves (McDowell *et al.*, 1983; 1993); cows (McDowell *et al.*, 1984; 1993)

These results (Table 4.6) revealed that 7 % of the forages had sodium concentrations below the ruminants' needs of 0.60-1.0 g/kg DM required for growing calves (McDowell *et al.*, 1983; 1993) while 93 % of the forage samples had sodium concentrations below the required 1.8 g/kg DM for lactating cows (McDowell *et al.*, 1984; 1993). About 7 % of forages had potassium concentrations below a range of 5.0-7.0 g/kg DM recommended for growing calves (McDowell *et al.*, 1983; 1993) while 36 % of the forages sampled had potassium concentrations below 9 g/kg DM for lactating cows (McDowell *et al.*, 1984; 1993). The results also revealed that 86 % of forages had calcium concentrations below the range of 0.80-15.0 g/kg DM for both growing calves (McDowell *et al.*, 1983; 1993) and lactating cows (McDowell *et al.*, 1984; 1993). Majority of the samples (86 %) had magnesium concentrations below a range of 0.50-3.0 g/kg DM for growing calves (McDowell *et al.*, 1983; 1993) while 89 % of samples were also below 2.10 g/kg DM for lactating cows (McDowell *et al.*, 1984; 1993). The 4 % and 25 % of forages sampled had phosphorus concentrations below the recommended range of 2.0-6.0 g/kg DM for growing calves (McDowell *et al.*, 1983; 1993) and 3.40 g/kg for lactating cows respectively (McDowell *et al.*, 1984; 1993). All the three regions recorded calcium levels below the recommended 0.8-15.0 g/kg for both calves and cows while all magnesium concentrations were below the required 2.10 mg/kg for lactating cows. This shows that grazing cattle in Uasin Gishu district suffer from calcium and magnesium deficiencies similar to those in Trans Nzoia district.

Macro element deficiencies were also detected in forages from Mt. Elgon region in western Kenya (Oduor, 2002), which reported major deficiencies in calcium (78 %),

magnesium (70 %) and phosphorus (95.5 %). These macro element deficiencies seem to be widespread in the tropics since similar studies have recorded lower magnesium levels of 1.7 g/kg and 3.8 g/kg in Dominican Republican and Guatemala (Valdez *et al.*, 1988), respectively. A similar study reported wide variations in magnesium content among several plants and concluded that plant breeding had considerable potential for reducing mineral composition in many forage species (Hacker, 1982). Nitrogen level, stages of maturity, excessive potassium level and readily fermentable carbohydrates have also been considered as factors influencing magnesium utilization (Rosero *et al.*, 1980). Excessive or high levels of potassium usually antagonize magnesium utilization. The higher potassium levels from this study could be responsible for lower magnesium levels due to K-Mg interactions. High levels of potassium in soils could cause low uptake of magnesium by plant species, which could be antagonistic to magnesium absorption hence limiting its availability to plants (Gatahi, 1986).

The concentration of calcium in forages (0.57 g/kg) was lower than 4.8 g/kg and 3.5 g/kg of Dominican Republican (Jerez *et al.*, 1984) and Guatemala (Valdez *et al.*, 1988), respectively. The low levels of calcium in forages could be due to the stage of maturity at which forage was sampled (hay stage). The mineral content in mature plants is usually lower than in young forages (Tergas and Blue, 1971). The levels of calcium in forages from this study imply that prospects of deficiency could be anticipated in ruminants in the study area. The forages had phosphorus concentration (6.34 g/kg) higher than 1.8 g/kg of Nakuru national park (Siva, 1996) and 1.0 g/kg of Guatemala (Valdez *et al.*, 1988) respectively. A positive relationship between amount of phosphorus absorbed and lost,

and the amount of phosphorus ingested increases with dry matter intake especially when the diet contains coarse roughage (Tergas and Blue, 1971). The increased availability of forage may increase phosphorus demand by either not releasing phosphorus in the gastro-intestinal tract or phosphorus sticking to soil particles or molecules as it passes down the tract. Such molecules include complexes of calcium, aluminium and iron (Rosa *et al.*, 1982; Field *et al.*, 1983). This means that dietary composition of the interfering elements of Ca, Al and Fe needs to be controlled for efficient phosphorus utilization. Research revealed that endogenous feacel phosphorus is increased as dietary phosphorus absorption is increased especially when phosphorus is sequestered within the gastro-intestinal tract (Braithwaite, 1985). Soil ingestion has also been found to increase iron, which can antagonize utilization of essential mineral such as phosphorus (McDowell, 1985). The fact that only 7 % and 36 % of the forage sampled had potassium concentrations below the recommended standards for calves and cows, respectively indicates no major deficiency problems in the area.

4.2.2.2 Trace elements

Table 4.7 gives the mean concentrations of trace elements in forages considered in this study (Appendix VIII). The iron concentration in forages had a mean of 56 mg/kg with a range of 10.25-231.00 mg/kg while that of manganese was 105 mg/kg with values ranging from 38.25-246.50 mg/kg (Table 4.7). The mean concentration of copper and zinc in the forages were 5.32 mg/kg and 19.50 mg/kg with ranges of 2.00-15.50 mg/kg and 8.30-39.78 mg/kg, respectively.

Table 4.7 The mean concentrations and ranges of trace elements in forages (mg/kg DM)

Parameter	Mean±SD (n=28)	Range (n=28)	Central±SD (n=16)	North±SD (n=8)	South±SD (n=4)
Fe	56.00 ± 0.53	10.25-231.00	50.00±10.36	57.16±26.60	80.56±20.59
Mn	105.00 ± 0.58	38.25-246.50	119.00±30.65	85.59±21.07	89.63±30.80
Cu	5.32 ± 2.84	2.00-15.50	4.95±1.09	5.56±2.06	6.31±2.15
Zn	19.50 ± 8.26	8.30-39.78	20.12±7.59	17.20±6.70	21.65±10.01

The southern region recorded the highest concentrations ($P<0.05$) of iron (80.56 mg/kg), copper (6.31 mg/kg) and zinc (21.65 mg/kg) relative to central and northern regions. The northern region was lowest ($P<0.05$) in zinc (17.20 mg/kg) when compared to central (20.12 mg/kg) and southern (21.65 mg/kg) regions. Analysis of the data revealed some percent trace element deficiencies in the forages (Table 4.8). All the regions recorded copper concentration of 4.95 mg/kg (central), 5.56 mg/kg (north) and 6.31 mg/kg, (south) which were far below the critical level of 9 mg/kg, hence indicating deficiency. The Northern region recorded 17.20 mg/kg for zinc, which was below the recommended 18 mg/kg for both calves and cows. This was in agreement with the study in Trans Nzoia bordering Uasin Gishu, which revealed copper and zinc deficiencies (Jumba, 1989).

Table 4.8 The mean concentrations and ranges of trace elements in forages (mg/kg DM) in relation to the critical levels of calves and cows needs

Parameter	Mean ±S.D (n=28)	Range (n=28)	C L Calves/cows	%Samples deficient
Fe	56.00 ± 5.30	10.25-231.00	<13.00	7
Mn	105.00 ± 5.80	37.75-246.50	<13.00	0
Cu	5.32 ± 2.84	2.00-15.50	<9.00	93
Zn	19.50 ± 8.26	8.30-39.78	<18.00	39

CL –Critical level based on calves and cows needs (NRC, 2001)

The results (Table 4.8) revealed acute deficiencies in copper (93 %) and no major deficiency in zinc (39 %). The deficiencies could possibly be due to the 14 % and 4 % of the soil samples, which had copper and zinc concentrations, respectively below the recommended requirements (Table 4.4). Available Zinc could also be further lower in the event of high concentrations of interfering (antagonistic) elements such as Ca, Cu, Fe and Cd being present in the ruminants diet (Appleton, 1994). Deficiencies in Cu and Zn seem to be widespread since similar studies from Mt. Elgon (Oduor, 2002) and Trans Nzoia (Jumba, 1989) regions in western Kenya bordering Uasin Gishu in the north, revealed copper and zinc deficiencies. This could also be true for other tropical regions since copper levels in this study (3.30 mg/kg) were higher than 0.7 mg/kg of Columbia (Pastrana *et al.*, 1991b) and 1.18 mg/kg of South Eastern Venezuela (Rojas *et al.*, 1993). The iron concentration in forages (56 mg/kg) was lower than 760 mg/kg of Mt. Elgon region in western Kenya, 135 mg/kg of Columbia (Pastrana *et al.*, 1991b) and 579 mg/kg of South Eastern Venezuela (Rojas *et al.*, 1993). The manganese concentration in forages (105 mg/kg) was also lower than 220 mg/kg of Mt. Elgon region in western Kenya, 309 mg/kg and 203 mg/kg of Columbia and Malawi (Mtimuni *et al.*, 1990), respectively.

The main problem with iron and manganese is usually that of excess intakes in feeds (McDowell *et al.*, 1993). This is evident from this study in which 93 % and 100 % of forage samples had high levels of iron and manganese minerals, respectively (Table 4.8). Concentrations of 500 mg/kg or above in feeds usually depress appetite and retard growth (McDowell and Conrad, 1989). The critical levels of iron for growing calves and lactating cows are 25 mg/kg DM and 40 mg/kg DM, respectively (ARC, 1980;

McDowell *et al.*, 1993). Although high iron levels have been reported in forage, high plant accumulated iron may also be as a result of soil-forage contamination during grazing. This study has revealed high concentrations of iron and manganese in soil with no prospects of deficiency. However the high iron levels may cause interference in absorption and utilization of copper in plants (Bremner *et al.*, 1983).

On the other hand, forage had generally low levels of copper, which is usually antagonized by high iron-copper concentrations through Fe-Cu interactions. Though molybdenum was not analyzed in the forage samples, its presence in high quantities is known to affect copper absorption and metabolism through possible formation of insoluble copper thiomolybdate complexes in the gut (Suttle, 1975). Copper deficiency has been categorized into four groups according to the composition of feed: i) high level of Mo and Cu (>20 mg/kg DM), ii) low Cu but significant amount of Mo (e.g. ratio <2:1), iii) insufficient Cu (<5 mg/kg DM) and iv) normal Cu and low Mo, with high levels of soluble protein (Suttle, 1975). The results of this study fall in category iii) corresponding to insufficient copper levels. Distribution of copper deficiency problems worldwide has been associated with various types of soils (McDowell *et al.*, 1993). The occurrence of acute Cu-Mo problems could arise from either Molybdeniferous clays as in England (Thornton, 1977). The present study area contained poorly drained, almost clay soils that are more likely to cause Cu-Mo interaction problems (MPND, 2002).

4.2.3 SERUM ANALYSIS

Macro and trace element analysis was done on serum of selected animal blood in this study. The results are discussed in the following sections.

4.2.3.1 Macro elements

Table 4.9 and 4.10 give concentrations of the macro elements in animal serum investigated in this study (Appendix IX; X).

Table 4.9 The mean concentrations and ranges of macro elements of serum (g/l) in young calves

Parameter	Mean±SD (n=18)	Range (n=18)	Central±SD (n=10)	North±SD (n=5)	South±SD (n=3)
Na	2.45 ± 0.42	1.91-3.64	2.42±0.42	2.18±0.26	2.60±0.46
K	0.37 ± 0.08	0.25-0.58	0.37±0.09	0.32±0.04	0.41±0.02
Ca	0.68 ± 0.21	0.29 –1.14	0.66±0.22	0.53±0.22	0.74±0.09
Mg	0.02 ± 0.01	0.001-0.09	0.02±0.02	0.03±0.03	0.01±0.00
P	0.17 ± 0.10	0.07-0.52	0.17±0.10	0.12±0.04	0.16±0.06

Table 4.10 The mean concentrations and ranges of macro elements of serum (g/l) in lactating cows

Parameter	Mean±SD (n=24)	Range (n=24)	Central±SD (n=14)	North±SD (n=7)	South±SD (n=3)
Na	2.38 ± 0.61	1.13-4.04	2.56±0.62	1.99±0.54	2.44±0.21
K	0.39 ± 0.06	0.28-0.58	0.39±0.07	0.38±0.03	0.44±0.04
Ca	0.81 ± 0.32	0.49-2.16	0.84±0.41	0.75±0.11	0.78±0.06
Mg	0.02 ± 0.01	0.003-0.04	0.02±0.01	0.02±0.01	0.03±0.01
P	0.15 ± 0.05	0.07-0.22	0.15±0.05	0.13±0.05	0.13±0.04

The concentration of sodium in calves (Table 4.9) ranged from 1.91-3.64 g/l having a mean of 2.45 g/l while lactating cows (Table 4.10) had a mean concentration of 2.38 g/l with a range of 1.13-4.04 g/l. Serum samples from young calves revealed potassium concentrations in the range of 0.25-0.58 g/l with a mean of 0.37 g/l while lactating cows recorded a mean of 0.39 g/l with a range between 0.28-0.58 g/l. The concentration of calcium in calves ranged from 0.29-1.14 g/l having a mean of 0.68 g/l while lactating cows had concentration of 0.81 g/l with a range of 0.49-2.16 g/l. Serum samples from young calves revealed magnesium concentration in the range of 0.001-0.09 g/l with a mean of 0.02 g/l while lactating cows recorded a mean of 0.02 g/l with a range between 0.003-0.04 g/l. Serum samples had phosphorus concentrations, which ranged from 0.07-0.52 g/l with a mean concentration of 0.17 g/l in calves while lactating cows' serum had mean concentration of 0.15 g/l with a range of 0.07-0.22 g/l.

The southern and central regions recorded the highest concentrations of sodium in calves (2.60 g/l) and cows (2.56 g/l), respectively ($P<0.05$). The results show that the southern region recorded the highest potassium concentration in both calves (0.41 g/l) and cows (0.44 g/l) with significant differences when these concentrations were compared with other regions ($P<0.05$). The results revealed that the southern and central regions recorded the highest calcium concentrations in calves (0.74 g/l) and cows (0.84 g/l), respectively. Also there were significant differences when these values for calves were compared with central and northern regions and cows' values with southern and northern regions ($P<0.05$). The northern and southern regions recorded the highest magnesium concentrations (0.03 g/l) in both calves and cows. There were no significant differences

when these calves' values were compared with central and southern, and cows' values compared to central and northern ($P<0.05$). The concentration of phosphorus was highest in central for both calves (0.17 g/l) and cows (0.15 g/l) with significant differences when these values were compared to northern (calves:0.12 g/l; cows:0.13 g/l) and southern (calves:0.16 g/l; cows:0.13 g/l) regions.

Analysis of the results (Table 4.11) revealed that none of the calves' and cows' serum samples had macro elements' concentrations below the recommended levels (McDowell *et al.*, 1985; 1993) except for magnesium. These findings are in agreement with those of Mt. Elgon region in western Kenya, which reported no deficiencies in sodium and potassium (Oduor, 2002). Young calves and lactating cows from the three regions considered in this study recorded magnesium concentrations of 0.001-0.09 g/l and 0.003-0.04 g/l, respectively which revealed deficiencies.

Table 4.11 Mean serum macro-element concentrations (g/l) for calves and lactating cows in relation to recommended levels in animal blood

Parameter	Mean ±S.D (Calves (n=18)	Mean ±S.D (Cows) (n=24)	C L for Calves/ Cows	% Samples deficient (Calves)	%Samples deficient (Cows)
Na	2.45 ± 0.42	2.38 ±0.61	0.25	0	0
K	0.37 ± 0.08	0.39± 0.06	0.03[a]	0	0
Ca	0.68 ± 0.21	0.81 ±0.32	0.08	0	0
Mg	0.02 ± 0.01	0.02 ±0.01	0.015	50	21
P	0.17 ± 0.10	0.15 ±0.05	0.045	0	0

C L-Critical level (McDowell et al, 1993); [a]Critical level (McDowell et al, 1985)

The calves' and cows' serum revealed no deficiencies in sodium, potassium calcium and phosphorus elements. Sodium requirement for ruminants is usually at 0.25 g/l of diet (McDowell *et al.*, 1993) while that of potassium is 0.03 g/l (McDowell *et al.*, 1985). Initial sodium deficiency sign is demonstrated in animal craving for salt by licking wood, and soil while potassium deficiency is shown by reduced feed intake (McDowell *et al.*, 1993). This was confirmed by the entire serum sample having Na and K concentrations above the critical levels. The fact that 93 % of forages sampled were deficient in sodium for lactating cows contradicts results from soil analysis, which showed no deficiency. This could be due to the type and stage of forage species sampled which influence mineral concentration. Evaluation of potassium deficiency is usually difficult since the high serum potassium may have some diagnostic value for establishing deficiency and may also be caused by malnutrition, negative nitrogen balance, gastro-intestinal losses and endocrine malfunction. The high potassium levels obtained in cattle from this study may lead to adverse implications including mineral interactions such as Na-K and Mg-K. This usually leads to a reduction in sodium and magnesium levels.

The results further revealed 50 % of the calves' and 21 % of the cows' serum sample had magnesium levels below the CL of 0.015 g/l (McDowell *et al.*, 1993; Mtimuni, 1982). This correlates to forage analysis, which revealed 86 % and 89 % of samples with magnesium concentrations below critical levels for calves and cows, respectively. Prospects of tetany needs to be investigated since levels of Mg below 1.0 g/Kg DM in forages and in some blood samples indicated lower serum magnesium levels than the recommended requirement of 0.015 g/l. This suggests occurrence of magnesium

deficiency in calves and lactating cows. Given that magnesium concentrations in blood do not fall until there is severe deficiency, levels of below 0.015 g/l obtained in this study indicates magnesium deficiency in the region.

While soil analysis revealed no calcium deficiency, 86 % of forages sampled were found to be deficient in calcium for both young calves' and lactating cows' requirements. However, cattle serum samples revealed no deficiencies in calcium. This could be attributed to the supply of calcium through other sources such as soil ingestion. The high serum calcium could also be as a result of other diet given to the animals. Typical symptoms of calcium deficiency may not develop immediately. It may be true that grazing cattle in the region could be experiencing calcium deficiency. This is because animals usually tolerate low dietary concentrations of Ca for fairly long periods (even up to six months) by increasing capacity for intestinal absorption of calcium (Siva, 1996). The results obtained further revealed that while 25 % of forages were deficient in phosphorus, none of the serum sampled were low in phosphorus for lactating cows. This could be due to the fact that ruminants usually accumulate phosphorus in their tissues during periods of generous dietary forage supply and mobilize them in periods of shortage as the bone responds to the demands of phosphorus (Jumba, 1989).

4.2.3.2 Trace elements

Table 4.12 and 4.13 give concentrations and ranges of the trace elements in animal serum investigated in this study (Appendix XI; XII). The concentration of iron in calves (Table 4.12) ranged from 0.20-7.30 µg/ml having a mean of 2.28 µg/ml while lactating cows (Table 4.13) had concentration of 2.43 µg/ml with a range of 0.40-6.10 µg/ml. Serum

samples had manganese concentrations, which ranged from 0.10-0.60 µg/ml with a mean concentration of 0.27 µg/ml in calves while lactating cows' serum had mean concentration of 0.26 µg/ml with a range of 0.10-0.70 µg/ml.

Table 4.12 The mean concentrations and ranges of trace elements in calves (µg/ml)

Parameter	Mean±SD (n=18)	Range (n=18)	Central±SD (n=10)	North±SD (n=5)	South±SD (n=3)
Fe	2.28± 1.13	0.20 –7.30	1.98±0.91	2.44±0.38	2.73±0.90
Mn	0.27 ±0.15	0.10-0.60	0.27±0.16	0.26±0.13	0.23±0.05
Cu	0.60 ±0.19	0.30-1.00	0.65±0.19	0.66±0.20	0.40±0.10
Mo	0.32± 0.38	0.07-1.16	0.36±0.13	0.41±0.05	0.27±0.10
Zn	3.11 ±1.23	0.93-5.00	3.17±1.30	3.36±1.20	3.16±0.57

Table 4.13 The mean concentrations and ranges of trace elements in cows (µg/ml)

Parameter	Mean±SD (n=24)	Range (n=24)	Central±SD (n=14)	North±SD (n=7)	South±SD (n=3)
Fe	2.43 ± 1.53	0.40-6.10	1.86±1.20	3.13±1.92	3.43±0.08
Mn	0.26 ± 0.14	0.10-0.70	0.28±0.15	0.27±0.12	0.17±0.05
Cu	0.60 ± 0.17	0.20-1.00	0.56±0.17	0.61±0.08	0.77±0.20
Mo	0.36 ± 0.35	0.07-1.31	0.33±0.03	0.56±0.13	0.07±0.00
Zn	3.71 ± 1.63	0.65-6.84	4.04±0.43	2.84±1.87	4.23±1.43

The serum samples from young calves revealed copper concentration in the range of 0.30-1.00 µg/ml with a mean of 0.60 µg/ml while lactating cows recorded a mean of 0.60 µg/ml with a range between 0.20-1.00 µg/ml. Serum had molybdenum concentrations of 0.32 µg/ml in calves and 0.36 µg/ml for cows. These molybdenum concentrations were approximately half of those for copper in both calves and cows. Serum samples from young calves revealed zinc concentration in the range of 0.93-5.00 µg/ml with a mean of

3.11 µg/ml while lactating cows recorded a mean of 3.71 µg/ml with a range between 0.65-6.84 µg/ml.

The southern region recorded higher iron concentrations (2.73 µg/ml) than central region (1.98 µg/ml) in calves with the difference being significant ($P<0.05$). Cows serum on the other hand also recorded higher iron concentrations in the southern region (3.43 µg/ml) than the central region (1.86 µg/ml) with the difference being significant ($P<0.05$). The concentration of manganese was highest in central in both calves (0.27 µg/ml) and cows (0.28 µg/ml) and lowest in southern in calves (0.23 µg/ml) and cows (0.17 µg/ml) with significant differences ($P<0.05$) when these values were compared. The results further revealed that the northern and southern regions recorded the highest copper concentrations in both calves (0.66 µg/ml) and cows (0.77 µg/ml), respectively. Significant differences were observed when these values compared with those of central and southern (calves) and central and northern regions ($P<0.05$). The northern and southern regions also recorded the highest zinc concentration in calves (3.36 µg/ml) and cows (4.23 µg/ml), respectively. However, analysis of data (Table 4.14) revealed that 28 % of the calves' and 29 % of the cows' serum samples had iron concentrations below the critical level of 1 µg/ml (McDowell *et al.*, 1993). This may be due to the fact that the 7 % of the forage samples (Table 4.8) were deficient in iron for both young calves and lactating cows (NRC, 2001).

Table 4.14 Mean serum trace element concentrations (µg/ml) for calves and lactating cows in relation to recommended levels in animal blood

Parameter	Mean ±S.D (Calves)	Mean ±S.D (Cows)	C L Calves/ Cows	% Samples deficient (Calves)	%Samples deficient (Cows)
Fe	2.28± 1.13	2.43± 1.53	<1.00	28	29
Mn	0.27 ±0.15	0.26 ±0.14	<1.00	100	100
Cu	0.60 ±0.19	0.60± 0.17	<0.65	61	42
Mo	0.32± 0.31	0.36±0.35	-	-	-
Zn	3.11 ±1.23	3.71 ±1.63	<0.70	0	4

CL-Critical level for calves/cows (McDowell *et al.*, 1993)

Iron deficiency was detected in calves with 28 % of the serum samples having iron levels below 1 µg/ml. This is because when calves are fed on an exclusive whole milk diet (milk is low in Fe), they can develop iron deficiency causing anaemia within 2 to 3 months (McDowell *et al.*, 1993). However, iron deficiency in adult cows is rare unless there is blood loss from the animal (Oduor, 2002). This is due to adequate pasture iron concentration usually found together with contaminated iron-rich soil. The fact that this study revealed generally lower manganese levels in serum than the recommended standard levels of 1 µg/ml (McDowell *et al.*, 1993) means that immediate investigations into contributing factors be carried out. The study has obtained lower levels of manganese in serum (0.26 µg/ml), which is in agreement with the study previously carried out Mt Elgon region in western Kenya, which obtained Mn concentrations as low as 0.16 µg/ml in cows (McDowell *et al.*, 1993). While none of the forages sampled had manganese concentrations below critical level for both growing calves and lactating cows (Table 4.8), all the serum samples had manganese concentrations below critical level of 1

µg/ml. This implies serious manganese deficiency for both calves and cows. Comparison of soil manganese concentration revealed no deficiency at all.

This study has also revealed acute copper deficiency, with 93 % of the forage having copper levels below 9 mg/kg DM (Table 4.8); the critical level below which deficiency occurs in both growing and lactating cows (NRC, 1985; NRC, 2001). This could be due to the fact that 14 % of the soil samples were deficient in copper. However, 61 % of the calves' and 42 % of cows' serum samples (Table 4.14) had copper levels below the CL of 0.65 µg/ml (McDowell *et al.*, 1993). This correlates with the forage analysis, which revealed that 93 % of samples had copper concentrations below the critical levels for both calves' and cows' needs. It can therefore be concluded that soil copper deficiency influenced forage and serum copper concentrations.

Only 4 % of the cows' serum sampled had zinc levels below the CL of 0.7 µg/ml indicating no major deficiency problems (McDowell *et al.*, 1993). Forage analysis (Table 4.8) on the other hand revealed that 39 % of samples were below the critical level of 18 mg/kg for both calves' and cows' needs (NRC, 2001). The majority of soil samples (96 %) had zinc concentrations above the deficiency critical level of 2 mg/kg DM (McDowell *et al.*, 1983). These values are similar to those from other findings, which have reported zinc deficiency in tropical Sudanese sheep (Mahmoud *et al.*, 1983) and in Southern America (McDowell *et al.*, 1984, 1993). Zinc deficiency is usually manifested in depressed diet intake, retarded growth, reproductive disorders, loss of hair and wool (Underwood, 1981). It is therefore interesting to note that while major deficiencies were

reported in forages, only 4 % of serum samples from cows were deficient. This indicates no obvious relationship between pasture concentration and zinc response conditions or on onset of clinical abnormalities. A possible explanation for higher serum zinc and lower forage zinc levels could be due to poor correlation between clinical deficiency and dietary zinc intakes in which the zinc released by tissue catabolism (associated with insufficient intakes of energy or protein) in animals is available to supplement dietary zinc (Masters, 1984). It can also be stated that the 39 % of samples deficient in zinc, could be due to the corresponding 4 % of the soils (Table 4.4), which revealed zinc deficiency. It can therefore be inferred that soil zinc influenced forage and serum concentrations.

4.3 FACTORS THAT INFLUENCE MINERAL CONCENTRATIONS IN FORAGE AND SERUM

Studies on soil and forage mineral status have shown their contributions towards mineral status in gazing livestock (Plant *et al.*, 1988). It is possible to estimate mineral deficiencies in animal from the average elemental concentrations of soils and plants although other factors have been found to influence their levels. The objective of this section is to discuss the role of soil pH, forage species, age and breed of animals on mineral deficiencies detected in soil, forage and animal serum.

4.3.1 Soil minerals and soil pH

The mean soil pH was found to be 5.8 with a range of 5.1-7.2 (Table 4.15). All the pH values were above 5.0, the range that enables maximum absorption of ionic species by plants to occur (Reid and Horvarth, 1980). Most mineral elements exist in soil in varying concentrations depending on the pH of that soil (Oduor, 2002). All the soils (Appendix V, VI) were assessed for the mineral concentrations at various pH levels (Table 4.15). The results show that the pH values of soils were fairly acidic ranging from 5.1-7.2. The soils revealed deficiencies in only Cu and Zn over this pH range. Alkaline metals are usually soluble in acidic medium, a condition that increases their availability (Cox, 1973; Pastrana *et al.*, 1991a). High levels of Ca, Fe and Al usually influence availability of Zn, Cu and P through Ca-Zn, Fe-Cu and $AlPO_4$ interactions (Sachez, 1981).

Table 4.15 The mean mineral concentrations and ranges (mg/kg), and pH of soil in relation to the recommended levels

Parameter	Mean ±SD (n=28)	Range	% Samples deficient	% Samples deficient
pH	5.80±0.44	5.10-7.20	>5.00	-
Na	101.00±32.00	58.13-155.00	55.00	0
K	709.00±24.40	189.10-1135.00	60.00	0
Ca	720.00±33.30	280.00-1840.00	71.00[a]	0
Mg	330.00±11.90	161.30-627.70	30.00	0
Fe	551.00±22.80	228.30-1198.30	30.00[b]	0
Mn	630.00±34.50	239.70-1583.50	10.00[c]	0
Cu	3.30±0.90	1.60-5.13	2.00[d]	14
Zn	6.70±0.40	1.97-22.06	2.00[d]	4

CL Source: Rhue and Kidder, 1983; [a]Breland, 1976; [b]Bahia, 1978; [c]Mtimuni, 1982; [d]McDowell et al., 1983)

Such mineral interactions reduce availability of Zn and Cu in plants thus influencing plants' nutrition.

4.3.2 Soil-forage mineral concentrations relationships

A soil-plant study on the mineral interrelationships has revealed a positive trend in most minerals (Jumba, 1989). A summary of the relationship between soil minerals and forage minerals is presented using SPSS program in which correlation analysis of their mean values (Table 4.16) was done. Correlation coefficients (r) between soil and forage

element concentrations were mainly positive with their differences being significant ($P<0.05$). This shows that an increase in soil mineral concentration leads to an increase in plants' concentration.

Table 4.16 Correlation analyses between mean mineral concentrations of soils and forage

Parameter	Soil (mg/kg) (n=28)	Forage (mg/kg) (n=28)	Correlation (r)
Na	101.00±32.00	1000±33.00	0.094
K	709.00±24.40	11800±50.00	0.201
Ca	720.00±33.30	570.00±19.00	-0.002
Mg	330.00±11.90	1350.00±71.00	0.005
Fe	551.00±22.80	56.00±0.53	0.205
Mn	630.00±34.50	105.00±0.58	0.281
Cu	3.30±0.90	5.00±2.84	-0.257
Zn	6.70±0.40	19.00±8.67	0.078

Negative relationship between soil and herbage Ca and Cu implies that an increase of these minerals in soil leads to their decrease in plants. This may be due to the varying stages at which various forages were sampled since similar studies have revealed that plants in their advanced maturity stage contain less concentration of minerals than young forages (Minson, 1990). Such a relationship can help increase the element concentration in forages for example by increasing the mineral that may be deficient in soils. However, the small correlation coefficients revealed weak relationships.

4.3.3 Forage species

Different plant species contain varying mineral concentrations (Jumba *et al.*, 1995a, b; Oduor, 2002). All forage species sampled (Appendix II) were classified in terms of their botanical species and species mixtures. The mineral concentration of these species: Rhodes (*Chloris gayana*), Kikuyu grass (*Pennisetum Clandestinum*), and mixtures; mixed natural grasses and Rhodes/Kikuyu grasses were compared (Table 4.17a, b). The results show that Kikuyu grass was superior in most minerals with the differences being significant ($P<0.05$) for P, Fe, Mn and Zn. Kikuyu grass was however inferior in Na, K, Ca, Fe, Cu and Zn minerals ($P<0.05$) when it was compared to Kikuyu/Rhodes grass mixtures.

Table 4.17a The mean macro element concentrations (g/kg DM) of forage species

Element	**Concentrations (g/kg DM)**							
	Pasture species				Supplement species			
	Rhodes	Kikuyu	Mixed Natural	Rhodes/ Kikuyu	Maize	Oats	Wheat	Silage
Na	1.03	1.01	0.93	1.25	0.83	0.80	0.81	1.90
K	14.93	5.29	21.65	13.07	10.58	14.93	13.07	5.60
Ca	0.61	0.49	0.66	0.54	0.42	0.78	0.47	0.56
Mg	1.51	1.93	1.32	1.64	1.13	0.93	1.15	0.10
P	7.29	9.38	5.87	7.93	4.50	4.04	4.50	3.20

The Rhodes grass had a better Na content than Kikuyu grass, which agrees with findings of western Kenya (Oduor, 2002). In terms of mineral deficiencies, most samples had low concentrations of magnesium with 80 % of samples species having concentrations below the minimum dietary allowance of 2.0 g/kg DM for cattle (TCORN, 1991). Kikuyu and

Rhodes grass mixtures were rich in all minerals except copper. Analysis of mineral concentration of plant species (oats, wheat, maize and silage) fed to cattle as mineral supplements during the dry season revealed some variations. These supplements were potential source of most mineral elements except magnesium, copper and zinc. This is due to the fact that botanical effects of a plant may affect its mineral concentrations (McDowell *et al.*, 1993).

Table 4.17b The mean trace element concentrations (mg/kg DM) of forage species

Element	**Concentrations (mg/kg DM)**							
	Pasture species				Supplement species			
	Rhodes	Kikuyu	Mixed Natural	Rhodes/ Kikuyu	Maize	Oats	Wheat	Silage
Fe	43.84	51.75	72.42	164.80	47.08	19.67	25.75	66.00
Mn	101.30	129.10	179.40	109.60	58.00	110.30	61.42	56.00
Cu	6.06	6.50	5.67	9.42	3.00	3.83	3.30	2.50
Zn	21.64	23.64	21.15	27.00	14.93	18.59	12.75	9.05

Differences between species in terms of mineral availability may be associated with physiological traits because minerals are unevenly distributed within the plant. The species also differ in their rates of maturity and hence their leaf: stem mineral level ratios (Minson, 1990).

4.3.4 Age of the animal

Mineral concentrations in animals vary depending on the stage of growth of the animal (Oduor, 2002). All the blood samples independent of the breeds were classified

depending on whether the animals were young growing calves (2-14 months old) or mature lactating cows (2-14 years old). The results (Table 4.18) revealed that lactating cows had higher levels of K, Ca, Fe and Zn while young growing calves were superior in Na and Mn although the differences in concentrations between calves and cows were not significant ($P<0.05$). On the other hand, there were no significant differences for Mg and Cu between calves and cows.

Table 4.18 Means of macro (g/l) and trace elements (μg/ml) concentrations in serum of young calves and lactating cows

Element	Young calves±SD	Lactating cows±SD
Na	2.45±0.42	2.38±0.61
K	0.37±0.08	0.39±0.06
Ca	0.68±0.21	0.81±0.32
Mg	0.02±0.01	0.02±0.01
Fe	2.28±1.13	2.43±1.53
Mn	0.27±0.15	0.26±0.14
Cu	0.60±0.19	0.60±0.17
Zn	3.11±1.23	3.71±1.63

The differences could be partly due to the type of feed given to the animal. Lactating cows depend wholly on forages, which may have high mineral element levels while calves depended mostly on available mother's milk and little of forages (McDowell and Conrad, 1989).

4.3.5 Cattle breeds

The effect of animal breed differences on mineral requirements has been observed in ruminants (Miller, 1979). All the blood serum from young growing calves and lactating

cows were categorized on the basis of the breed of the grazing animals (Table 4.19). The breeds sampled were Friesian, Ayshire and Friesian/Ayshire crossbreeds (Appendix III, IV).

Table 4.19 Means of macro element (g/l) and trace element (µg/ml) concentrations in serum of various cattle breeds

Parameter	**Growing calves**			**Lactating cows**		
	Friesian	Ayrshire	Friesian/ Ayrshire	Friesian	Ayrshire	Friesian/ Ayrshire
Na	2.52±0.28	2.35±0.26	2.00±0.82	2.35±0.45	2.58±0.78	2.13±0.59
K	0.39±0.05	0.38±0.12	0.33±0.06	0.41±0.06	0.38±0.07	0.37±0.02
Ca	0.69±0.13	0.81±0.26	0.48±0.15	0.70±0.10	0.99±0.50	0.77±0.12
Mg	0.02±0.01	0.02±0.01	0.04±0.03	0.03±0.01	0.02±0.01	0.02±0.01
P	0.20±0.13	0.17±0.05	0.13±0.05	0.15±0.05	0.16±0.05	0.15±0.05
Fe	1.73±0.00	3.00±2.71	2.44±1.45	2.90±1.12	2.20±1.97	2.14±1.55
Mn	0.26±0.16	0.22±0.04	0.34±0.19	0.24±0.12	0.30±0.17	0.26±0.13
Cu	0.54±0.17	0.58±0.16	0.72±0.23	0.59±0.20	0.66±0.12	0.52±0.17
Mo	0.25±0.31	0.26±0.20	0.49±0.11	0.34±0.35	0.37±0.40	0.36±0.19
Zn	2.89±1.11	4.15±0.54	2.44±1.42	4.59±0.99	3.24±1.03	2.78±0.79

The results revealed that Friesian calves were superior in Na, K, and P while Friesian/ayrshire calves had higher levels of Mg and Mn although the differences in concentrations between the breeds were significant ($P<0.05$). The result reveals no significant differences in the concentrations of minerals in mature breeds whose concentrations were almost the same. However, all the species had low levels of trace elements especially manganese whose levels were below the recommended 1µg/ml (McDowell *et al.*, 1993). The variations between the breeds in mineral levels could be due to the efficiency of absorption of minerals from the diet. Studies have shown that 3-

35 % of Mg in dairy cows, 40-80 % of P and 2-10 % of Cu in adult sheep are absorbed from the diet (McDowell and Conrad, 1989; Field, 1984).

CHAPTER FIVE

CONCLUSIONS AND RECOMMENDATIONS

5.1 CONCLUSIONS

Soil analysis revealed deficiencies in copper and zinc minerals while the forages were deficient in all minerals except manganese. However, serum analysis revealed deficiencies in magnesium, iron, manganese and copper in both calves and lactating cows. Deficiencies depicted in serum analysis could partly be due to inadequate dietary supply since the proportion of herbage that could not meet required levels for lactating cows was 89 % for magnesium and 93 % for copper.

The study revealed the soil pH and mineral interrelationships within samples, as factors influencing mineral concentrations in the study area. The soil pH range of 5.1-7.2 was favourable for most minerals except copper and zinc. Forage species analysis revealed that mixed grasses of Rhodes/Kikuyu had generally higher mineral concentrations while Friesian breeds were superior in most mineral levels. However, Cu: Mo levels in blood samples were approximately 2:1 implying no incidences of molybdenum poisoning through Cu-Mo interactions. However, correlation analysis of soil and forage mineral concentrations revealed mostly positive relationships which were not significant.

5.2 RECOMMENDATIONS

5.2.1 Direct mineral supplementation to grazing cattle

Results in this study have revealed deficiencies in magnesium, iron, manganese and copper in cattle serum in Uasin Gishu district. These deficiencies require possible supplementation trials for confirmation before long-term remedies can be set. The supplementation can be done by direct administration of the deficient minerals through either water, mineral licks or drenches of injections. The quantities of minerals depend on the stage of development of the particular animal, the average mineral status of the forages in the area and the mineral ingredient labels by particular feed manufacturers. A survey of mineral levels in feed concentrates has revealed that there are serious deviations from the information supplied on ingredient labels by some manufacturers in Kenya (Wasike, 1998). Supplementation has revealed that female reproductive performance may be improved by availability of specific trace elements. In cattle for example, Mn supplementation resulted in increased calving rates.

5.2.2 Indirect mineral supplementation to grazing cattle

Indirect mineral supplementation to grazing cattle involves use of inorganic fertilizers and altering soil pH in the region. Applying inorganic fertilizers to soils can help provide minerals to grazing livestock only if such fertilizer contains the mineral that is deficient in both soils and forages. Soil pH range of 5.1-7.2 was found sub-optimal for the availability of copper and zinc in the soils. Application of fertilizers containing Cu and Zn can increase the availability of such minerals by reducing the interactive effects of Ca

and Fe. This could influence mineral uptake by forage thereby reducing their deficiencies in plants (McDowell *et al.*, 1993).

5.2.3 Encouraging specific pasture species

This study has shown Rhodes/Kikuyu grass mixtures were superior in most bio-essential elements covered. It is noted that mixture of individual species may yield even better results than when they are individually available for grazing cattle.

5.2.4 Encouraging certain animal breeds

This study has revealed that Friesian breeds were superior in most mineral concentrations. It is suggested that replacement of indigenous breeds with Friesian breeds would lead to increased production.

5.3 Further work

There is need to investigate the effect of seasonal changes on mineral concentrations and determine the other factors contributing to lower levels of manganese and iron in calves and lactating cows in the district.

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APPENDICES

APPENDIX I
GPS IDENTIFICATION OF STUDY SITES

Farm ID No.	Farm Name		Location	Altitude (M)
1	VIL-ELDORET	SOY	00^0 31. 664N 035^0 16. 800E	2176
2	KIPKABUS	AINABKOI	00^0 17.113N 035^0 29.776E	2129
3	ELSO	KAPSARET	00^0 29.061N 035^0 24.503E	2501
4	MOIBEN	MOIBEN	00^0 51.563N 035^0 24.503E	2150
5a	BESERO	MOIBEN	00^0 34.723N 035^0 20.126E	2016
5b	BESERO	MOIBEN	00^0 34.671N 035^0 20.175E	2212
6	CHEMOSET	SOY	00^0 40.384N 035^0 10.638E	2174

APPENDIX II
SOIL/GRASS SPECIES LOCATION DESCRIPTION

Farm ID	Soil sample No.	Forage No./species
1	1	1 Oats (O)
	2	2 Maize (M)
	3	3 Maize (M)
	4	4 Mixed Natural Grass (N/G)
2	5	5 Oats seeds (O)
	6	6 Oat straws (O)
	7	7 Silage (Sil.)
	8	8 Rhodes/Kikuyu (R/K)
3	9	9 Rhodes (R)
	10	10 Kikuyu (K)
	11	11 Rhodes (R)
	12	12 Mixed Natural Grass (N/G)
4	13	13 Wheat straws
	14	14 Rhodes (R)
	15	15 Rhodes/Kikuyu (R/K)
	16	16 Rhodes (R)
5a	17	17 Maize straws (M)
	18	18 Kikuyu (K)
	19	19 Rhodes (R)
	20	20 Rhodes/Kikuyu (R/K)
5b	21	21 Rhodes re-growth (R)
	22	22 Rhodes (R)
	23	23 Kikuyu (K)
	24	24 Mixed Natural Grass
6	25	25 Wheat hay (W)
	26	26 Rhodes (R)
	27	27 Wheat straws (W)
	28	28 Rhodes/Hay (R)

APPENDIX III
SERUM SAMPLES FOR YOUNG GROWING CALVES

Farm ID	Serum sample No./Breed
1	1. Friesian Calf (F.C) 2. Friesian Calf (F.C) 3. Ayrshire Calf (A.C)
2	4. Friesian Calf (F.C) 6. Friesian Calf (F.C) 6.Friesian Calf (F.C)
3	7.Ayrshire Calf (A.C) 8. Friesian Calf (F.C) 9. Friesian Calf (F.C)
4	10. Ayrshire Calf (A.C) 11. Friesian/ Ayshire calf (F.A.C)
5a	12. Friesian/ Ayshire calf (F.A.C) 13.Ayrshire Calf (A.C)
5b	14. Friesian Calf (F.C) 15.Friesian/ Ayshire calf (F.A.C)
6	16. Ayrshire Calf (A.C) 17. Friesian/ Ayshire calf (F.A.C) 18. Friesian/ Ayshire calf (F.A.C)

APPENDIX IV

SERUM SAMPLES FOR LACTATING COWS

Farm ID	Serum sample No./Breed
1	1. Friesian (F) 2.Friesian (F) 3.Ayshire (A)
2	4. Friesian (F) 5.Friesian (F) 6. Friesian (F)
3	7.Ayrshire (A) 8. Friesian (F) 9. Friesian (F)
4	10. Ayshire (A) 11. Friesian (F) 12. Friesian (F) 13.Friesian/ Ayshire (F/A)
5a	14.Friesian/ Ayshire (F/A) 15. Ayrshire (A) 16. Ayrshire (A) 17.Ayrshire (A)
5b	18. Ayrshire (A) 19. Friesian (F) 20.Friesian (F) 21.Friesian/ Ayshire (F/A)
6	22. Ayrshire (A.) 23. Friesian/ Ayshire (F/A) 24. Friesian/ Ayshire (F.A.)

APPENDIX V
)IL MACRO ELEMENT CONCENTRATION (mg/kg DM) and SOIL pH

ıple No.	Na	K	Ca	Mg	pH
	96.89	567.3	440	182.3	5.1
	77.51	945.5	280	200.8	5.2
	58.13	1134	600	417.7	5.8
	58.13	567.3	760	362.9	5.9
	96.89	756.4	520	347.9	5.8
	155.0	378.2	520	210.6	6.3
	155.0	756.4	1260	558.8	6.1
	135.7	567.4	760	407.3	5.9
	135.7	756.47	760	396.4	5.8
	58.13	189.1	880	453.3	6.1
	116.3	756.4	480	303.4	5.1
	116.3	567.3	280	194.9	5.1
	135.7	756.7	760	461.9	6.1
	77.51	945.5	1240	262.9	6.0
	58.13	945.5	520	276.3	5.6
	135.7	378.2	520	161.3	5.9
	96.89	378.2	480	187.2	5.7
	77.51	567.3	680	185.1	5.8
	96.89	1134	720	409.0	5.8
	77.51	756.4	720	286.5	5.9
	77.51	945.5	440	254.3	5.6
	96.89	567.3	320	214.6	5.1
	77.51	1135	1840	398.9	7.2
	96.89	378.2	1000	627.7	6.3
	58.13	756.4	760	363.7	5.9
	116.3	756.4	840	396.2	6.0
	135.7	756.4	960	363.9	5.9
	155.0	756.4	840	376.5	6.1

APPENDIX VI
SOIL TRACE ELEMENT CONCENTRATION (mg/kg DM)

Sample No	Fe	Mn	Cu	Zn
1	636.6	505.1	2.966	3.108
2	428.8	607.7	2.789	2.704
3	537.5	669.2	3.297	8.888
4	779.1	1583.5	4.301	22.063
5	328.6	564.8	2.363	4.604
6	228.3	256.1	5.128	2.300
7	707.8	915.1	1.926	6.717
8	385.3	364.9	4.538	3.113
9	1061.8	1011.6	3.557	21.271
10	437.3	440.1	4.041	4.658
11	471.1	431.3	2.541	3.813
12	541.2	571.5	3.013	2.233
13	668.0	445.8	4.526	5.113
14	1198.3	1305.7	4.313	18.583
15	521.8	644.0	2.871	4.342
16	523.0	593.1	3.190	5.388
17	300.8	239.7	1.595	1.971
18	515.8	425.5	2.623	4.438
19	860.0	1200.9	1.843	12.8
20	625.7	1022.2	3.049	6.650
21	532.7	855.4	2.800	3.571
22	504.9	629.4	3.025	4.733
23	688.5	803.7	4.951	13.20
24	657.1	257.4	1.855	12.30
25	310.4	330.8	3.639	2.996
26	405.9	421.5	4.171	2.329
27	332.2	289.3	3.273	2.967
28	251.3	263.6	4.278	2.121

APPENDIX VII
FORAGE MACRO ELEMENT COMPOSITION (g/kg DM)

Sample No	Na	K	Ca	Mg	P
1	0.545	18.67	0.608	0.530	2.516
2	0.860	16.80	0.355	1.885	5.489
3	0.820	6.53	0.406	0.790	4.803
4	0.800	7.47	0.710	1.348	5.489
5	0.913	14.00	0.558	0.708	6.404
6	0.950	12.13	1.166	1.550	3.202
7	1.900	5.60	0.558	0.100	3.202
8	2.118	10.27	0.608	2.368	4.117
9	0.775	15.87	0.659	1.668	7.548
10	1.130	3.73	0.507	2.850	8.920
11	0.958	20.53	0.811	0.763	2.973
12	0.983	5.60	0.710	1.445	5.261
13	0.900	12.13	0.558	1.623	2.745
14	1.378	11.20	0.558	3.133	4.575
15	0.738	11.20	0.558	0.798	9.149
16	1.148	19.60	0.659	0.420	5.489
17	0.800	8.40	0.507	0.713	3.202
18	0.708	7.47	0.811	0.920	5.489
19	1.063	10.27	0.811	1.183	5.947
20	0.908	17.73	0.456	1.768	10.522
21	1.028	11.20	0.558	1.160	8.234
22	1.085	14.93	0.304	1.645	10.004
23	1.195	4.67	0.152	2.010	13.724
24	1.000	7.47	0.558	1.160	6.862
25	0.875	19.60	0.507	0.455	1.830
26	0.880	14.00	0.558	1.828	6.404
27	0.668	7.47	0.355	1.360	8.920
28	0.913	16.80	0.558	1.745	14.410

APPENDIX VIII

FORAGE TRACE ELEMENT COMPOSITION (mg/kg DM)

Sample No	Fe	Mn	Cu	Zn
1	34.00	124	4.25	18.00
2	35.25	47.75	2.50	13.33
3	20.00	46.25	2.50	9.88
4	79.50	178.00	5.25	19.38
5	10.25	63.25	4.00	25.45
6	14.75	143.50	3.25	12.33
7	66.00	56.00	2.50	9.05
8	231.25	95.75	15.50	39.78
9	70.75	114.25	4.50	21.88
10	87.75	246.5	9.75	32.53
11	12.50	138.25	2.75	9.88
12	43.00	193.00	5.25	22.08
13	21.25	64.25	2.25	11.73
14	54.00	38.25	6.50	11.60
15	218.75	195.25	6.00	18.95
16	28.25	127.00	5.50	29.75
17	860.00	80.00	4.00	21.58
18	19.00	56.75	3.00	13.45
19	30.75	131.15	4.25	11.50
20	44.25	37.75	6.75	22.28
21	35.75	196.25	4.25	22.28
22	52.25	63.25	7.00	37.05
23	48.50	84.00	6.75	24.93
24	94.75	167.25	6.50	21.98
25	15.25	63.00	2.00	8.30
26	29.75	72.50	8.25	17.40
27	40.75	57.00	5.75	18.25
28	49.25	67.50	8.25	21.63

APPENDIX IX
SERUM MACRO ELEMENT COMPOSITION (g/l) IN YOUNG GROWING CALVES

Sample No.	Na	K	Ca	Mg	P
1	2.622	0.337	0.820	0.038	0.087
2	2.289	0.299	0.800	0.033	0.146
3	2.301	0.576	0.560	0.028	0.117
4	2.184	0.401	0.640	0.028	0.087
5	2.501	0.392	0.820	0.008	0.223
6	3.105	0.441	0.760	0.008	0.174
7	2.596	0.404	0.950	0.014	0.223
8	2.639	0.453	0.600	0.001	0.515
9	2.484	0.387	0.560	0.009	0.243
10	2.379	0.365	0.510	0.012	0.175
11	2.002	0.345	0.290	0.085	0.136
12	3.648	0.402	0.670	0.024	0.204
13	1.912	0.251	1.140	0.016	0.223
14	2.373	0.400	0.480	0.025	0.126
15	2.493	0.505	0.760	0.014	0.126
16	2.537	0.314	0.890	0.013	0.126
17	1.939	0.309	0.450	0.008	0.068
18	2.056	0.247	0.490	0.032	0.097

APPENDIX X
SERUM MACRO ELEMENT COMPOSITION (g/l) IN LACTATING COWS

Sample No.	Na	K	Ca	Mg	P
1	2.294	0.578	0.650	0.035	0.068
2	2.346	0.361	0.790	0.035	0.097
3	2.507	0.321	1.070	0.044	0.107
4	2.274	0.472	0.830	0.003	0.175
5	2.372	0.463	0.800	0.041	0.117
6	2.669	0.398	0.700	0.032	0.087
7	3.190	0.332	0.770	0.020	0.165
8	2.768	0.349	0.490	0.034	0.223
9	1.685	0.395	0.740	0.028	0.105
10	1.615	0.413	0.800	0.020	0.223
11	1.620	0.400	0.730	0.017	0.194
12	2.576	0.420	0.630	0.039	0.165
13	1.131	0.364	0.710	0.025	0.117
14	2.278	0.365	0.840	0.020	0.185
15	1.891	0.395	0.520	0.025	0.117
16	2.870	0.515	2.160	0.019	0.185
17	2.041	0.275	0.840	0.012	0.087
18	4.041	0.412	0.920	0.021	0.185
19	3.173	0.384	0.550	0.017	0.223
20	2.021	0.329	0.770	0.023	0.175
21	2.705	0.413	0.700	0.013	0.185
22	2.453	0.369	0.780	0.011	0.185
23	2.405	0.363	0.660	0.014	0.175
24	2.123	0.357	0.950	0.033	0.068

APPENDIX XI
SERUM TRACE ELEMENT COMPOSITION (µg/ml) IN YOUNG GROWING CALVES

Sample No.	Fe	Mn	Cu	Mo	Zn
1	0.80	0.50	0.60	0.07	4.40
2	1.50	0.10	0.70	0.07	2.24
3	1.60	0.20	0.40	0.07	3.57
4	1.80	0.30	0.50	0.44	3.13
5	3.60	0.20	0.40	0.29	3.75
6	2.80	0.20	0.30	0.07	2.61
7	7.30	0.30	0.70	0.07	5.00
8	2.00	0.20	0.80	0.07	3.77
9	0.80	0.10	0.60	0.07	2.27
10	3.80	0.20	0.50	0.22	4.01
11	3.30	0.50	0.70	0.07	1.29
12	0.20	0.20	0.90	0.07	2.19
13	2.00	0.20	0.80	0.44	4.26
14	0.50	0.50	0.40	0.95	0.93
15	3.90	0.60	0.40	1.09	1.09
16	0.30	0.20	0.50	0.51	3.90
17	1.90	0.20	1.00	0.07	4.57
18	2.90	0.20	0.60	1.16	3.04

APPENDIX XII
SERUM TRACE ELEMENT COMPOSITION (µg/ml) IN LACTATING COWS

Sample No.	Fe	Mn	Cu	Mo	Zn
1	3.00	0.20	0.40	0.07	5.82
2	2.90	0.20	0.40	0.07	4.75
3	0.40	0.40	0.80	0.36	3.49
4	4.20	0.10	1.00	0.07	5.69
5	3.50	0.20	0.70	0.07	2.83
6	2.60	0.20	0.60	0.07	4.16
7	3.00	0.20	0.70	0.15	4.68
8	0.80	0.20	0.40	0.15	5.45
9	4.00	0.40	0.40	0.80	4.15
10	6.10	0.20	0.60	0.44	3.65
11	3.70	0.50	0.80	0.87	5.34
12	1.00	0.20	0.60	0.51	3.45
13	2.20	0.20	0.60	0.07	4.58
14	0.80	0.40	0.60	0.07	6.84
15	0.70	0.70	0.70	0.15	3.64
16	0.80	0.20	0.50	0.07	2.91
17	2.40	0.20	0.70	0.15	3.41
18	0.70	0.30	0.80	0.36	3.09
19	3.10	0.20	0.60	0.44	3.59
20	1.20	0.20	0.60	0.80	4.12
21	2.30	0.10	0.20	0.95	0.65
22	3.50	0.20	0.50	1.31	1.04
23	0.80	0.20	0.60	0.15	1.00
24	4.60	0.40	0.60	0.58	0.82