MINERAL STATUS OF SOILS AND FORAGES IN LAKE NAKURU NATIONAL PARK AND IMPLICATIONS TO ANIMAL HEALTH

BY

SILVESTER SIVA KISIA

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DECLARATION

I declare that this thesis is of my own composition and, apart from the acknowledged assistance, is a record of my own research. The material has never been presented before the University of Nairobi or any other institution for an academic award.

Silvester K. Siva_____

Department of Chemistry

University of Nairobi

This Thesis has been Submitted with Knowledge of:

Dr. I.O. Jumba_____

Department of Chemistry

University of Nairobi

Dr. S. Kisia_____

Department of Veterinary Medicine

University of Nairobi

DEDICATION

This thesis is dedicated to Clement, Jerida and Damaris.

ABSTRACT

A study was conducted in which forty-four (44) soil and sixty-seven (67) forage samples were collected at different sites within the Lake Nakuru National Park. Sampling sites were classified in terms of distance from the lake and location (east, west or north) while forages were categorized according to species most favoured for grazing by wildlife. Soils were analyzed for extractable sodium (Na), potassium (K), calcium (Ca), magnesium (Mg), phosphorus (P), iron (Fe), manganese (Mn), copper (Cu), cobalt (Co), zinc (Zn), cadmium (Cd) and lead (Pb); while forages were assayed for the same elements plus aluminium (Al) and molybdenum (Mo) as total concentrations on a dry matter(DM) basis. The objectives of this work were to assess the status of both macro- and trace elements in soils and forage and identify those that might be limiting animal health and nutrition within the Park.

Soil analysis indicated that pH, extractable Ca, Mg, Co, Cu, Mn and Zn were significantly higher ($P<0.05$) in samples from the east than from the north and west of the Park. Potassium and Na decreased significantly ($P<0.05$) with distance (away) from the lake; whereas the converse was true of Mn and Fe. Differences in soil concentrations for the elements P, Cd, Pb and Fe were insignificant irrespective of sampling region or distance from the lake.

Forage analysis revealed wide variations in elemental contents. Macromineral concentrations (g/kg) were Na(mean: 1.73; median: 0.855), K(mean: 8.11; median: 7.00), Ca(mean: 4.02; median: 2.89), Mg(mean: 0.941; median: 0.860) and P(mean: 1.80; median: 1.69). Levels of trace elements (mg/kg) were Al(mean: 489; median: 431), Fe(mean: 529; median: 424), Mn(mean: 89; median: 78), Cu(mean: 9.92; median: 8.21), Co(mean: 0.649; median: 0.624), Zn(mean: 18.1; median: 15.4), Mo(mean: 5.85; median: 3.71), Cd(mean: 0.191; median: 0.181) and Pb(mean: 0.721; median: 0.644).

Comparison with nutrient standards set by various authorities such as the National Research Council suggested that most of the forages were generally deficient in terms of requirements for grazing ruminants. The percentage of forages with deficient levels were 68(K), 58(Na), 52(Ca), 87(P), 95(Mg), 94(Zn) and 63(Cu). No significant differences ($P<0.05$) were noted between forage mineral concentrations and distance from the lake or location. However, analysis of species effects showed that the concentration of only one mineral, K was significantly higher ($P<0.05$) in *Digitaria swazilandensis* than in the other major species (*Cynodon dactylon, Chloris gayana* and *Sporobolus spicatus*). This difference was attributed to the influence of high soil composition, especially on the eastern side of the Park. Strategies and priorities have been suggested for further investigations concentrating on blood and tissue analysis (e.g. bone, liver and kidney) and response to supplementation in some of the animal species such as the waterbuck. In this way any constraints on animal health will be identified and alleviated.

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CONTENTS

LIST OF TABLES

LIST OF FIGURES

CHAPTER 1

INTRODUCTION

Nutritional disorders account for a major segment of the factors affecting animal health and productivity in almost all regions of the world (McDowell *et al.*, 1993). Problems of low fertility, bone abnormalities, wasting diseases such as nutritional muscular dystrophy, retarded growth and maturity, hair disorders, pica, sudden death, low meat and milk production, general weakness and predisposition for the occurrence of bacteriological, viral and parasitic diseases (Faria *et al.*, 1981) have been recorded among grazing animals with a high degree of prevalence especially in tropical regions. These problems can be attributed to an imbalance or insufficient availability of one or a combination of several nutritional factors such as protein, fibre, energy (carbohydrates and fats), vitamins and minerals. For example, evidence from animal trials indicates that mineral supplementation can reduce mortality significantly, increase the calving percentage by 20 - 100%, and promote growth rates by 10 - 25% (McDowell and Conrad, 1977). These results emphasize the fact that this class of nutrients accounts for one of the crucial factors in animal diets. A specific example is provided by McDowell *et al.*(1985) who reported increased milk yields by 7 - 260% and meat production by 6 - 400% following sulphur supplementation of cattle diets.

For all elements, the surest diagnostic method of determining a mineral deficiency, toxicity or imbalance would be to monitor growth and health responses following administration of a specific supplement. This is especially important in the tropics where many types of soils occur originating from a variety of bedrocks.

These soils have varying chemical compositions and are deficient in a number of mineral elements, including sulphur (Whanger, 1972; Paladines, 1984) and trace elements (Jumba *et al.*,

1995b). And since grazing ruminants in these areas do not usually receive mineral supplements except for common salt, they must depend almost exclusively on forage to meet their nutritional mineral requirements. Forages on the other hand do rarely satisfy all mineral needs of the grazing animal, making deficiencies a prevalent problem in several tropical regions. For example, many investigators have reported sub-optimal levels of copper and zinc in forages (McDowell *et al.*, 1989; McDowell, 1992; Rojas *et al.*,1993; Jumba *et al.*, 1996a) whereas iron and manganese concentrations have usually been found to be adequate in terms of animal requirements (Jumba *et al.*, 1995b).

The assessment of the mineral status of grazing animals involves sampling of forage consumed by the animals and soil upon which the forages grow. A sample of greatest value from soil, forage and animal tissues depends on the mineral in question (McDowell, 1985). Since the soil-plant-animal interaction is a complex system which has not been adequately investigated especially in developing countries, it would be worthwhile obtaining greater information on interretionships among soils, plants and animals and then use the information to assess the nutrition problems and animal productivity. However, sampling of animals requires handling facilities and labour especially when more informative samples such as liver and bone biopsy specimens are required.

Furthermore, for wild animal species, the animal has to be sacrificed for the study and this can threaten the population of the particular species under national protection. These drawbacks point to the fact that the first step in mineral studies would be to assess soil and forage and by implication determine the elements that are likely to be limiting animal health and productivity. This would then be followed by specific supplementation trials. Such an approach has the following additional advantages over direct mineral supplementation trials:

1) It limits supplementation schemes to a few specific minerals.

2) It eliminates the problems associated with direct mineral supplementation in areas where toxicity of certain elements could be caused by imbalanced intake (Suttle, 1986a).

3) It reduces time of obtaining specific evidence from animal trials for a range of minerals.

4) It eliminates uncertainties involved in supplementation due to inaccurate and unreliable information on mineral ingredient labels (McDowell *et al.*, 1993).

The Lake Nakuru National Park (LNNP) is one of the highest revenue generating parks in Kenya. Situated in the central Rift Valley, its proximity to Nakuru town offers a strategic advantage to tourists visiting the Western part of Kenya. However, there has been recent concern over the health status of wild herbivore and by implication the future suitability of the park for certain endangered animal species.

Maskall and Thornton(1989; 1991) reported low copper levels in the blood of the impala species and attributed the anomaly to poor forage composition and variations in the geochemistry of the area. In another study Kock *et al.* (1994) attributed the loss of body condition among the waterbucks (*Kobus ellipsipyrmnus defassa*) to the possible occurrence of hepatitis, helminthiasis and normocytic anaemia. However, recent but preliminary analysis of the liver and kidney has indicated high levels of cadmium and lead in the tissues but the source of these pollutants has not yet been established (R.A.Kock: Personal Communication).

The waterbuck at LNNP compete intra- and inter- specifically for available forage around the lake. They are restricted by a fence and territorial behaviour in their feeding range. They are also reported to feed on a variety of dryland forage, selection being dependant on relative crude fibre,

energy, protein and moisture content (Tomlinson, 1980). Recent observations however indicate health differences in waterbuck populations in different areas of the park. Animals residing in the east appear emaciated, weak and generally in poor health with loss of hair colour; while those residing in the western part of the park are in better health with little or no evidence of loss of body condition. This makes the park a good study site for investigations involving minerals and environmental pollution.

The objectives of the work presented in this thesis were therefore to (1) assess the elemental status of soils and forages from LNNP (2) identify those minerals that might be limiting animal health and nutrition in different locations within the park (3) investigate the factors contributing to the problem and (4) determine the possible sources of cadmium and lead to the animals.

CHAPTER 2

LITERATURE REVIEW

IMPORTANCE OF MINERALS IN BIOLOGICAL SYSTEMS AND DIAGNOSTIC METHODS OF MINERAL IMBALANCES IN GRAZING ANIMALS

2.1 CLASSIFICATION OF MINERAL ELEMENTS

Although protein and calorie availability is accepted as the most limiting factor to animal health in tropical areas, mineral deficiencies and imbalances in soils and forage have long been thought to be responsible for low production and reproduction among grazing ruminants in many developing countries (McDowell *et al.*, 1983).

Minerals can either be considered as essential (having some form of function in the body for example, Ca in the skeleton of vertebrates) or non essential (no known physiological function eg. Cd and Pb). A mineral is considered to be essential if its deficiency consistently results in impairment of a function from optimal to suboptimal (Mertz, 1970). It must also meet the following criteria: (i) should be present in all healthy tissues of living things; (ii) its concentration from one animal to the other must be fairly constant; (iii) its withdrawal from the body induces reproducibly the same physiological and structural abnormalities regardless of the species studied; (iv) its addition either reverses or prevents these abnormalities; (v) the abnormalities induced by deficiency are always accompanied by pertinent, specific biochemical changes and (vi) these biochemical changes can be prevented or cured when the deficiency is prevented or cured (Cortzias, 1967).

At least 15 mineral elements are nutritionally essential for ruminants. There are seven major (sometimes called macrominerals): Ca, P, K, Na, Cl, Mg and S; and eight trace elements (microminerals):- Fe, I, Zn Cu, Mn, Co, Mo and Se. By definition, macrominerals are those elements that are present in the body in large quantities and whose daily requirement exceeds 100mg while trace elements are those required in very small (μg/g) amounts for optimum body function (Cotzias, 1967; Mertz, 1974) but they are equally important in animal nutrition and each deficiency can lead to lowered productivity and clinical disease. In addition there are a number of newly discovered essential trace elements. These include Chromium, Vanadium, Nickel, Tin, Silicon and Arsenic and their characteristics have been reviewed by Underwood (1977) and McDowell *et al.* (1978). However, their practical significance for ruminants has not been demonstrated since evidence for essentiality has been based almost exclusively on growth effects in "laboratory" animals receiving highly purified diets in controlled growth environments.

Mineral deficiencies and excesses have been reported from most regions of the world, but the problem appears to be particularly severe in developing tropical countries because ruminants have to depend on local forage for almost all their requirements. McDowell (1976) and McDowell et el.(1984) showed that within the tropical regions of Africa, iodine deficiency had been reported in 20 countries; Co, Cu, Se and Zn deficiency in 34 countries and Fe and Mn deficiency in 7 countries.

Mineral deficiencies may also be confounding factors in animal diseases caused primarily by other factors. In the Eastern Plains of Columbia, for example, a wasting disease (secadera) caused by acute thiamine deficiency is complicated by Zn, Cu and less frequently Co and Se deficiencies. In that region, McDowell *et al.* (1985a) showed that 80-100% of soils and 70-100% of forage samples were

deficient in Cu, Zn, Co and Se. Excess Mo, with 26-72% of samples above the critical level, could complicate the secadera problem because it could produce a secondary Cu deficiency.

2.2 FUNCTIONS OF MINERALS IN BIOLOGICAL SYSTEMS

Minerals in general tend to: play a structural role in the skeleton, connective tissue and proteins; function as osmotic and ionic regulators in body fluids; and act as integral parts of enzymes.

2.2.1 MAJOR ELEMENTS

(a) Calcium and Phosphorus

Ninety-nine percent of Ca and 75% of P in the body are found in the skeleton and it is this tissue which reveals signs of deficiency. In bones calcium and phosphorus provide strength and density: a deficiency can lead to their fracture and deformation (Little, 1984, Read *et al.*, 1986). Both elements are present in combined form as the bone mineral, hydroxyapatite, in the ratio of 1:2.2 (P:Ca). Therefore typical bone ash contains about 24% Ca and 11.5% P.

In addition Ca is essential for normal blood clotting, rhythmic heart action, neuro-muscular excitability, enzyme activation and permeability of membranes. On the other hand P in soft tissue is essential for proper functioning of rumen micro-organisms (especially those which digest cellulose), utilization of energy from feeds, buffering of blood and other fluids, many enzyme systems and protein metabolism. Consequences of dietary P deficiency on the skeleton are generally the same as those of Ca deficiency and include rickets in the young growing animals, and osteomalacia in the old animals. Severe P deficiency can cause loss of appetite and depraved appetite, manifested in the chewing of bones of dead animals and other debris, a situation called 'pica'. Pica is a wasting disease which occurs worldwide in

cattle, sheep and other animals (Russel and Duncan, 1956, Buttler and Jones, 1973). Loss of appetite may provide some protection from P deficiency by reducing both maintenance and production needs (Suttle 1987a). Under conditions of extreme P shortage, however, animals may go for 2 or 3 years without producing a calf or even coming into oestrus (Phillips, 1956).

(b) Sulphur

Sulphur, like nitrogen, is essential for microbial synthesis because it contributes to microbial S-containing amino-acids. Failure to meet the microbial needs for S may depress the rate of digestion in the rumen (Bray and Hamsley, 1969; Kennedy and Siebert, 1973) with consequent depression of feed intake.

Blood lactate and dietary sulphur levels are considered to be the most reliable indicators of S status. Evidence presented by Paladines (1984) and McDowell *et al.* (1984) has shown that S deficiencies appear to be frequent in Savanna-type soils of the tropics. Sulphur is implicated in Se toxicity and also in Cu deficiency, where high levels of sulphide produced in the rumen generate high levels of insoluble copper sulphide, a form in which Cu is unavailable for absorption.

(c) Magnesium

Hypomagnesaemia and the associated tetany is a metabolic disorder encountered in both grazing adult ruminants and calves reared too long on milk without access to other foods. Magnesium is widely distributed among plant and animal tissues. In vertebrates, some 70% of total body Mg is present in bones. Typical bone ash contains an average 0.7% Mg. Magnesium has many diverse physiological functions. The Mg in the skeleton is important for the integrity of bones and teeth.

Magnesium plays a key role as an essential metal ion in many fundamental enzymatic reactions, in intermediary metabolism and also as an "activator" of enzymes such as kinases involved in the metabolism of carbohydrates and lipids. It is also involved in protein synthesis through its action on ribosomal aggregation; in binding messenger RNA to ribosomes; and in the synthesis and degradation of DNA. Magnesium also plays an important role in neuromuscular transmission as well as neuromuscular activity.For grazing ruminants, grass tetany occurs when analysis of blood or urine samples shows abnormally low levels of Mg (12-16 mg/kg). Likewise, a reduction in Mg content of cerebrospinal fluid (i.e. values < 1.6 mg/100ml) is an indication of Mg deficiency (Meyer, 1976; McDowell *et al.*, 1984). Grass tetany is usually prevented by Mg fertilization of pastures, adding Mg to feed or salt blocks, and avoiding the use of high K fertilizers.

(d) Potassium

Potassium is the third most abundant mineral in the animal body and is the principal cation of intracellular fluid. It is also a constituent of extracellular fluid where it influences muscle activity. Potassium is thus essential for life, being required for a variety of body functions including osmotic balance, acid-base equilibrium, several enzyme systems, and water balance. An ionic balance exists between K, Na, Ca, and Mg.

Potassium requirement appears to be increased for animals under stress. Excitement tends to increase the urinary loss of K. Fever and diseases such as diarrhoea also increase K loss. Potassium deficiency is not considered to be a major problem for grazing ruminants in tropical countries since young forages generally contain adequate levels of K. However, it is possible that K deficiency may occur during extended dry seasons as K decreases with increasing forage maturity. Deficiency of

potassium results in non-specific signs such as slow growth, reduced feed and water intake, lowered feed efficiency, muscular weakness, nervous disorders, stiffness, emaciation, intracellular acidosis, and degeneration of vital organs (McDowell *et al.*, 1993).

Detection of K deficiency is difficult. Low serum K values have some diagnostic value for establishing a deficiency but these may be caused by malnutrition, negative nitrogen balance,gastrointestinal losses, and endocrine malfunction. Reduced feed consumption is an early sign of inadequate dietary K. Because reliable data on evaluation of K deficiency based on tissue analyses are not available, dietary K concentration apparently is the best indicator of K status of grazing animals (McDowell *et al.*, 1984).

(e) Sodium and Chlorine

Sodium and Cl (in addition to K) are the elements responsible for maintaining osmotic pressure and regulating acid-base equilibrium. These two mineral elements (Na^+ and Cl^-) function as electrolytes in body fluids and are specifically involved at the cellular level in water metabolism, nutrient uptake and transmission of new impulses. Chloride is necessary for activation of amylase and is essential for formation of gastric hydrochloric acid (Underwood, 1977; Mills, 1983).

Sodium deficiency is very common amongst grazing animals in the tropics and can easily be overcome by provision of common salt licks. Natural forage is generally deficient in Na and in the tropics high Na losses occur in sweat. The initial sign of Na deficiency is a craving for salt, demonstrated by avid licking of wood, soil and sweat from other animals, and also drinking a lot of water. A prolonged deficiency causes loss of appetite, decreased growth, unthriftness, reduced milk production, and loss of weight. More pronounced signs of Na deficiency include shivering,

incoordination, weakness, and cardiac arrhythmia, which can lead to death. For most grazing animals sodium deficiency is most likely to occur (a) in lactating animals due to increased secretion of sodium in milk; (b) in animals that are rapidly growing such as calves; (c) during loss of water and sweating and (d) where animals are forced to graze pastures low in Na and heavily fertilized with K (McDowell *et al.*, 1993). A criterion for assessment of Na deficiency status is the low level of Na and K in saliva. Deficiency also causes a rise in K in blood. Khalili (1991) suggested that faecal Na concentration could be an accurate and practical method of detecting deficiency of the mineral.

2.2.2 TRACE ELEMENTS

Trace elements act primarily as catalysts in cellular enzyme systems and as integral parts of specific proteins. However, Si which is one of the newer essential trace elements, has been found to have a structural role in connective tissue and the organic matrix of the skeleton (Underwood, 1977; Mills, 1983).

(a) Cobalt

Rumen organisms require Co for the synthesis of vitamin B_{12} (Andrews, 1956). 4.5% of vitamin B_{12} is the cobalt complex referred to as cobalamin; hence for grazing animals, synthesis of vitamin B_{12} entirely depends on the content of Co in their diet. Vitamin B_{12} is an essential component of several enzyme systems which carry out a number of very basic metabolic functions.

most reactions of this vitamin involve transfer or synthesis of one-carbon units, for example methyl groups. Though the most important task of vitamin B_{12} concern metabolism of nucleic acids and proteins, it also functions in metabolism of fats and carbohydrates (Andrews, 1956).

Summarily vitamin B_{12} functions in metabolic processes including:- purine and pyrimidine synthesis; transfer of methyl groups in synthesis of methionine, formation of proteins from amino-acids and metabolism of fats and carbohydrates (McDowell *et al.*, 1993)

Cobalt deficiency is largely restricted to grazing ruminants that have little or no access to mineral concentrates. The clinical signs of Co deficiency include poor appetite, lack of vigour, muscle wasting and low body weight, and are usually difficult to distinguish from those of malnutrition related to low protein and low calorie diet. Sub-clinical deficiency, characterised by low production rates, is very common and causes considerable economic losses. However, Co-deficient ruminants respond quickly to Co supplementation.

b) Copper and Molybdenum

The roles of copper in the animal body include cellular respiration, bone formation, proper cardiac function, connective tissue development, myelination of the spinal cord, keratinization, and tissue pigmentation. Its physiological importance is mainly the functional role in the metalloenzymes which include ceruloplasmin, cytochrome oxidase, dopamine-_-hydroxylase, lysyl oxidase, superoxide dismutase, and tyrosinase (Underwood, 1981; McDowell *et al.*, 1993).

Decreased production of antibody producing cells is a characteristic of Cu deficiency since the level of the T and B cells, neutrophils and macrophages are normally affected.

Molybdenum on the other hand, has been identified as an integral part of six enzymes including:- Xanthine oxidase, aldehyde oxidase, and sulphite oxidase. These enzymes help in

metabolism of purines, pyrimidines, pteridines, aldehydes and in oxidation of sulphite. Electron transport chain in cells, involving cytochrome C (Rajagopalan, 1980) is carried out by Xanthine oxidase and aldehyde oxidase.

Copper is probably the second most deficient trace element for grazing ruminants in the tropics, although most cases are related to constituents such as Mo, S and other factors which produce a conditioned Cu deficiency even though normal amounts of Cu (6-16mg/kg DM) may occur in forage. In general, Cu deficiency usually occurs when forage Mo exceeds 3mg/kg DM and Cu is below 5 mg/kg DM. Ward (1978) recognised four categories of Cu deficiency in which feed contained high Mo (> 20mg/kg), low but relatively high Mo in a ratio of 2:1, very low Cu (< 5mg/kg) and normal Cu with low Mo but high levels of soluble protein related to high intake of fresh pasture. In this situation Cu deficiency results from the high levels of sulphide produced by reduction of sulphate in the rumen which in turn produces high levels of copper sulphide and thiomolybdates from which the Cu cannot be absorbed. The clinical signs of Cu deficiency include scouring, rough and bleached hair, slow growth and loss of body weight, pale membranes of the eyes and mouth and in some cases heart lesions (Mills, 1983).

(c) Iodine

Iodine is essential in the synthesis of thyroid hormones namely thyroxine and triiodothyroxine. The roles of these hormones are thermoregulation, intermediary metabolism, reproduction, growth and development, circulation and muscle function. Its primary function as a component of the thyroid hormone is to control the rate of oxidation of all cells. Iodine deficiency is characterised by general weakness, stunted growth, still born animals with goitre, suppression of oestrus periods in the female

and lack of libido in the male. Iodine deficiency remains a serious problem in many tropical countries, both for animals and humans. Low I intake is the primary cause of deficiency but the intake of goitrogens that interfere with its utilization, also cause deficiency problems. Diagnosis of severe deficiency is based on the clinical evidence of goitre; subclinical iodine deficiency may be detected by low serum iodine (thyroxine) or milk iodine concentration which is very sensitive to dietary intakes (McDowell *et al.*, 1993).

(d) Iron and Manganese

Iron occurs in the animal body complexed with the heme compounds: haemoglobin and myoglobin, as heme enzymes or as non-heme compounds. Animal metabolism and cellular respiration are some of the functions played by iron. Deficiency is rare in grazing ruminants unless loss of blood occurs.

Manganese on the other hand is important in maintaining normal bone structure, reproduction and normal functioning of the central nervous system. The metal also acts as a co-factor for enzymes involved in carbohydrate metabolism and mucopolysaccharide synthesis. Acid tropical soils are generally rich in Fe and Mn so that forage levels are generally in excess of requirements and deficiencies are unusual in the tropics (McDowell *et al.*, 1993). Clinical signs suggesting manganese deficiency have been reported from Costa Rica and the Mato Grosso regions of Brazil. Detection of manganese deficiency is aided by liver analysis and iron by haemoglobin and percent saturation of transferrin. Excesses of Fe and Mn may interfere with metabolism of other minerals including copper (McDowell *et al.*,1993).

(e) Selenium

Tissue breakdown or degeneration which occurs by oxidative stress of biological membranes is brought about by lack of vitamin E and Se. Selenium in the body promotes growth, reproduction, disease prevention and protection of integrity of tissues. It is a constituent of glutathione peroxidase (GSH-Px) - the enzyme that destroys organic peroxides before they can attack cellular membranes. Forages with >5 mg/Kg selenium are usually toxic to grazing ruminants whereas deficiency symptoms commonly occur when Se is below 0.1 mg/Kg (ARC, 1980; Chesters and Arthur, 1988). Selenium toxicity is usually associated with seleniferous soils, especially where these are alkaline. Liming can increase selenium toxicity by increasing plant absorption.

Chronic selenium poisoning is characterised by dullness, emaciation, rough hair coat, soreness, stiffness and lameness (caused by erosion of the joints), atrophy of the heart and cirrhosis of liver (MacPherson *et al.*, 1988). Remedial measures include soil treatment by lowering pH to reduce availability, treatment of the animal with arsenic to reduce absorption and increase excretion, and modification of the diet by rotation to low selenium pastures.

(f) Zinc

The key roles of zinc in the body are related to its association with enzymes as part of the molecule and as an activator. The stable structures of RNA, DNA and ribosomes are due to firmly bound zinc (Prask and Plocke, 1971).

Zinc deficiency in grazing animals is characterized by reduced feed intake, growth rate and feed efficiency, followed by skin disorder, hair loss, inflammation of the nose and mouth and stiffness of joints. Zinc deficiency also affects various stages of the reproductive process and development in both

males and females. All animals with these symptoms respond positively to Zn adminstration. Zinc deficiency is widespread in the tropics (Arora, 1988). Supplemental Zn can be provided by feeding mineral salts containing 20 to 30mg/kg zinc.

2.3 FACTORS INFLUENCING MINERAL REQUIREMENTS

A wide variety of factors influence mineral requirements of grazing ruminants. These include type and level of production, age of the animal, chemical form of elements, dietary intake level, breed and adaptation, interactions between elements (e.g. Ca-P, Fe-P, Al-P, Ca-Zn, Cu-Mo, Cu-Fe, Se-As-S, K-Na-Mg), as well as vitamins D, E and B_{12} availability. In addition, goitrogenic substances, oxalic acid and phytic acid influence the availability by acting as binders. Mineral deficiencies vary with season, in some cases being lower in the wet season and in other cases being higher in the dry season (Appleton, 1994).

2.4 SOURCES OF MINERALS TO GRAZING RUMINANTS

a) Water

Although soil is the principal source of trace elements entering the food chain, drinking water may be a significant source of minerals. For humans public water supply is normally low in trace elements and generally provides less than 10% of the required levels (Allaway, 1986). For animals, usually less than 2% of the trace element requirements for Fe, Zn, Cu and Se are provided in water. On average, water provide 3-12% of Mn and Co requirements of ruminants.

Water occasionally contains trace elements in toxic concentrations. The best example of the devastating effects to both humans and animals is the endemic fluorosis which occurs in several regions

of the world. In these regions, high fluoride in water seldom bears any relationship to the F status of soil.

In addition to F, wells and springs have been found to contain high concentrations of As, Li, Sr, B, Se and Pb. Water naturally high in Mo and/ or S can induce Cu deficiencies in grazing animals (McDowell, 1987; 1992).

b) Soil

Soils can influence animal nutrition status through the quantity and quality of the herbage they produce and the amount and type of soil ingested by grazing animals. Ingested soil can either be beneficial by improving performance from various rations through availability of essential trace elements like Co and Se (McDonald and Suttle, 1986) or injurious through excess uptake of toxic substances such as pesticides, cadmium and lead, or through abrasion of teeth (Healy and Ludwig, 1965). The bulk of ingested soil is probably taken in accidentally along with herbage and not because of depraved appetite. Under certain circumstances, soil ingestion by the grazing animal can constitute a substantial fraction of the diet. Deliberate soil consumption (geophagia), on the other hand, is classified as a form of "pica" which is defined as animals chewing objects and eating materials not considered to be natural feedstuff. The simplest and most common explanation for these behaviours is that the animals need some nutrient not available in the diet in sufficient quantities, and that they eat the abnormal substances to correct these shortages. Deliberate soil intake, in contrast to inadvertent soil intake with contaminated forage is normally selective. It usually concerns exploitation of special local deposits and sometimes even particular subsoils (McDowell, 1985).

c) **Forage**

Grazing ruminants do not often receive mineral supplementation except for common salt and must depend almost exclusively upon forages for their requirements. Only rarely, however, can tropical forages completely satisfy all mineral requirements (McDowell *et al.*,1993). Reduced forage intake due to factors such as low protein content (<7.0%) and increased degree of lignification is often responsible for decreased mineral amounts consumed.

Since tropical forages contain less minerals during the dry season, it is logical to assume that ruminants would most likely suffer mineral inadequacies during this time. On the contrary, numerous reports, including those from Kenya, Brazil and South Africa, have noted specific mineral deficiencies more prevalent during the wet season (McDowell, 1985).

2.5 METHODS OF ASSESSING MINERAL IMBALANCES IN ANIMALS

Three principal ways of assessing the incidence and impact of mineral imbalances in animals are:

(i) Use of clinical symptoms

(ii) Use of biochemical data from soil, plant and animal (blood or tissue) in relation to establishment criteria of normality.

(iii) Use of supplementation trials to check response.

There are problems associated with these methods and these have been discussed in several reports (Suttle, 1980; Suttle, 1986b; Suttle, 1987a; Langlands, 1987; Suttle, 1988).

2.5.1 USE OF CLINICAL SYMPTOMS

Problems associated with the use of symptoms as a diagnostic aid are:

(i) Some symptoms may overlap: the occurrence of anaemia does not give a clue as to which particular element is involved or the physiological or biochemical function impaired. Anaemia is a characteristic of Fe, Cu, and Co deficiencies and of Zn, Mo and Se toxicities.

(ii) The same deficiency does not always give the same symptom.

Mills *et al.*(1976), found a differential decline in the activity of dependent enzymes when they experimentally induced copper deficiency in calves. Effects of reduced activities of different enzymes depend on the duration and severity of the deficiency and the physiological development of the animal during that period. Symptoms of acute deficiency are often different from those observed with longer term, gradual chronic deficiency, possibly because different enzymatic activities are impaired (Suttle, 1983a).

It is important to note that not all deficiencies occur in isolation: dual deficiency cases involving Cu and Co have been reported in the "Coast disease" (Reuter, 1975b). Symptoms such as debility and starvation can be confused with "ill-thrift" syndromes unrelated to mineral disorders. Inconsistencies have been observed in diagnosing Co deficiency in "Coast disease" due to confounding symptoms of internal parasitism and protein malnutrition (Reuter, 1975b).

Copper deficiency can lower the production of wool in healthy sheep (Reuter, 1975b); however, other tests are needed to confirm areas of marginal deficiency because many nutritional and non-nutritional factors affect wool strength. Failure to conclusively assess the extent of mineral imbalances from

diagnostic records of visual symptoms has led to use of alternative procedures such as plant, soil and animal data assessment.

2.5.2 GEOCHEMICAL AND REGIONAL RECONNAISSANCE TECHNIQUES

The increasing awareness that quite subtle variations in both absolute and relative abundances of minor and trace elements in soils play a major part in determining the thriftiness of plants and animals has generated considerable interest in regional variations in soil chemistry. A major problem is posed by the fact that soils are capable of profound variations, both vertically and laterally, even when developed over homogeneous parent material. Consequently in many areas the density of sampling required to confidently describe the chemistry of soil by direct sampling is prohibited. A better method is by switching to indirect sampling techniques which give an average value for a large area with minimum samples. Four basic methods are used to achieve this:

(a) Geological mapping technique

Mineral deficiencies or toxicities in grazing ruminants can be predicted by use of systematic mapping survey techniques or regional reconnaissance especially within the tropics. Such mapping techniques were undertaken for Ca and P in Brazil, Se in Venezuela, and Co and Se in U.S.A. soils where concentrations have been related to Se and Co responsive diseases (McDowell *et al.*, 1984; Kubota, 1968; Kubota *et. al*, 1967). Deficiencies of P were discovered by mapping in Venezuela (Chico and French, 1959) and in Africa. Regional reconnaissance has also been used to identify mineral

problem areas. Van der Merwe (1967) and Boyazoglu (1973) reported likely deficiencies of Cu, Zn and Co in South African soils.

(b) Remote Sensing

Using remote sensing air craft or space craft, area identification based on colour or reflectivity variation induced in plants by variations in soil chemistry has been made. However, this method is limited to areas nurturing a very restricted variety of plants, and not all plants show significant colour or reflectivity responses to chemical characteristics of the soil (Joyce, 1975).

The disadvantage of this method is that soils of different chemical and physical character can develop over homogenous parental rock within a small area with some soils of agricultural importance being transported. Rock types show considerable chemical variation, but detailed chemical data are rarely available for map areas of significant size.

(c) Airborne Radiation Surveys

Airborne Scintillometer surveying based on radiation from Uranium (U) and K has been demonstrated as a reconnaissance technique for mapping soils in the U.S.A. This method has potential for rapid assessment of homogeneity and patterns of soil types but is limited to U and K only (Joyce 1975).

(d) Stream Sediment Geochemistry

Stream sediment geochemistry has offered a successful method of prospecting for concealed mineral deposits. The technique is based on the fact that the chemistry of a sediment sample

approximates to the average composition of the soils within the catchment area upstream of the sampling point (Thornton *et al*, 1969; Thomson *et al*, 1972; Thornton *et al*, 1972; Appleton, 1994).

2.5.3 USE OF BIOCHEMICAL INDICES (MARKERS) IN ANIMALS

The analysis of animal blood or tissue is generally considered to be more reliable than the visual symptoms or geochemical reconnaissance approach. Tissue samples analyzed may be blood, liver or fluids such as saliva and milk. These may be analyzed for the mineral itself, metabolite, enzymes, hormones or vitamins. Elemental status in blood reflects intake over an extended period. Payne *et al.*(1970) assessed the mineral contribution to metabolic or nutritional disorders using animal data from the "Compton Metabolic Profile Test" to identify abnormalities in the animal blood composition.

However, this test was criticized on the grounds of cost and the number of elements it could cover (Adams *et al*, 1978; Wolf *et al*, 1978). As previously mentioned, most of the elements deposit themselves in certain fluids or tissue in the body hence the choice of tissue or fluid for analysis is critical. Typical indices are given in Table 1.

Table 1: Best Tissue for Elemental Analysis

Element	Tissue/fluid
Calcium	Bone
Phosphorus	Bone
Magnesium	Urine/bone/plasma
Copper	Liver
Cobalt	Vitamin B_{12}/blood
Selenium	Whole blood/plasma
Sodium/Potassium	Saliva

Some of the disadvantages of using fluid and tissue analysis techniques are:-

(i) the specimen may not be easily obtainable unless by biopsy or by sacrificing the animal (Tartour, 1975).

(ii) there are high variations in concentrations of copper in the liver between animals.

(iii) concentrations can fall to very low values before health is affected since threshold values are ill-defined (Suttle, 1986a). In such cases plasma copper assessments are necessary.

2.5.4 USE OF RESPONSIVE CONDITIONS IN THE ABSENCE OF CLINICAL SIGNS.

In this technique mineral supplementation is administered and certain parameters such as female reproductive performance, wool production and growth monitored (Underwood and Somers, 1969; Reuter, 1975b; Suttle, 1986a). However, McDowell and Conrad (1977) cited some problems associated with this method with reference to tropical regions. These included:-

(i) Insufficient chemical analysis and biological data for determination of the required minerals and their content.

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

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

(iv) Supplements containing inadequate amounts or imbalances of certain elements.

(v) Standardized mineral mixtures that are inflexible for diverse ecological regions.

(vi) Farmers amending commercial mixtures thereby disregarding recommendations by the manufacturer.

(vii) Difficulty concerned with transportation, storage and cost of mineral supplements. Supplementation programmes can also be extremely expensive if the limiting elements have not been identified.

2.5.5 USE OF SOIL ANALYSIS

The chemical composition of soil offers alternatives for predicting mineral imbalances in grazing animals in various localities. Watkinson (1983) for example, used soil data to map Se deficient areas in New Zealand. Since soils are formed from the rock of parent material the mineral composition is similar to that of the bedrock. Mineral deficient areas have been associated with a number of factors including geological formations (Hartmans, 1970), climate and the leaching and weathering processes (Pfander, 1971; Langlands *et al*, 1981b), and soil drainage (Latteur, 1962; Pfander, 1971, Miller *et al*, 1972; Mitchell and Burridge, 1979). Assessment of soil mineral status (total or extractable) can indicate the need to apply deficient elements at forage establishment or sowing time. Most assessments are however based on availability, determined equilibrium and extraction of soils with chemical reagents. Procedures used tend to be empirical due to an incomplete understanding of the soil/soil solution/plant system since most tests are calibrated in glass house experiments where conditions may differ from those in the field. Therefore, extractable soil element concentrations correlate poorly with

plant uptake due to wide variations in soil properties including pH (Beckwith, 1963; Reith, 1965; Fleming, 1973; Loneragan, 1975), organic matter (Schnitzer and Khan, 1972), and uptake capacity of plant species (Allaway, 1968; Fleming, 1973; Reid *et al*, 1970; McDowell *et al.*, 1983)).

Since plants are the final arbiter of available mineral elements in the soil, assessment can be achieved by measuring plant content and relating this to the elemental status of the soil. The quantity of any element that is absorbed depends on the plant and the soil micro-climate at the time of sampling. The choice of extractant depends on both the element and the characteristics of the soil (Mitchell, 1971; Scott *et al*, 1971; West 1981). Critical soil concentration, below which deficiency can be expected, is estimated by regression analysis or by separating deficient and non-deficient soils into classes on the basis of plant analysis (Mitchell *et al*, 1957).

2.5.6 USE OF FORAGE ANALYSIS

Concentrations of minerals are dependent upon the type of soil, plant species, stage of maturity, yield, pasture management and climate. Mitchell *et al.* (1957) successfully used herbage analysis to assess mineral status of pastures to explain disorders in grazing ruminants. Mineral deficiencies in grazing animals are associated with certain regions although variations in mineral content have been reported in different plant species growing on the same soil. Disadvantages of herbage mineral analysis in assessing the mineral adequacy for grazing ruminants include:

(i) Uncertainty in taking samples representing what livestock consume because ruminants select certain species in periods of pasture abundance and also favour digestible protein herbage (Egan, 1975). In addition, P, Mg, K, Na, Cl, Cu, Co, Fe, Se, Zn and Mo tend to decline with forage maturity (Underwood, 1981).

(ii) It is difficult to estimate forage intake.

(iii) There is a variation in the availability of forage elements brought about by interactions between dietary components.

(iv) There is the possibility of taking soil-contaminated forages.

2.5.7 THE SOIL-PLANT-ANIMAL RELATIONSHIP

Investigations carried out by Russel and Duncan (1956) on the relationships between mineral levels in pastures and soils and the occurrence of mineral deficiency and toxicity diseases in grazing ruminants led to the recommendation that an integrated investigation of soil, plant and animal factors was needed for diagonising and resolving mineral imbalance diseases in animals. This approach was latter used by Kubota and Allaway (1972), Allaway (1975), Reuter (1975) and Underwood (1977). In the study by Reuter (1975) the approach involved determination of water and exchangeable mineral concentrations in soils and its relationship to plant, blood and tissue composition. Determination of enzyme activity and metabolite accumulation in plant and animal tissues was also thought to be of importance in understanding the relationship.

Whereas the most reliable method of confirming deficiency is probably by mineral supplementation trials, the problem with this is its high cost in time and resources. As already mentioned, clinical, pathological, biochemical, soil, water, plant, animal tissue and animal fluid analysis have all been used to diagonise trace element deficiencies and excesses.

Although extreme cases of trace deficiency and toxicity are often readily diagonised on the basis of clinical or pathological characteristics, diagnosis of subclinical cases is complicated by the fact that many of the symptoms of mild and transient mineral imbalances such as unthriftness, may also be caused by energy and protein deficiency as well as parasitism. Diagnosis of subclinical cases must

therefore rely on chemical and biochemical analysis for additional evidence. However, sampling of animals requires handling facilities and labour especially when more informative samples such as liver and bone biopsy specimens are required. Furthermore, for wildlife, the animal has to be sacrificed for the study and this can threaten the population of the particular species under national protection. These drawbacks point to the fact that the first step in mineral studies would be to assess soil and forage and by implication determine the elements limiting animal health and productivity.

CHAPTER 3

MATERIALS AND METHODS

3.1 LOCATION AND CHARACTERISTICS OF THE STUDY AREA

Nakuru district (7200 km^2) is located north west of Nairobi in the Rift Valley Province of Kenya. Most of the district lies within the floor of the Rift Valley at an average altitude of 1800 m. The district is bordered to the east by Nyandarua (elevation 2000-3000 m) and to the west by the Mau Ranges (elevation 2500 m).

3.1.1 Climate

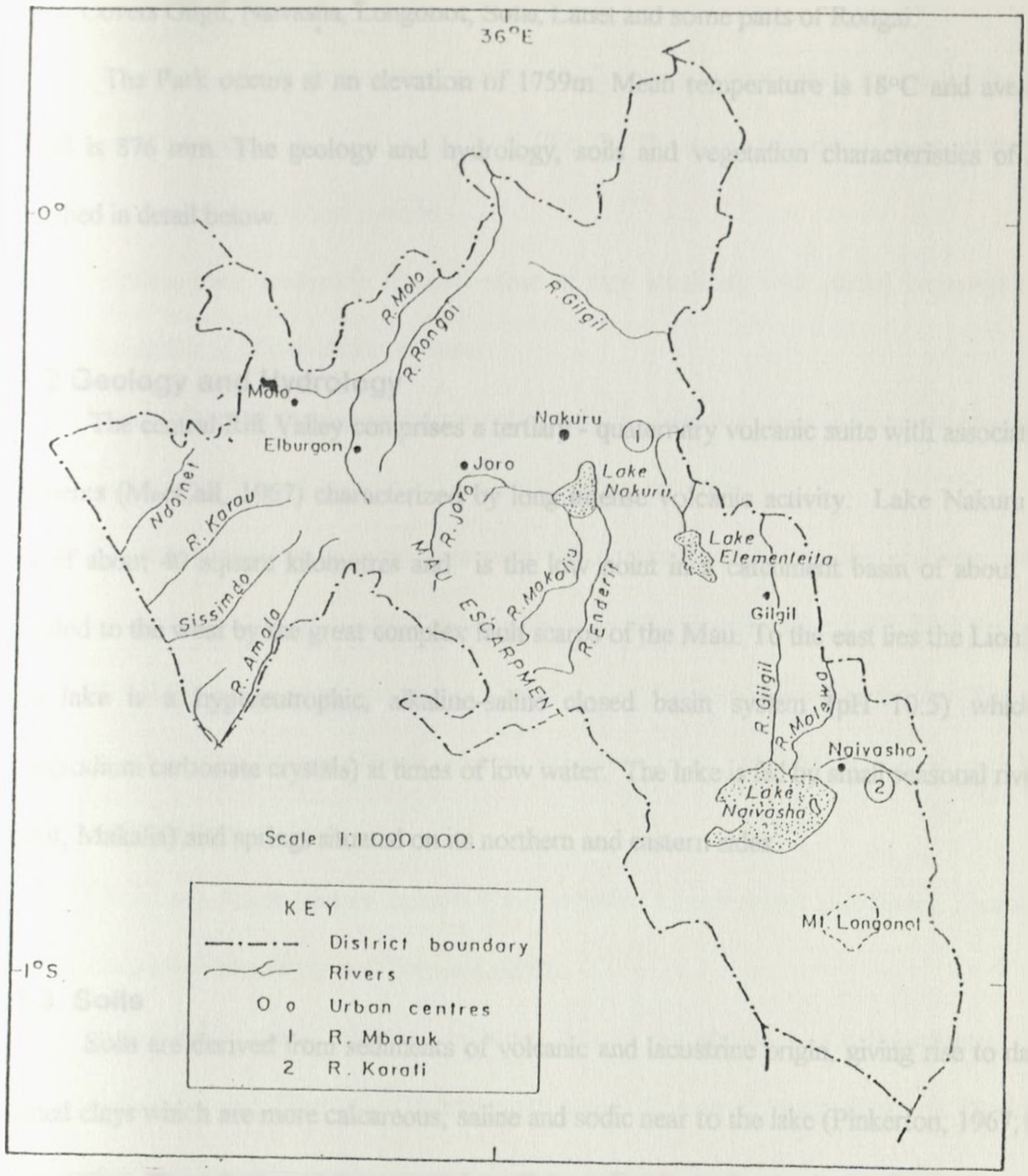
Climate in the district is strongly influenced by considerable altitudinal variation. The district is characterised by a bimodal rainfall pattern. The long rains fall between March and June while the short rains fall between October and December. There are large variations in average annual rainfall amount particularly in the marginal areas of the Rift Valley floor.

The district can be divided into three climatic zones as follows:

ZONE I: Rainfall over 1000 mm annually. Humid to semi-humid equatorial climate with a moisture index of less than 10. Covers Mau Narok, Molo, Olenguruone and Lower Subukia.

ZONE II: Rainfall between 700 and 1000 mm annually. Dry sub-humid equatorial climate with a moisture index of 10-30. Covers Bahati, Subukia, Njoro, Kinangop and parts of Rongai.

Figure 1. The watershed of Nakuru district.



In Figure 1, the position of Lake Nakuru is shown. This lake is fed by three rivers, all flowing from the south-west (Mau Escarpment).

ZONE III: Rainfall below 700 mm annually. Semi-arid climate with a moisture index of 30-42. Covers Gilgil, Naivasha, Longonot, Solai, Lanet and some parts of Rongai.

The Park occurs at an elevation of 1759m. Mean temperature is 18°C and average annual rainfall is 876 mm. The geology and hydrology, soils and vegetation characteristics of LNNP are described in detail below.

3.1.2 Geology and Hydrology

The central Rift Valley comprises a tertiary - quaternary volcanic suite with associated alkaline sediments (MacCall, 1967) characterized by long intense volcanic activity. Lake Nakuru covers an area of about 40 square kilometres and is the low point in a catchment basin of about 1,800 km^2 bounded to the west by the great complex fault scarps of the Mau. To the east lies the Lion Hill range. The lake is a hypereutrophic, alkaline-saline closed basin system (pH 10.5) which deposits trona(sodium carbonate crystals) at times of low water. The lake is fed by small seasonal rivers (Njoro, Nderit, Makalia) and springs situated on its northern and eastern sides.

3.1.3 Soils

Soils are derived from sediments of volcanic and lacustrine origin, giving rise to dark, poorly drained clays which are more calcareous, saline and sodic near to the lake (Pinkerton, 1967; Sombroek *et al.*, 1982). Plains further to the east and south have friable sandy clay loam, developed mainly from volcanic ashes (Sombroek *et al.*, 1982). The Mau escarpment and Lion Hill are mantled by andosols; reddish brown clays with a high humic horizon over phonolites, and phonolitic trachytes (Gethin-Jones and Scott, 1962; Sombroek *et al.*, 1982).

3.1.4 Vegetation

Around the lake is alkaline flats, marsh and alkaline grassland whilst the remainder of the Park is a mixture of grassland, shrub and woodlands [Fig.2]. The major vegetation zones of the Park were described by Kutilek (1974) and include:-

(1) Alkaline flats: seasonally flooded areas of high alkalinity with partial coverage of *Cyperus laevigatus* and *Sporobolus spicatus*.

(2) Marsh: flooded areas except during dry periods. Major species is *Cyperus laevigatus* which mixes with *Pluchea bequaertii* further away from the lake.

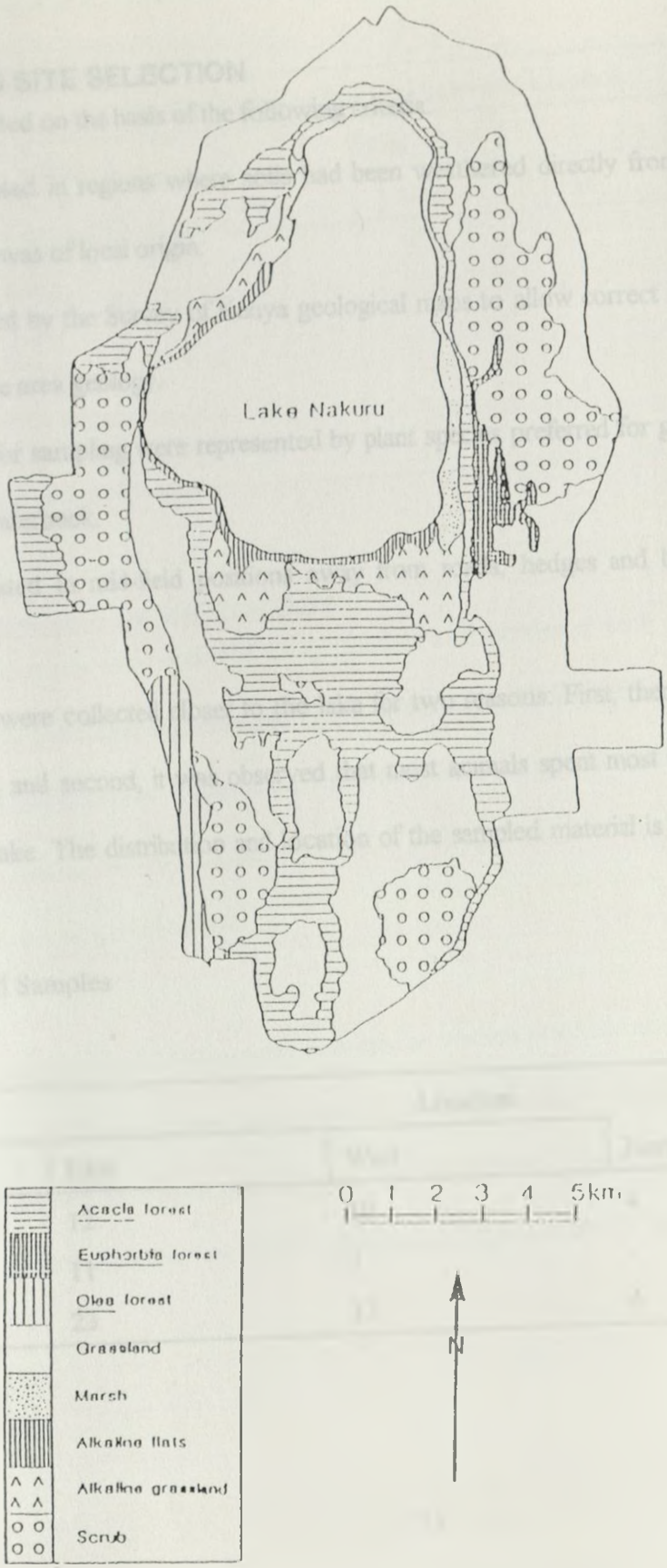
(3) Alkaline grassland: Alkaline flats with full coverage of *Sporobolus spicatus* and *Cyperus laevigutus* which mixes with *Cynodon dactylon* (Star grass) further away from the lake.

(4) Grassland: Major grass species are *Cynodon dactylon* and *Themeda triandra* (Red oat grass). Other species include *Chloris gayana* (Rhodes grass), *Hyparrhenia hirta* and *Pennisetum squamulatum*.

(5) Shrub: the major species include *Tarchonanthus camphoratus* (lelechwe), *Psaidia arabica, Kalanchoe densiflora* and *Rhus natalensis*.

(6) Woodlands: major species include Acacia xanthphloea (Yellowthorn), Euphorbia candelabra and Olea sp. Undergrowth plants feature Solanum incanum (Sodom apple), Justice flava, Achyranthes aspera, Senecio petitianus and Urtica massaica.

Figure 2. Vegetation of Lake Nakuru National Park.



3.2 SAMPLING SITE SELECTION

Sampling sites were selected on the basis of the following criteria:

a) Areas were situated in regions where soils had been weathered directly from the underlying rock or any drift was of local origin.

b) Area was covered by the Survey of Kenya geological maps to allow correct identification and description of the area geology.

c) Areas selected for sampling were represented by plant species preferred for grazing by wildlife especially the waterbuck.

d) Sites were located in mid-field positions away from roads, hedges and buildings to avoid contamination.

e) Most samples were collected closer to the lake for two reasons: First, there was evidence of active grazing, and second, it was observed that most animals spent most of their idling time closer to the lake. The distribution and location of the sampled material is shown in Table 2a and Table 2b.

Table 2a. Soil Samples

	Location		
Relative Dist.	East	West	North
Near	12	10	4
Far	11	7	-
Total	23	17	4

Table 2b. Forage Samples

	Relative Distance from the Lake and Location					
	East		West		North	
Species	Far	Near	Far	Near	Far	Near
Cynodon	7	9	4	7	-	2
Chloris	1	5	1	3	-	2
Digita.	2	-	-	-	-	-
Sporob.	-	2	-	-	-	-
Total	11	15	5	10	-	4

3.3 SAMPLING METHODOLOGY

a) Vegetation samples were collected using stainless steel sickles at each site on the grid at all locations. The dominant species(either *Cynodon dactylon, Chloris gayana, Themeda triandra, Digitaria swazilandensis* or *Sporobolus spicatus*) were sampled at several points within a 4 x 4m square where there was evidence of active grazing. Samples were kept in clean polythene bag sealed and properly labelled.

b) Soil samples (0-30cm) were collected at random positions within the same square from where the forages had been collected to give a representative sample of about 10kg. This was sub-sampled to 500gm according to the Hesse(1971) quartering method as described by Jumba (1989). Samples were kept in clean paper bags and labelled prior to storage.

3.4 SAMPLE TREATMENT

3.4.1 Soils

Soils stored under cool dry conditions were later dried at room temperature for three days in a dust free atmosphere. Once dry the soils were disaggregated with a pestle and mortar and screened through a 0.2mm (80 mesh) nylon sieve. The fraction passing through was retained for laboratory analysis.

3.4.2 Herbage

Herbage samples were dried in an oven at 70°C for forty-eight hours to a millable state. They were then ground in a Wiley Laboratory Mill, sieved through a 0.5mm stainless steel screen and stored in mini-grip polythene bags ready for elemental analysis

3.5 REAGENTS AND GLASSWARE

3.5.1 Chemicals

Chemicals were of Analar, Aristar, Pronalys or HPLC grade. They were obtained from normal laboratory suppliers including, BDH, Sigma, May and Baker, Aldrich Chemical Company and Koch-Light Ltd, UK.

3.5.2 Glassware

All glassware were thoroughly washed in laboratory detergent, rinsed in hot water and finally in de-ionized water(DIW). To leach out any contamination, they were soaked in 3M nitric acid overnight.Plastics were washed thoroughly in hot water and rinsed with de-ionized water before use.

3.6 INSTRUMENTATION TECHNIQUES

Concentrations of the trace and macro elements in the samples were determined using the following techniques:-

a) A Shimadzu Model AA-680 atomic absorption spectrophotometer (AAS) fitted with a graphic printer PR-5 (Shimadzu Corporation, 1991), and a Perkin-Elmer Model 2380 (AAS) fitted with a background correction accessory (Perkin Elmer, 1976) were used with appropriate burners for air/ acetylene, and nitrous oxide/ acetylene flames.

Where necessary, Programmable Autocalibration and Direct Concentration Readout modes were used.

b) Colorimetric determinations were performed on a Pye Unicam Model SP8-150 uv/visible spectrophotometer using 10mm pathlength quartz cells and background correction system.

3.7 pH DETERMINATION

The pH of each soil sample was determined according to the methods of Hesse (1971) and Black (1965). Each 20g sample portions was shaken with 50mls of water for 1hr. The pH of the 1:2.5 soil-water suspension was measured using a standard glass pH electrode and Pye Unicam Model 292 MK2 pH meter after standing overnight.

3.8 EXTRACTION OF MINERALS FROM SOILS

3.8.1 Available Phosphorus

Phosphorus was extracted with 0.5M sodium bicarbonate, pH 8.5 according to the procedure of Olsen *et al.*(1954). The volumetric soil:extractant ratio was 1:20 and shaking time 1hr at 27r.p.m. Phosphorus was measured colorimetrically with ammonium molybdate-ascorbic acid reagent

(Watanabe and Olsen, 1965. Excessively coloured extracts were decolorised with acid-leached activated charcoal.

3.8.2 Available Sodium, Potassium, Calcium and Magnesium

Sodium, potassium, calcium and magnesium were extracted with 1M ammonium acetate adjusted to pH 7 (Cahoon, 1974). The volumetric soil : extractant ratio was 1:10 and shaking (end over end) time 1hr at 27r.p.m. The extracts were filtered (Whatman No.42) and samples and standards made 0.2M with respect to HCl. For Ca and Mg sample aliqouts were made 0.25% with respect to Lanthanum Chloride before analysis.

3.8.3 Available Iron, Manganese and Zinc

Iron, manganese and zinc were extracted from the soils using the acid ammonium acetate-EDTA solution containing 0.5M CH_3COONH_4, 0.5M CH_3COOH, 0.02M Na_2EDTA. This solution was made by diluting 57.1ml glacial acetic acid, 37.3ml of 25% ammonia solution and 7.44g Na_2EDTA (ethylene diamine-tetraacetic acid disodium salt) to 1 litre with DIW. The pH was adjusted to 4.65 with acetic acid or ammonium hydroxide (Lakanen and Ervio, 1971). The volumetric soil: extractant ratio was 1:10 and shaking (end over end) time 1hr at 27r.p.m. The suspension was filtered using Whatman No.42 filter paper.

3.8.4 Available Copper

Copper was extracted from the soil samples using the acid ammonium acetate-EDTA extracting solution (Lakanen and Ervio, 1971) with a volumetric soil: extractant ratio of 1:5. The extracts were filtered through Whatman No.42 filter paper prior to analysis.

3.8.5 Total Cobalt, Cadmium and Lead

Cobalt, cadmium and lead were extracted with 4M HCl(Fordyce *et al.*, 1993). The volumetric soil: extractant ratio was 1: 5. The contents were boiled with ammonium acetate for ten minutes and filtered using Whatman No. 42 filter paper.

3.9 EXTRACTION OF MINERALS FROM FORAGE

For extraction of macro-(Ca, Mg, Na, K, P) and trace elements(Cu, Mn, Fe, Zn, Pb, Cd, Co, Mo, Al) from forage wet digestion was employed with lower temperatures and liquid conditions maintained throughout the oxidation process. Unless otherwise stated concentrated reagents were used viz: HNO_3(1.50), $HClO_4$ (s.g 1.54), H_2SO_4 (s.g 1.84). All samples were digested in pyrex boiling tubes, with de-ionised water substituting the sample for the blanks. The metals were extracted from 1g aliquots except for Co where digestion was done on 5g (with appropriate adjustment of acid volumes). Charing was prevented by dropwise addition of HNO_3 (Milner and Whiteside, 1984; Jumba, 1989).

After overnight predigestion with HNO_3 (10ml), (1g) samples were heated at 150°C for 1 hour. H_2SO_4 (0.5ml) and $HClO_4$ (1ml) were added and digestion continued until white fumes of $HClO_4$ appeared consistently for 30 minutes (reflux conditions). Digests were finally boiled in 1M HCl (15ml), filtered and stored in plastic vials awaiting analysis.

3.10 CHEMICAL DETERMINATION

3.10.1 Sodium and Potassium

Test solutions, calibrating standards and blanks were further diluted to give a suitable working range of 0-10mg/l Na and K. For soil extracts the dilutions were made in ammonium acetate, while the herbage digests were diluted with 1M HCl.

The metals were determined by flame emission spectrophotometry (Shimadzu Corporation, 1991) at 589nm (Na) and 766.5nm (K), respectively.

3.10.2 Calcium and Magnesium

Test solutions, calibrating standards and blanks were brought to the recommended working range of 0-5mg/l (Perkin- Elmer, 1976) by further dilution. For soil extracts dilutions were made in ammonium acetate while for herbage the dilutions were made in 1M HCl. La^{3+} was incorporated to a concentration of 0.31% to eliminate phosphate interference during analysis.

Calcium and Mg were determined by AAS at 422.7nm and 285.2nm, respectively using the air/acetylene flame (Price, 1972).

3.10.3 Iron, Manganese, Zinc, Cobalt, Copper, Lead and Cadmium

Test solutions, calibrating standards and blanks were further diluted where necessary to give a suitable working concentration range of 0-5mg/l for all the elements except Pb whose range was 0-20mg/l. For soil extracts, the dilutions were made with the appropriate extracting solutions, while herbage digests were diluted with 1M HCl.

The working solutions were atomised directly for the determination of Fe at 248.3nm, Zn at 213.9nm, Mn at 279.5nm, Co at 240.7nm, Cu at 324.8nm, Pb at 283.3nm and Cd at 228.8nm (Allan, 1959) by flame AAS. Analytical conditions were established as recommended by the instrumental manufacturers (Perkin-Elmer, 1976; Shimadzu Corporation, 1991).

3.10.4 Aluminium

Aluminium was determined by AAS at 309.3nm using a rich nitrous oxide/ acetylene flame (Price, 1972). Calibrating standards and blanks in the working concentration range 0-50mg/l were prepared in 1M HCl. All solutions were however made 1% with respect to K^+ ions by mixing 5ml aliqouts with 0.1ml of 19.6% KCl before analysis to prevent ionization interference.

3.10.5 Determination of Phosphorus

(i) Soil Extracts

Phosphorus in the soil extracts was determined by reacting it with excess molybdate ions in the presence of ascorbic acid to form a blue-coloured complex whose intensity was measured colorimetrically (Murphy and Riley, 1962; Watanabe and Olsen, 1965).

To 5mls of unknown extract was added 8mls of ammonium molybdate-ascorbic acid reagent and the contents mixed thoroughly. The solution was diluted to 100mls with DIW. The intensity of the coloured solutions was measured at 660nm and compared with that of a range of standards (0-2 mg/kg) prepared in the same matrix as the samples and similarly treated.

(ii) Forage Digests

Phosphorus in the plant 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. The intensity of the colour was measured at 470nm (Jumba, 1989).

To 1ml sample solution was added 1ml of stock vanado-molybdate reagent (a 50:50 mixture of 0.25% ammonium metavanadate in 33% nitric acid and 5% w/v aqueous ammonium molybdate). The contents were then diluted to 5mls and shaken thoroughly on a vortex mixer. The intensity of the coloured solutions was measured at 470nm. This procedure was a modifications of the method recommended by Gerike and Kurmies (1952) and Hesse (1971).

Standards 0-20mg/l were prepared in the same matrix as the samples and treated similarly.

3.10.6 Molybdenum

Molybdenum in the digests was determined spectrophotometrically by the dithiol method of Bingley (1959; 1963), with some minor modification as described by Jumba (1989).

To 15mls of each test solution was added 0.25ml Fe(III) solution (10% ferric ammonium sulphate in 2% H_2SO_4) and the volume adjusted to 25mls with DIW. 0.25ml of 50% KI was added and the mixture allowed to stand for ten minutes with occasional swirling. Iodine liberated was then discharged by dropwise addition of 10% sodium thiosulphate before addition of 0.25ml of 50% tartaric acid followed by 2mls of 10% thiourea. 2mls of dithiol reagent(2% 4-methyl-1,2-dimercaptobenzene in 1% sodium hydroxide) prepared according to the procedure of Bingley (1959) was added and the contents mixed and allowed to stand for 30 minutes. The complexed Mo was extracted by shaking with 5mls of isoamyl acetate (boiling point range 136-142°C) for thirty seconds.

The organic layer was drawn off and filtered (Whatman No. 1) before measuring the intensity of the green Mo-dithiol complex at 680nm using amyl acetate in the reference channel. Standards 0-10μg Mo were prepared in the same acid matrix as the samples and treated similarly.

3.11 STATISTICAL ANALYSIS OF DATA

Data were analyzed by the Generalized Linear Model (GLM) of the Statistical Analysis Systems (SAS, 1985). For any unequal subclass representations yielding unbalanced data, hypothesis testing based on sum of squares was applied.

Samples were classified according to location (i.e north, east, west or south) and forage by species. Duncan's Multiple Range Test was then used to test significant differences between regional means (Duncan, 1955).

Determination of the significance of differences entailed comparing the means of relevant quantities. Since means tended to be grossly affected by unusually low or high values, a preliminary analysis was carried out on the distribution of the data, using the UNIVERIATE procedure in SAS in order to detect the existence of extreme values. Consequently, extreme values were replaced by the averages of the non-extreme values for a given site.

Since in all cases under investigation more than two means were being compared, use was made of multiple comparison procedures as implemented in the GLM procedure in SAS (SAS, 1985). As the sample sizes were not necessarily equal, the current analysis used Duncan's multiple range test which minimizes the comparisonwise error rate and whose power does not diminish as the number of means increases (Duncan, 1955).

Analysis was carried out at the significance levels P=0.05 and P=0.01. Particular note was taken of highly significant differences, namely differences detected with P=0.01.

CHAPTER 4

RESULTS AND DISCUSSION

4.1 SYMBOLS AND UNITS

Although stated previously, the parameters determined, on dry matter (DM) basis, together with their symbols and units of expression are presented here for ease of reference (Table 3).

Table 3. Symbols and units of Parameters Determined

Symbol	Parameter	Soil Units	Forage Units
Al	Aluminium	-	g/kg
Cd	Cadmium	mg/kg	mg/kg
Ca	Calcium	g/kg	g/kg
Co	Cobalt	mg/kg	mg/kg
Cu	Copper	mg/kg	mg/kg
Fe	Iron	mg/kg	mg/kg
K	Potassium	g/kg	g/kg
Mg	Magnesium	mg/kg	g/kg
Mn	Manganese	mg/kg	mg/kg
Mo	Molybdenum	-	mg/kg
Na	Sodium	g/kg	g/kg
P	Phosphorus	mg/kg	g/kg
Pb	Lead	mg/kg	mg/kg
Zn	Zinc	mg/kg	mg/kg

4.2 RESULTS

4.2.1 Soil Analysis

(a) Soil Acidity and Macromineral Composition

Results of the batch analysis of soils are presented in appendices I and II. A summary is presented in Tables 4a and Table 4b

Table 4a. Mean Soil pH and Macromineral Data: Soil Regional Data

Parameter	ov./mean	East	North	West
Ca	2.705	3.066	1.801	2.431
Mg	113	170	55.6	50.9
Na	1.823	2.307	1.210	1.643
K	2.020	2.275	1.714	1.780
P	28.9	30.0	37.4	25.5
pH	7.671	8.089	6.475	7.388

Table 4b. Mean Soil pH and Macromineral Data: Soil Distance Data

Parameter	Overall mean	Far	Near
Ca	2.705	2.584	2.790
Mg	113	169	74.9
Na	1.823	1.168	2.243
K	2.020	1.711	2.243
P	28.9	27.6	29.9
pH	7.67	7.57	7.74

ov./mean = overall mean

Soil pH

The mean soil pH was 7.67 with a range of 3.97-10.2. Most pH values were above 5-7, the range which enables maximum absorption of ionic species by plants to occur (Reid and Horvath, 1980). The pH was highest in the east (8.09) followed by west (7.39) and north (6.47) but the differences were not significant. The pH values tended to be slightly higher towards the lake (mean: 7.74) but the difference between these and those of soils further away from the lake (mean: 7.57) was not significant.

Calcium

Concentrations of Ca in the sampled soils ranged from 0.863-4.85g/kg with a mean of 2.71g/kg. None of the samples had Ca below the critical level of 71 mg/kg which signifies deficiency (Breland, 1976). The mean Ca in soils was highest in the east (3.066), followed by west (2.431) and North (1.801), with the difference between east and north being significant ($P<0.05$). The mean Ca concentration was higher near the lake (2.790) than it was far from the lake (2.584) but the difference was not significant.

Magnesium

Concentrations of Mg in soils ranged from 6.53-405 mg/kg with a mean of 114mg/kg. 27% of the samples were found to have concentrations below 30 mg/kg, a level suggested to be critical for most plant species in terms of deficiency (Rhue and Kidder, 1983).

The concentrations were highest in the east (mean: 170), followed by north (mean: 55.6) and west(mean: 50.9), with the difference between the east and the other two areas being significant

(P<0.05). The concentrations also increased significantly (P<0.01) with distance from the lake, from 74.9 to 169mg/kg

Sodium

The mean Na concentration in the soils was 1.82g/kg while the range was 0.198-5.27g/kg. The concentrations were highest in the east (mean: 2.307), followed by north (mean: 1.643) and west (mean: 1.210) but the differences were not significant. Concentrations were higher towards the lake (mean: 2.276) than farther away from the lake (mean: 1.168), with the difference being significant (P<0.05).

Potassium

The mean K concentration in the soils was 2.02g/kg. Concentrations ranged from 0.495 to 4.44g/kg. None of the samples had concentrations below the critical level of 0.06g/kg (McDowell and Conrad, 1977; Bahia,1978; Rhue and Kidder, 1983).

Concentrations were highest in soils from the east (mean: 2.275), followed by north(mean: 1.780) and west(mean: 1.714) but the differences were not significant. Concentrations were higher near the lake(mean: 2.243) but decreased with distance away from the lake(mean: 1.711) although the difference was not significant.

Phosphorus

Soil extractable P ranged from 9.12-1210 mg/kg with a mean of 28.9mg/kg. 11% of the samples were found to have concentrations below the critical level of 17mg/kg (Rhue and Kidder, 1983). Concentrations were highest in the north (mean: 37.45), followed by the east(mean: 30.01) and

the west (mean: 25.51) but the differences were not significant. Concentrations were slightly higher near the lake (mean: 29.9) but decreased to 27.6mg/kg farther away from the lake. However, the difference was not significant.

(b) Soil Trace Element Composition in Relation to Distance and Region

Table 5a. Concentrations of Trace Elements in the Soils: Soil regional data

Element	ov./mean	East	North	West
Co	1.03	1.301	0.821	0.713
Cu	0.784	1.105	0.654	0.381
Cd	0.134	0.158	0.142	0.099
Fe	141	175	137	97.7
Mn	408	542	271	259
Pb	5.95	6.68	5.84	5.00
Zn	4.23	6.18	4.69	1.49

Overall Means

Element	Overall mean	Near	Far
Co	1.030	0.959	1.133
Cu	0.784	0.780	0.791
Cd	0.134	0.122	0.152
Fe	141	108	190
Mn	408	381	447
Pb	5.95	5.51	6.60
Zn	4.23	3.26	5.65

ov./mean (overall mean)

Cobalt

The mean Co concentration was found to be 1.03mg/kg with a range of 0.459-14.8mg/kg. None of the samples had a concentration below the critical level of 0.1mg/kg (Mtimuni, 1982). Concentration were highest in the east (mean: 1.301), followed by north (mean: 0.821) and west (mean: 0.713). The difference between the east and the other two areas was highly significant ($P<0.01$). Concentration were also slightly higher in soils collected farther away from the lake (mean: 1.133) than near the lake (mean: 0.959), but the difference between the levels was not significant.

Copper

Concentrations of copper ranged from 0.17-2.28mg/kg with a mean of 0.784mg/kg. If 2mg/kg is taken as critical level that would reflect deficiency (Bahia, 1978), then only two sites satisfied the requirement. If 1mg/kg is considered as the critical value (McDowell *et al.*, 1983), then sixteen sites satisfied the requirement. If 0.6mg/kg is considered (Mtimuni, 1982), then twenty-five sites (57%)

were adequate in Cu; but if 0.3mg/kg is considered as the critical value (Rhue and Kidder, 1983) then thirty-five (80%) of the sites were adequate in Cu. Whatever the case, it would appear that most forages in the Park may be deficient in Cu. Concentration were highest in the east (mean: 1.105), followed by the north (mean: 0.654) and the west (mean: 0.381). The difference between east and west was significant ($P<0.05$). Mean Cu concentrations were approximately the same (about 0.8) irrespective of the distance of the sampling site from the lake.

Iron and Manganese

Concentrations of iron in the soil fell in the range 21-395mg/kg with a mean of 141mg/kg. One sample was found to be below the critical deficiency level of 30mg/kg suggested by Bahia (1978). Concentrations were highest in the east (mean: 175), followed by north(mean: 137) and west (mean: 97.7) but the differences were however not significant. Concentrations were higher farther away from the lake(mean: 190) compared to samples collected closer to the lake (mean: 108) with the difference being significant($P<0.05$).

Concentrations of Mn ranged from 20-1141mg/kg with a mean of 408mg/kg. None of the sample concentrations was found to be below 10mg/kg, the critical deficiency level (Mtimuni, 1982). Concentrations were highest in the east (mean: 542), followed by north (mean: 271) and west (mean: 259), with the difference between the east and the other two areas being significant ($P<0.05$). Concentrations also increased with distance from the lake from 381 to 447mg/kg but the difference was not significant.

Zinc

Concentrations of Zn ranged from 0.22-18.3 with a mean of 4.23mg/kg. Compared with the suggested critical level of 8mg/kg (Bahia, 1978), 84% of the soils were deficient in this element. However, if 1mg/kg (Rhue and Kidder, 1983) is considered, then 18% were deficient. And if 6mg/kg (McDowell *et al.*, 1983) is taken as the critical value, then 77% were deficient compared to 41% for a 2mg/kg critical level (Mtimuni, 1982). Whatever the case, the results indicated prospects of prevalent deficiency in the soils.

Concentration were highest in soils in the east (mean: 6.18), followed by north (mean: 4.69) and west (mean: 1.49). The differences between east and west were significant ($P<0.05$). Although concentrations were higher in samples collected far from the lake (mean: 5.65) than closer to the lake (mean: 3.26), the difference was not significant.

Cadmium

Cadmium concentrations in soils were highest in the east (mean: 0.158), followed by north (mean: 0.142) and west (mean: 0.099), but the differences between these levels were not significant. Concentrations were also higher far from the lake (mean: 0.152) than it was near the lake (mean: 0.122), but the difference was not significant.

Lead

Mean Pb concentrations in the soils were highest in the east (6.68), followed by north (5.84) and west (5.00), but the differences were not significant. Concentration were also higher in samples collected far from the lake (mean: 6.60) than closer to the lake(mean: 5.51) but the difference was not significant.

4.2.2 Forage Analysis

(a) Major Elements

Results of the forage analysis are presented in appendices III and IV. A summary is given in Tables 6a and 6b.

Calcium

Concentrations of Ca ranged from 1.09-14.8g/kg DM with a mean of 3.02g/kg DM. According to ARC (1980) a range of 1.7-2.8g/kg DM is recommended for growth, while lactating animals require higher dietary concentrations, in the range 2.8-3g/kg DM. Based on these requirements, 95% and 48% of the forages could be considered adequate for growing and lactating animals, respectively in the study area.

Concentrations were highest in the east (mean: 3.422), followed by the north (mean: 2.505) and the west (mean: 2.477), but there were no significant differences. The Ca concentrations were highest in *Sporobolus spicatus* (mean: 3.290), followed by *Cynodon dactylon* (mean: 3.217), *Chloris gayana* (mean: 2.635) and *Digitaria swazilandensis* (mean: 2.320) but the differences were not significant.

Table 6a. Forage Mean Macromineral Concentrations (g/kg DM) in Relation to Region

Element	Ov./mean	East	North	West
Ca	3.025	3.422	2.505	2.477
Mg	0.856	0.940	0.926	0.693
Na	0.947	0.959	0.959	0.924
K	7.349	6.836	6.105	8.569
P	1.847	1.923	1.802	1.727

Table 6b. Forage Mean Macromineral Concentrations (g/kg DM) in Relation to Species

Element	Ov./mean	*Chloris*	*Cynodon*	*Digitaria*	*Sporo.*
Ca	3.025	2.635	3.217	2.320	3.290
Mg	0.856	0.763	0.888	0.933	0.875
Na	0.947	0.887	1.014	0.631	0.685
K	7.349	8.050	6.694	12.90	7.085
P	1.847	1.726	1.930	2.120	1.090

Ov./mean (overall mean)

Magnesium

The mean forage Mg concentration was 0.856g/kg DM with a range of 0.149-2.66g/kg DM. Majority (65%) of the samples had concentrations between 0.149 and 1g/kg DM. 63% of the samples were below 1.0g/kg DM the level considered critical in animal diets by the Agricultural Research Council (ARC, 1980) compared with 95% if a level of 2g/kg DM is considered as critical according to

other reports (McDowell *et al.*, 1984). However, only 15% of the forages had concentrations that met the requirement for lactating animals (1.4-2.1g/kg DM, McDowell *et al.*, 1993).

Forage Mg concentrations were highest in the east (mean: 0.940), followed by north (mean: 0.926) and west (mean: 0.693) but the differences were not significant. And according to species, concentrations were highest in *Digitaria swazilandensis* (mean: 0.933), followed by *Cynodon dactylon* (mean: 0.888), *Sporobolus spicatus* (mean: 0.875) and *Chloris gayana* (mean: 0.763) but the differences were again not significant.

Sodium

The mean forage concentration was 0.947g/kg with a range of 0.054-14.5g/kg DM. 42% of the forages had concentrations below the critical level of 0.6g/kg recommended for growth (McDowell, 1992; McDowell *et al.*, 1984) and 72% of the forages were below the minimum requirement of 1.8g/kg DM for lactating animals (McDowell *et al.*, 1993). And according to NRC (1985), 52% of the forages could not meet the minimum requirements for grazing ruminants. Na was generally found to be deficient in the forages but this could be offset by the salty water available to the animals for drinking.

Mean concentrations in forage were slightly higher in the north and east (0.959) than in the west (0.924) but the difference was not significant. Concentrations were highest in *Cynodon dactylon* (mean: 1.014), followed by *Chloris gayana* (mean: 0.887), *Sporobolus spicatus* (mean: 0.685) and *Digitaria swazilandensis* (mean: 0.631) but the differences were not significant.

Potassium

The mean forage K concentration was 7.35g/kg DM with a range of 2.67-26.1g/kg DM. 68% of the forages were found to have concentrations below 8g/kg recommended for lactating animals (McDowell *et al.*, 1984; 1993), while 85% of the forages had concentrations above the 5g/kg DM, a level necessary to support growing animals.

Concentrations in the forages were highest in the west (mean: 8.569), followed by east (mean: 6.836) and north (mean: 6.105) but the differences were not significant. For species, concentrations were highest in *Digitaria swazilandensis* (mean: 12.9), followed by *Chloris gayana* (mean: 8.05), *Sporobolus spicatus* (mean: 7.085) and *Cynodon dactylon* (mean: 6.694). The difference between *Digitaria swazilandensis* and the other species was significant ($P<0.05$).

Phosphorus

Forages sampled had a mean P level of 1.8g/kg DM with a range of 0.243-4.89g/kg DM. 87% of the forages had concentrations below the deficiency critical level of 2.5g/kg DM recommended by ARC (1980), while 82% were below the minimum maintenance limit (without lactation) of 2.3g/kg DM (McDowell *et al.*, 1993).

Only 3% of the forages had concentrations above the minimum requirement of 3.9g/kg DM (Suttle, 1983b) for lactating animals. In general, the forages were found to be deficient in P.

Concentrations in forage were highest in the east (mean: 1.923), followed by north (mean: 1.802) and west (mean: 1.727) but the differences were not significant. The mean P concentration was highest in *Digitaria swazilandensis* (2.120), followed by *Cynodon dactylon* (1.930), *Chloris gayana* (1.726) and *Sporobolus spicatus* (1.090) but again the differences were not significant.

(b) Trace Elements

Tables 7a and 7b give the results of trace element concentrations in the forages

Cobalt

Concentrations of Co ranged from 0.202-2.03mg/kg DM with a mean of 0.573mg/kg DM. None of the forages had concentrations below 0.08mg/kg DM, the deficiency critical level for grazing ruminants (ARC, 1980; NRC, 1985; McDowell and Conrad, 1977). Concentrations in forage were highest in the west (mean: 0.674), followed by east (mean: 0.54) and north (mean: 0.405) but the differences were not significant. The Co concentration was highest in *Cynodon dactylon* (mean: 0.631), followed by *Digitaria swazilandensis* (mean: 0.534), *Sporobolus spicatus* (mean: 0.519) and *Chloris gayana* (mean: 0.448) but the differences were not significant.

Copper and Molybdenum

Forage Cu ranged from 1.79 mg/kg level to 43mg/kg DM with a mean of 8.56mg/kg DM. 63% of the forages had Cu level below 10mg/kg DM, the critical level below which deficiency symptoms occur in grazing animals (McDowell *et al.*, 1984; 1993; McDowell and Conrad, 1977). Considering a dietary allowance of 8mg/kg DM for both growing and lactating species (ARC, 1980; NRC, 1985), 45% of the herbages were deficient in Cu.

The mean Cu concentration was highest in the east (9.412), followed by west (8.153) and north (4.588) but the differences were not significant. Concentrations were highest in *Sporobolus spicatus* (mean: 14.00), followed by *Chloris gayana* (mean: 9.137), *Cynodon dactylon* (mean: 8.163) and *Digitaria swazilandensis* (mean: 5.495) but the differences were also not significant.

Concentrations of Mo in the herbage ranged from 0.37-23.7 mg/kg DM with a mean of 4.44mg/kg DM. 27% of the samples contained > 6mg/kg DM of Mo, a level regarded as toxic to grazing animals (ARC, 1980; NRC, 1985; McDowell *et al.*, 1993).

Table 7a. Trace Element Composition of LNNP Forage in Relation to Species

Element	Ov./mean	*Chloris*	*Cynodon*	*Digitaria*	*Sporo.*
Al	0.432	0.484	0.417	0.457	0.312
Cd	0.149	0.115	0.164	0.172	0.128
Co	0.573	0.448	0.631	0.534	0.519
Cu	8.564	9.137	8.163	5.495	14.00
Fe	518	598	509	445	251
Mn	78.8	74.1	80.1	55.8	112
Mo	4.442	3.772	4.703	5.233	3.890
Pb	0.722	0.513	0.799	0.647	0.927
Zn	16.8	16.6	16.3	18.8	22.8

Table 7b. Trace Element Composition of LNNP Forage in Relation to Region

Element	Ov./mean	East	North	West
Al	0.432	0.386	0.531	0.486
Cd	0.149	0.164	0.116	0.133
Co	0.573	0.540	0.405	0.674
Cu	8.564	9.412	4.588	8.153
Fe	518	440	603	633
Mn	78.8	79.9	67.8	80.0
Mo	4.442	3.916	3.473	5.613
Pb	0.722	0.920	0.321	0.485
Zn	16.8	20.2	14.6	11.6

The mean Mo concentration was highest in the west (5.613), followed by east (3.916) and north (3.473) but the differences were not significant. Concentrations were highest in *Digitaria swazilandensis* (5.235), followed by *Cynodon dactylon* (4.703), *Sporobolus spicatus* (3.890) and *Chloris gayana* (3.772) but the differences were not significant.

Iron and Manganese

The mean herbage Fe was found to be 318mg/kg DM with a range of 30-1698mg/kg DM. All the samples met the requirements for grazing ruminants of 30mg/kg DM (ARC, 1980; NRC, 1985). There may be a problem of Fe overload as 25 out of the 67 sampled forages had concentrations above 500mg/kg DM, the level regarded as the maximum tolerable by ruminants (ARC, 1980).

Mean Fe concentrations in forages were highest in the west (633), followed by the north (603) and east (440) but the differences were not significant. Concentrations were highest in *Chloris gayana* (mean: 598), followed by *Cynodon dactylon* (mean: 509), *Digitaria swazilandensis* (mean: 445) and *Sporobolus spicatus* (mean: 252) but the differences were not significant.

Concentrations of Mn in the sampled herbage ranged from 22.9-294mg/kg DM with a mean of 78.8mg/kg DM. Examination of the results revealed that majority of the samples had concentrations above 25mg/kg DM, the critical level below which growing animals show deficiency symptoms (ARC, 1980). 9% of the forages had levels below 40mg/kg DM, the critical level for lactating animals (McDowell *et al.*, 1993). Manganese was generally sufficient considering that even a lower value (20mg/kg DM) has been suggested by different investigators as being critical (NRC, 1985; McDowell and Conrad, 1977).

Concentrations were highest in the west (mean: 80.0), followed by east (mean: 79.8) and north (mean: 67.8) but the differences were not significant. The mean Mn concentration was highest in *Sporobolus spicatus* (112), followed by *Cynodon dactylon* (80.1), *Chloris gayana* (74.1) and *Digitaria swazilandensis* (55.8). However, the differences were not significant.

Zinc

Concentrations of Zn ranged from 2.2-55.8mg/kg DM with a mean of 16.8mg/kg DM. 94% of the samples were below 40mg/kg DM, the deficiency critical level (McDowell and Conrad, 1977) while none of the sample concentrations was above 150mg/kg DM, hence Zn toxicity not a problem (McDowell *et al.*, 1993).

The mean Zn concentration was highest in the east (20.2), followed by north (14.6) and west (11.6) but the differences were not significant. According to species, concentrations were highest in *Sporobolus spicatus* (mean: 22.8), followed by *Digitaria swazilandensis* (mean: 18.8), *Chloris gayana* (mean: 16.6) and *Cynodon dactylon* (mean: 16.3) but the differences were not significant.

Aluminium

The Al concentrations in forage were highest in the north (mean: 0.531), followed by west (mean: 0.486) and east (mean: 0.386) but the differences were not significant. *Chloris gayana* had the highest mean concentration of Al (0.484), followed by *Digitaria swazilandensis* (0.457), *Cynodon dactylon* (0.417) and *Sporobolus spicatus* (0.312) but the differences were also not significant.

Cadmium

The mean Cd concentration in forage was highest in the east (0.164), followed by west (0.133) and north (0.116) but the differences were not significant. The Cd concentration was highest in *Digitaria swazilandensis* (0.172), followed by *Cynodon dactylon* (0.164), *Sporobolus spicatus* (0.128) and *Chloris gayana* (0.115) but the differences were not significant.

Lead

The mean Pb concentration in forage was highest in the east (0.920), followed by west (0.485) and north (0.321) but the differences were not significant. Concentrations were highest in *Sporobolus spicatus* (mean: 0.927), followed by *Cynodon dactylon* (mean: 0.799), *Digitaria swazilandensis* (mean: 0.647) and *Chloris gayana* (mean: 0.513); however the differences were not significant.

4.3 DISCUSSION

4.3.1 Criteria for Assessing Mineral Requirements of Grazing Animals

In animal nutrition, mineral requirements are usually based on factors determined using a factorial approach (ARC,1980). The requirements entail:

(1) Net minimum endogenous requirement (E) taken as the inevitable loss of the element from the body in faeces and urine

(2) Net requirement for body growth (G) taken as the daily retention of the element at the specified rate and stage of growth

(3) Net requirement for pregnancy (P) taken as the daily retention of the element in the foetus and adnexia at the specified stage of pregnancy

(4) Net requirement for lactation (L) taken as the daily secretion of the element in milk at the specified yield

(5) Total net requirement (N) taken as the sum of all the requirements above (i.e N=E+G+P+L).

(6) Dietary requirement (D) taken as the net requirement divided by the absorption coefficient,A, i.e D= N/A= (E+G+P+L)/A

(7) Absorption defined as the amount of a mineral supplied in the diet that enters the body through the gut, i.e the apparent absorption plus the net faecal endogenous excretion. The coefficient of absorption is the amount absorbed divided by the amount ingested.

Using animal trials in both field and laboratory experiments, different authorities have suggested different requirements for grazing animals in different parts of the world. The requirements for each element for the specified stage of development (whether young growing, body maintenance, pregnancy or lactating) have been determined. Using these requirements, it is therefore possible to predict whether a deficiency, toxicity or imbalance is prevalent in a given area. Authorities referred to most commonly are the Agricultural Research Council of the Commonwealth Bureaux, UK and the National Research Council of America.

4.3.2 Assessment of Results in Relation to Animal Requirements

Calcium

Soil Ca results found in this study (mean: 2.71g/kg) were similar to those obtained in Guatemala (mean: 2.22g/kg, Tejada *et al.*, 1987). However, they were higher than those obtained in Colombia (mean: 0.15g/kg, Vargas *et al.*, 1984) and USA (mean: 1.24g/kg, Espinoza *et al.*, 1991). Forage Ca (mean: 3.02g/kg DM) was however similar to that obtained in USA (mean: 4.2g/kg DM, Espinoza *et al.*, 1991) and Guatemala (mean: 4g/kg DM, Tejeda *et al.*, 1987). Concentrations were however higher than those obtained in Western Kenya (mean: 1.48g/kg , Jumba, 1989).

Concentrations of Ca were particularly low in *Chloris gayana* (mean: 2.63) and *Digitaria swazilandensis* (mean: 2.32). However, they were within the minimum dietary allowance (2.0 - 6.7g/kg DM) recommended for ruminants growth (ARC, 1980). The other species had levels above 3g/kg DM, a level recommended for lactating animals (McDowell *et al.*, 1984). Furthermore, soil Ca was much higher than the critical level (71mg/kg, Breland, 1976), implying that cases of Ca deficiency should not

be expected on the sampled sites. The low Ca levels in the forages could be attributed to the stage of maturity at which the grasses were sampled (hay stage) since mineral content of plants decline with age (Tergus and Blue, 1971).

The levels of Ca found in the sampled forages indicates prospects of Ca deficiency disorders in ruminants in LNNP. However, typical symptoms attributed to Ca deficiency may not develop abruptly since animals can tolerate low dietary Ca concentrations for fairly long periods (even upto 6 months).

Evidence from research experiments indicates that animals adapt to low Ca diets by increasing the capacity for intestinal absorption of Ca (Dowdle *et al.*, 1960; Kimberg *et al.*, 1961), a phenomenon which is considered to be related to the degree of depletion of the skeleton (Nicolayson *et al.*, 1953). Thus there is a wide margin between the low dietary concentrations shown to cause clinical abnormalities by experiment and the dietary requirements which various authorities recommend as the basis for ration formulation (ARC, 1980). The recommended allowances contain a margin of safety and animals apparently have there own buffering mechanisms which enable them to meet temporary shortfalls . However, with 52% of the sampled forages having Ca below 3g/kg DM (ARC, 1980), signs of ill-health may occur in this area, a situation more likely to be seen in domestic rather than wild animals.

Magnesium

The soil Mg results obtained in this study(mean: 114mg/kg) were higher than those obtained in Western Kenya because of the high soil pH which favours Mg availability (Jumba *et al.*, 1995a) and Colombia (mean: 39mg/kg, Vargas *et al.*, 1984). However, herbage Mg concentration (mean:

0.856g/kg DM) was similar to that obtained in Central and Southwestern regions of Guatemala(mean: 1.1g/kg DM, Tejeda *et al.*, 1987) but lower than that obtained in Western Kenya (mean: 1.61g/kg DM, Jumba *et al.*, 1995a).

Although 27% of the total soil samples were found to be deficient in magnesium, the overall mean was surprisingly high (114mg/Kg) compared to the critical level of 30 mg/Kg (Rhue and Kidder, 1983). Despite this however, 95% of the sampled forages had concentrations below critical level of 2g/kg when herbage assessment is based on requirements set by various authorities (McDowell *et al.*, 1983; 1984; Mtimuni, 1982). Furthermore, all the species studied had concentration levels below 1g/kg DM, the minimum level recommended for growing animals (ARC, 1980). As with previous studies (Jumba *et al.*, 1995; Long *et al.*, 1970), species differences were not marked but *Chloris gayana* (found on 26% of the sites) contained 0.763g/kg DM and could not meet Mg requirements for grazing animals (ARC, 1980). Hill and Guss (1976) and Hacker (1982) reported wide variations in the Mg content of various species and concluded that plant breeding had considerable potential for increasing the mineral composition in a number of forage species. Furthermore, nitrogen level, stage of maturity, excessive potassium level, form of magnesium and amount of readily fermentable carbohydrates have been considered as components of Mg utilization (Rosero *et al.*, 1980). Among the species, *Digitaria swazilandensis* (mean: 0.933g/kg DM) had a potential for development in the Park, however it was poorly spread in the entire Park and could not offset Mg deficiency. Moreover, the problem in this study area could further be confounded by high levels of K in forages. This element is antagonistic to Mg utilization.

Loneragen (1975) reported that Mg uptake is sensitive to temperature, aeration, soil pH and plant species while Gatahi (1986), noted that deficiency problems could be caused due to high levels of

Ca in soils (0.863-4.83g/kg) which could be antagonistic to Mg absorption thereby limiting Mg availability to plants. Therefore, these factors should be taken into account when explaining anticipated deficiency problems in the Park.

Phosphorus

The soil results (mean: 28.9mg/kg) obtained in this study were similar to those reported by several investigators in other parts of the tropics; for example, those reported in Guatemala(mean: 18.9mg/kg, Tejada *et al.*, 1987) and Colombia(mean: 25.4mg/kg, Pastrana *et al.*, 1991a). However, they were lower than those found in USA soils (mean: 67.9mg/kg, Merkel *et al.*, 1990). Herbage P concentrations found in this study (mean: 1.8g/kg DM) were similar to those reported in Colombia (Vargas *et al.*, 1984) and Guatemala (mean: 2.0g/kg DM, Tejada *et al.*, 1987) but lower than those reported by Jumba *et al.* (1995a) for Western Kenya (mean: 1.35g/kg DM).

87% of the sampled forages contained concentrations below the deficiency critical level of 2.5g/kg DM recommended by ARC (1980). Considering species, phosphorus levels were low (mean: 1.93g/kg DM) in the well represented species (*Cynodon dactylon*). *Digitaria swazilandensis* had an advantage over the other species, but it may not offset prevailing deficiency of P in the animals since it poorly spread in the entire Park. Nevertheless, 31% of the sampled forages had concentrations within the minimum recommended allowance (2.1-3.8g/kg DM) for growth.

However for lactating animals, 98% of the forages had concentrations below 3.9g/kg DM, which is minimum level recommended (Suttle, 1983b). The fact that species differences were not marked makes it difficult to suggest control of deficiency by choice of a particular botanical species (Jumba *et al.*, 1995a). Prospects of P deficiency disorders are therefore more likely to occur in wild

herbivore within the Park. However, as with Ca, animals are able to accumulate P in certain tissues during periods of generous dietary supply and mobilise them during periods of shortage(Jumba, 1989). Various investigators(Little, 1980; Ternouth, 1989) have observed that about 80% of the labile P is in the skeleton and that the bone responds to demands of P rather than variations in bone stress including changes in liveweight (Read *et al.*, 1986). This forces the grazing animal to cope with considerable fluctuations in the composition of herbage at various stages of growth.

The physical form of the diet fed to ruminants has also been shown to affect P absorbability and endogenous loss. Ternouth (1989) observed a positive relationship between the amount of P absorbed and lost, and the amount of P ingested, and concluded that requirements increased with dry matter intake, and when the diet contained coarse, long roughage.

The increased demand for P may also be aggravated by variability in herbage availability (Field *et al.*, 1983) due to either P remaining associated with some fraction of the diet and not being released in the gastro-intestinal tract or because the P becomes sequestered with molecules or soil particles as it passes down the tract. Such molecules include complexes of Ca, Al and Fe (Rosa *et al.*, 1982; Valdiva *et al.*, 1982).

Braithwate (1985) provided some evidence that endogenous faecal P is increased as dietary P absorption is increased when P is sequestered within the gastro-intestinal tract. These findings indicate that dietary composition of the interfering elements Ca, Al and Fe needs to be controlled for efficient P utilization, especially in such situations of dietary deficiency of the mineral.

Aluminium

Aluminium level (mean: 0.432g/kg DM) in the diet was higher than that found in Western Kenya forages (mean: 0.3g/kg DM, Jumba, 1989) despite the high pH of soils which tends to limit Al absorption by plants (Sanchez, 1976). Majority of the sampled forages had Al concentrations above 0.5g/kg DM, a level above which Al has been reported to cause metabolic problems in herbivores (Allen,1984). According to species, concentrations were highest in *Chloris gayana* (mean: 0.48g/kg DM) and lowest in *Sporobolus spicatus*(mean: 0.31g/kg DM) although these differences were not significant ($P>0.05$). This shows that particular species could not be regarded as Al accumulators to warrant further investigations.

Furthermore, herbage Al might be less portent than the inorganic salts of Al that were added to synthetic diets for studies on Al toxicity to ruminants (Allen, 1984; Wilkinson *et al.*, 1982). There was a possibility of over-estimation of herbage Al due to unavoidable trace contamination from soil or dust particles (Mitchell, 1960).

Whereas excess Al uptake may affect metabolism (Allen, 1984), small portions are recommended in regions with high fluoride content (McDowell *et al.*, 1993) such as the Lake Nakuru water (mean: 140mg/l, Njenga, 1989). Ruminants are usually more susceptible to F toxicity than are nonruminants (McDowell *et al.*, 1993).

Chronic fluorosis is generally observed under three conditions: 1) Continuous consumption of high F mineral supplements; 2) drinking water high in F (3 to 15μg/l or more); and 3) grazing F contaminated forages. With notable exceptions, the F content of plants is seldom more than 1 to 2mg/kg DM since most plants have a limited capacity to absorb this element (McDowell *et al.*, 1993).

Toxicity of fluoride is determined by the amount and duration of exposure, age of animal, nutritional status, solubility and form of fluorides ingested, stress factors and individual animal differences. For example, incisor teeth may show cavities due to fracture or wear, especially if excess F has been consumed prior to development of teeth. Jaws and long bones develop exostosis and joints may become thickened causing the animal to become stiff and lame with movement becoming difficult and painful. Growth may be subnormal, and weight loses may occur, together with a reduction in fertility and milk production. The impairment of these processes is mostly, but not entirely, secondary to the reduced feed intake brought about by the dental lesions and joint abnormalities, and the consequent inability and unwillingness of the animal to graze and masticate forage. Problems of these kind are usually investigated more exhaustively by culling a selected number of animals and examining the teeth for evidence of fluorosis, particularly for those areas where fluoride concentrations are high; as is the case with the present study area (Njenga, 1989).

Iron and Manganese

The soil Fe results obtained in this study (mean: 141mg/kg) were similar to those obtained in Colombia(mean: 132mg/kg, Vargas *et al.*, 1984) although lower than those recently reported in the same region (mean: 505mg/kg) by Pastrana *et al.*(1991b). However, forage iron (mean: 518mg/kg DM) was similar to that obtained in Southwestern Venezuela (mean: 579mg/kg DM, Rojas *et al.*, 1993) but much higher than that obtained in Western Kenya (mean: 300mg/kg DM, Jumba *et al.*, 1995b), Colombia(mean: 135mg/kg DM, Pastrana *et al.*, 1991b) and Central America(mean: 94.7mg/kg DM, Merkel *et al.*, 1990).

Soil Mn (mean: 408mg/kg) was higher than that obtained in Colombia(mean: 49mg/kg, Pastrana *et al.*, 1991b), Venezuela(mean: 49.5, Rojas *et al.*, 1993) and Central America(mean: 8.7mg/kg, Merkel *et al.*, 1990). Forage Mn (mean: 78.8mg/kg DM) was lower than that obtained in Malawi(mean: 203mg/kg DM, Mtimuni *et al.*, 1990), Western Kenya (mean: 220mg/kg DM, Jumba *et al.*, 1995b) and Colombia(mean: 309mg/kg DM, Pastrana *et al.*, 1991b). However, the level was similar to that obtained in Central America(mean: 66mg/kg DM, Espinoza *et al.*, 1991).

The main problems with these elements are therefore those due to excess intakes. Both elements at concentrations of 0.5g/kg DM and above have been shown to depress appetite and retard growth (Cunningham *et al*, 1966). Evidence of an antagonistic interaction between Mn and Fe has been reported in studies on Mn tolerance of lambs (Hartman *et al.*, 1955) in which higher levels of Mn greatly reduced serum Fe and prevented haemoglobin regeneration in anaemic lambs. Conversely, the high Fe concentrations in the sampled forages may offset any effects of high Mn.

Disagreement on dietary Mn concentrations that should be regarded as the maximum acceptable may be due to the Fe x Mn antagonism.

Chloris gayana and *Cynodon dactylon* which constituted majority of the sampled forages had Fe or Mn levels > 0.5g/kg DM. Most forages had concentration levels 15 times more than the recommended dietary allowance of 30mgFe/kg DM and 40mgMn/kg DM (ARC, 1980; NRC, 1984). Dietary concentrations above 0.4g/kg DM (NRC, 1980) are likely to present toxicity problems such as siderosis (Howell, 1983) or interference in Cu absorption by ruminants (Humphries *et al.*, 1983). Although high levels of Fe have been reported in herbage (Campbell *et al.*, 1974) plant-accumulated iron may also be confounded with soil contamination.

Zinc

Soil zinc contents(mean: 4.23mg/kg) were higher than those obtained in Colombia(mean: 2.8mg/kg, Pastrana *et al.*, 1991b) but lower in Malawi(mean: 7.47mg/kg, Mtimuni *et al.*, 1990). Herbage zinc (mean: 16.8mg/kg DM) was similar to that obtained in Malawi(mean: 17.4mg/kg DM, Mtimuni *et al.*, 1990) but lower than those in Colombia (mean: 20.7mg/kg DM, Pastrana *et al.*, 1991b) and Western Kenya(mean: 23.6mg/kg DM, Jumba, 1989) forages.

Concentrations of Zn were particularly low in the two well represented species (*Chloris gayana* and *Cynodon dactylon*). 94% of the sampled forages had concentration levels below the deficiency critical level of 30mg/kg DM (McDowell and Conrad, 1977) which may be attributed to deficiencies in the soils: the soil data revealed that 84% of the sampled sites were deficient when compared to the critical levels of 30mg/kg (Bahia, 1978).

The lower values obtained in this survey are of particular interest in view of the conflict between the ARC (1980) recommended dietary standards for ruminants (8-35mg/kg DM) and the findings of Mahmoud *et al.*(1983) who observed Zn deficiency in sheep grazing Sudanese pastures which contained 16-27mg/kg DM, and Legg and Sears (1960) who recorded a parakeratesis skin disorder in ruminants grazing forages containing 18-42mg/kg DM in Guyana.

Although zinc deficiency is relatively uncommon in grazing ruminants it has been reported in tropical South America (McDowell *et al.*, 1984; 1993) and even Kenya (Jumba *et al.*, 1995b). Deficiencies are difficult to diagnose and may be manifested by depressed intake, retarded growth, reproductive disorders, loss of hair and wool, the development of scaly skin which cracks readily and delayed healing of wounds (Underwood, 1981). However, there is no obvious relationship between forage concentrations and Zn responsive conditions or onset of clinical abnormalities.

There is no available literature that explains the effect of botanical differences on trace element composition in pastures (Langlands, 1987). Lack of species effects in the present data mean that deficiency problems cannot be corrected by choice of botanical species. Another possible explanation for poor correlation between clinical deficiency and dietary Zn intake is that zinc released by tissue catabolism (associated for example with insufficient intakes of energy or protein) in animals is available to supplement dietary Zn (Masters, 1984). Such interactions highlight one of the problems in assessing the nutrient status from plant and animal tissue analysis. Zinc may also be deficient in the event of high concentrations of interfering (antagonistic) elements such as Ca, Cu, Fe and Cd in the diet (McDowell *et al.*, 1993).

Copper and Molybdenum

With the soil results (mean: 0.784mg/kg) obtained in the study, it can be concluded that the mean Cu values were within the ranges quoted for the forages in problem areas such as Malawi(mean: 0.87mg/kg, Mtimuni *et al.*, 1990), Colombia (0.7mg/kg, Pastrana *et al.*, 1991b) and Southeastern Venezuela (mean: 0.77mg/kg, Rojas *et al.*, 1993). Forage Cu (mean: 8.56mg/kg DM) was similar to that obtained in Malawi (mean: 9.48mg/kg DM, Mtimuni *et al.*, 1990) and Colombia (mean: 7.18mg/kg DM, Pastrana *et al.*, 1991b) but it was higher than that obtained in Western Kenya (mean: 4mg/kg DM, Jumba *et al.*, 1995b) and Central America (mean:4.2mg/kg DM, Merkel *et al.*, 1990).

The herbage Mo content (mean: 4.44mg/kg DM) was higher than that obtained in Western Kenya (mean: 1.1mg/kg DM, Jumba, 1989), Malawi (mean: 0.6mg/kg DM, Mtimuni *et al.*, 1990) and Venezuela (mean: 0.31mg/kg DM, Rojas *et al.*, 1993).

Table 8. Copper: Molybdenum Ratios

Cu:Mo Ratio	% of Forage Samples within
Below 2:1	31
Within 2:1	19
Above 2:1	48

It can be observed that the sampled forages do not meet the Cu requirements of animals even before any antagonisms are taken into account. The quantity of supplementary Cu necessary to maintain a satisfactory Cu status of these animals must be modified according to the Mo and S contents of forages. Since both elements have adverse effects on Cu availability (Suttle, 1974; 1983b). Maltimore and Mason (1971) suggested a Cu:Mo ratio of 2:1 as being safe for ruminants. In the present study 31% of the sampled forages were below this safe ratio (Table 8), increasing fears of Cu deficiency among wildlife in the study area. The Cu concentration range obtained in the species (i.e from 5.49 mg/kg DM in *Digitaria swazilandensis* to 14 mg/kg DM in *Sporobolus spicatus*) indicated prospects of Cu deficiency in the survey area since most of the forages from the two well represented species were below the suggested minimum of 10mg/kg DM (McDowell *et al.*, 1984; 1993).

The effects of botanical composition on any mineral value of forages may be influenced by interactions between other minerals and this is particularly true for Cu. *Digitaria swazilandensis* which had the lowest Cu level (mean: 5.49mg/kg DM) contained the highest Mo value (mean: 5.32mg/kg DM), thus suggesting some anticipated effects of Mo antagonism (Whitelaw *et al.*, 1979). Thus animals grazing *Sporobolus spicatus* have an advantage in Cu availability over those

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grazing *Digitaria swazilandensis* because of its lower Mo content. Secondary or conditioned deficiency may be a problem on 27% of the sites, where forages had > 6mgMo/kg DM.

Copper deficiency usually occurs when forage Mo exceeds 3 or the Cu level falls below 5mg/kg DM (Cunha *et al.*, 1964). Ward (1978) categorized Cu deficiency into four groups according to composition of feed: (i) high levels of Mo (> 20mg/kg DM) and also Cu; (ii) low Cu but significant amounts of Mo (eg., ratio < 2:1); (iii) insufficient Cu (< 5mg/kg DM) and (iv) normal Cu and low Mo, with high levels of soluble protein. The last situation results from increases in the amount of sulphide produced in the rumen from fresh pasture, thus resulting in the formation of Cu sulphide in which Cu is unavailable (Dick *et al.*, 1975). The forages fell in category (ii), having insufficient amounts of Cu and high levels of Mo with Cu:Mo ratios below 2:1.

The geographical distribution of Cu deficiency problems, due to suboptimal herbage Cu content shows wide variations; in Kenya (Jumba *et al.*, 1995b), Australia (Lee, 1951), New Zealand (Cunningham, 1946) and Scotland (Mitchell, 1974), deficiency has been associated with sandy and/or organic soils. In other parts of the tropics (McDowell *et al.*, 1993) the primary cause of Cu deficiency has not been identified: It is possible that some cases may arise from simple soil and herbage deficiency and others from interfering elements such as Cd, Zn and Fe but with the Cu-Mo-S interactions being a dominant feature.

Beeson and Matrone (1976) observed that soils often associated with Cu-Mo problems are alkaline peats, muck soils and other poorly drained soils with high organic matter contents. Thornton (1977), when assessing the occurrence of acute Cu-Mo problems on molybdeniferous clays in England and Wales, and Kubota (1977) in USA mention the additional criteria of high soil

Mo content (> 20mg/kg) and neutral to alkaline soil pH. The study area contained poorly drained, alkaline soils more likely to cause Cu-Mo-S interaction problems. This was in agreement with results of Maskall and Thornton (1991), who found high levels of Mo (range: 0.45-6.35μg/g) in LNNP soils with very low Cu (range: 1.20-13.80mg/kg) content.

Cobalt

Soil Co (mean: 1.03mg/kg) was higher than that obtained in Colombia (mean: 0.04mg/kg, Pastrana *et al.*, 1991b) and Malawi (mean: 0.05mg/kg, Mtimuni *et al.*, 1990). Forage Co (mean: 0.573mg/kg DM) was higher than the typical levels obtained in Western Kenya (mean: 0.22mg/kg DM, Jumba *et al.*, 1995b), however, it was comparatively higher than that found in Colombian forages (mean: 0.4mg/kg DM, Vargas *et al.*, 1984) and Malawi (mean: 0.48mg/kg DM, Mtimuni *et al.*, 1990).

Cobalt deficiency commonly occurs in ruminants (Underwood, 1971) and responds completely to Co-containing anti-pernicious anaemia factor, vitamin B_{12}. The severity of deficiency varies with the animal species. Sheep require higher dietary levels, due partly to the low efficiency of absorption of the vitamin (Smith and Martson, 1970a). Severe deficiency in sheep leads to severe and progressive loss of appetite and ultimately to death from inanition. In less affected animals there is anaemia and anorexia, leading to depressed productivity and unthriftness. Young animals are particularly susceptible and symptoms appear when reserves are exhausted; not all animals within a herd or flock are unaffected. Pasture Co levels below 0.1mg/kg DM will likely produce a deficiency in lambs and calves but consistent intake of feeds containing

0.07mg/kg DM of Co and below are associated with widespread deficiency (McDowell *et al.*, 1993).

The forage species sampled in this study showed extreme mean differences although they were insignificant. *Cynodon dactylon* had the highest mean (0.63mg/kg DM), whereas *Chloris gayana* had the lowest level (0.448).

However, all the species had concentrations above the ARC (1980) requirements (0.08-0.11mg/kg DM). The high herbage concentrations (when compared with animal requirements) suggest that Co may not be a limiting mineral in the Park. This is in agreement with earlier reports by Maskall and Thornton (1991) who reported high total Co levels in soils (range: 2.40-13.20mg/kg) and forage (range: 0.3-1.92mg/kg). These observations are contrary to earlier reports by Horward (1970) and Russell and Duncan (1956) who attributed cattle health disorders to Co deficiency in a number of farms in Nakuru district. For LNNP, a lower incidence of Co dificiency disorders in wildlife animals is expected since additional Co may be obtained from soil through soil ingestion (McDonald and Suttle, 1986).

CONCLUSIONS

1) There were no forage mineral species differences in Lake Nakuru National Park except for K, where *Sporobolus spicatus* (with the highest mean) was significantly different (P>0.05) from the three other species.

2) The eastern side of the Park contained high levels of Pb and Cd in both soil and forage. Animals residing in this sector are likely to suffer from toxicity of these pollutants.

3) Significantly (P>0.05) higher levels of Ca, Co, Mn, Mg, Zn and Cu and pH were detected in the eastern side of the Park.

4) Comparison with critical values below which availability and deficiency problems are known to occur indicated that Mg, P, Cu and Zn were deficient in soils, whereas Ca, P, Mg, Cu and Zn were limiting in forages.

5) Except for Mg, all the soil macrominerals (Ca, K and P) and pH decreased with increasing distance away from the lake. However, the converse was true for the trace elements Co, Cu, Mn, Zn and Fe as well as Cd and Pb. Animal species having high residence times closer to the lake should be investigated for the status of these elements in view of deficiency (Cu, Mn, Zn, Ca, P, K) and toxicity (Cd & Pb) problems anticipated.

6) Cobalt which had for long been reported a deficiency problem in Nakuru district is according to the present findings, not a problem in the Park.

RECOMMENDATIONS

It is suggested that future investigations by the Kenya Wildlife Service (KWS) should involve culling some animal species (such as waterbuck and impala) of the same age (at least 6) from each region to obtain the following samples:

1) Sculls
2) Rib or Femur bone
3) Teeth
4) Urine
5) Blood.

These specimens should be assessed as follows:

(a) Teeth for evidence of fluorosis attributed to high fluoride intake from the lake water, and soil ingestion to confirm abrasion and wear.

(b) Bone for ash, Ca, P, F, Cu, Mg, Pb and Cd.

(c) Sculls for any abnormal development.

(d) Urine for cortisol and Mg analysis.

(e) Blood (serum) for Cu and Zn.

Results should be compared with those obtained for other animals in neighbouring Elementeita and Naivasha areas as well as distant sites such as Lewa Downs. If the data confirm the predictions made from the soil and forage analysis, then supplementation of the limiting elements should follow to check performance and improvement in health. In this way constraints on animal health can be alleviated. At the time of submitting this thesis, the KWS had already started taking action on the above recommendations, a step which is considered commendable in the interest of animal health in the Park.

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APPENDIX

Appendix I. Soil pH and Macromineral Composition*

Sample No.	Na (g)	K (g)	Ca (g)	Mg (mg)	P (mg)	pH
1	5.09	3.53	4.06	38.8	22.8	9.30
2	2.68	1.80	3.14	10.5	13.6	9.75
3	0.299	0.906	1.63	233	13.0	7.09
4	3.38	3.02	2.08	6.53	12.9	9.63
5	0.295	0.695	3.39	309	17.2	7.10
6	0.606	2.35	1.80	257	1210	6.84
7	2.05	4.44	4.81	334	35.1	7.98
8	4.81	2.93	2.82	11.1	22.7	9.69
9	4.52	3.60	3.72	40.3	36.3	8.28
10	0.577	1.56	3.53	405	66.2	7.38
11	3.03	2.85	3.36	16.8	40.6	8.83
12	1.94	2.45	4.59	188	31.7	7.84
13	0.866	2.23	4.11	258	26.9	6.97
14	2.87	3.08	4.11	166	21.7	7.88
15	0.296	0.896	1.55	202	27.2	6.76
16	0.587	1.68	2.84	316	54.6	7.21
17	0.489	1.48	2.46	348	145	7.14
18	3.90	2.76	3.32	37.9	22.4	8.74
19	5.27	2.91	1.83	18.1	48.2	9.44
20	0.294	0.495	3.30	371	22.6	6.95
21	0.498	1.11	1.75	208	44.6	7.25
22	5.01	3.57	2.94	115	27.4	8.23
23	3.72	1.98	3.38	21.7	40.4	9.77
24	0.198	-	4.85	124	45.8	8.18

Sample No.	Na (g)	K (g)	Ca (g)	Mg (mg)	P (mg)	pH
25	1.48	1.39	1.14	14.0	9.12	8.57
26	2.60	1.41	1.30	14.6	32.2	10.2
27	0.793	1.30	1.22	125	18.4	8.27
28	0.396	1.20	1.14	62.0	23.1	7.09
29	1.19	1.81	2.35	61.8	27.6	7.42
30	1.39	2.40	4.57	44.5	22.7	6.86
31	4.23	2.38	2.62	17.4	13.8	7.33
32	1.08	2.29	4.55	88.6	36.3	7.31
33	2.27	2.00	3.48	77.4	31.8	7.77
34	0.792	2.00	3.75	60.0	22.9	6.89
35	2.16	2.88	2.09	54.6	31.7	7.20
36	1.08	1.59	2.27	18.9	27.5	6.50
37	0.786	1.19	1.40	30.7	27.2	3.97
38	1.57	1.29	0.936	8.41	18.1	9.53
39	1.19	1.70	2.57	30.0	22.7	6.79
40	0.387	0.587	1.10	34.7	22.7	5.72
41	0.490	0.989	0.863	55.4	27.3	5.51
42	0.492	1.19	1.35	30.3	22.8	5.50
43	1.07	2.36	2.49	113	77.4	7.51
44	4.52	2.58	2.50	23.8	22.3	7.38

* All expressed on " per Kg dry weight basis"

Appendix II. Soil Trace Elements Composition (mg/kg)

Sample No.	Fe	Mn	Cu	Co	Zn	Pb	Cd
1	61.6	374	0.886	1.31	3.52	6.07	0.13
2	83.3	234	0.382	1.15	3.19	4.34	0.06
3	250	428	0.312	1.37	4.38	7.21	0.14
4	51.4	280	0.195	1.54	1.48	5.23	0.12
5	370	1141	0.899	1.90	14.9	9.57	0.33
6	305	413	0.763	1.54	7.05	6.85	0.10
7	159	782	1.14	1.39	9.72	6.54	0.27
8	129	315	1.31	1.75	3.65	7.05	0.05
9	97.0	469	1.45	1.56	2.33	7.63	0.02
10	238	595	1.51	1.25	17.8	7.92	0.33
11	55.3	179	0.917	1.31	1.57	5.83	0.08
12	81.8	663	2.28	0.874	5.41	4.57	0.10
13	178	802	1.37	1.76	4.46	7.94	0.19
14	122	902	1.04	1.47	4.00	6.87	0.17
15	218	165	1.01	0.720	6.39	6.04	0.06
16	290	583	1.25	1.26	9.23	7.45	0.18
17	395	467	2.09	0.916	18.3	8.75	0.24
18	47.4	680	1.16	0.720	2.05	4.20	0.14
19	69.6	148	0.788	1.21	1.42	6.56	0.08
20	369	982	1.47	1.50	8.05	8.07	0.30
21	224	606	1.12	1.39	7.45	7.33	0.23
22	132	771	1.47	1.40	3.39	8.43	0.20
23	107	493	0.601	0.634	2.44	3.04	0.17
24	95.7	20.0	1.76	14.8	0.34	0.30	0.04
25	42.4	212	0.429	0.881	1.40	4.14	0.02

Sample No.	Fe	Mn	Cu	Co	Zn	Pb	Cd
26	70.6	91.5	0.350	0.619	0.95	2.96	-
27	45.7	115	0.225	0.584	0.58	3.27	0.01
28	78.7	140	0.190	0.644	1.04	3.11	-
29	180	568	0.425	0.937	2.86	7.81	0.21
30	205	644	0.457	1.12	3.13	8.48	0.28
31	161	569	0.455	0.868	3.26	7.99	0.22
32	176	394	0.658	0.623	4.77	8.06	0.21
33	123	264	0.428	0.609	1.39	6.00	0.12
34	137	410	0.284	0.879	1.00	6.58	0.07
35	106	326	0.640	0.740	1.75	7.59	0.18
36	34.7	92.5	0.215	0.459	0.60	3.98	0.06
37	41.1	102	0.438	0.584	0.59	4.07	0.07
38	21.0	39.0	0.225	0.474	0.22	2.44	0.07
39	91.7	333	0.274	0.845	0.90	5.49	0.11
40	51.4	84.2	0.396	0.534	0.59	2.73	0.06
41	88.5	81.5	0.170	0.514	1.09	3.45	0.09
42	130	348	0.274	0.858	1.09	5.43	0.15
43	206	322	1.26	0.934	12.2	7.89	0.17
44	124	334	0.910	0.978	4.40	6.62	0.17

Appendix III. Forage Macromineral Composition (g/kg DM)

Sample No.	Na	K	Ca	Mg	Al	P
1	1.44	5.12	3.07	0.821	0.280	1.91
2	14.5	7.53	2.18	1.13	0.177	0.794
3	1.84	6.43	3.31	0.977	0.251	2.81
4	8.00	6.46	8.57	0.822	1.12	1.69
5	5.68	2.67	1.09	0.305	0.033	0.243
6	0.662	12.7	2.26	1.01	0.471	1.89
7	1.26	5.83	3.69	0.558	0.248	1.44
8	0.412	7.65	1.77	0.429	0.205	0.590
9	0.599	13.1	2.38	0.857	0.443	2.35
10	0.439	7.23	3.57	0.972	0.731	2.01
11	0.630	6.15	1.81	0.617	0.241	1.11
12	5.78	12.0	2.31	1.47	0.252	1.39
13	1.25	7.42	4.14	1.11	0.548	2.22
14	1.79	5.94	2.66	0.691	0.367	2.29
15	1.50	13.2	3.99	1.41	0.543	3.34
16	1.94	13.2	10.9	1.29	0.209	1.27
17	0.073	7.08	3.77	1.12	0.657	3.03
18	2.20	11.3	14.8	1.72	0.237	1.77
19	10.8	7.00	5.12	1.42	0.133	1.03
20	0.151	4.89	2.63	1.37	0.650	3.23
21	0.559	4.82	2.95	0.770	0.207	2.05
22	0.855	5.53	2.20	0.860	0.326	0.786
23	0.879	4.25	3.21	0.520	0.207	4.89
24	0.054	7.63	2.40	0.965	0.298	1.29
25	0.114	7.95	2.41	0.636	0.443	1.80

Sample No.	Na	K	Ca	Mg	Al	P
26	0.137	4.55	2.68	0.761	0.413	2.30
27	1.19	6.19	4.84	0.889	0.347	1.69
28	0.528	6.09	8.08	0.844	0.721	2.28
29	1.03	2.85	3.99	0.593	0.175	0.936
30	0.977	3.95	2.64	0.723	0.306	0.982
31	0.257	6.76	6.78	2.02	0.846	1.55
32	0.350	5.90	3.97	1.13	0.522	2.53
33	2.34	8.20	2.19	0.775	0.251	0.996
34	4.70	6.52	4.81	1.32	0.419	1.59
35	2.08	5.08	4.81	0.831	0.206	0.953
36	1.12	6.77	3.01	0.558	0.465	2.16
37	0.819	7.59	2.29	0.410	0.189	2.25
38	4.27	5.00	3.69	0.405	0.547	1.33
39	0.523	12.6	2.03	0.512	0.237	2.04
40	0.279	14.4	1.88	0.495	0.104	2.44
41	0.261	7.01	2.82	0.660	1.20	1.22
42	0.811	12.5	2.89	0.556	0.747	1.53
43	0.591	13.5	10.7	2.66	1.64	2.73
44	0.274	4.30	3.18	0.572	0.907	1.13
45	0.694	12.9	2.57	0.754	0.773	1.71
46	0.548	14.6	7.33	0.935	0.267	1.71
47	0.265	7.44	7.10	2.52	0.902	1.57
48	0.220	6.70	3.53	1.02	0.667	1.89
49	0.150	6.69	1.72	0.881	0.747	1.95
50	0.429	12.8	2.12	0.921	0.242	1.58
51	0.162	6.70	12.9	1.32	1.19	1.26
52	1.06	9.26	1.96	0.524	0.653	1.51
53	3.09	5.15	1.67	1.44	0.715	1.44

Sample No.	Na	K	Ca	Mg	Al	P
54	2.71	4.72	4.55	0.590	0.398	0.985
55	1.04	7.36	1.97	0.307	0.310	1.39
56	0.403	8.03	2.62	0.489	0.354	1.95
57	3.09	5.75	1.98	1.08	0.285	1.11
58	0.346	5.48	1.54	0.664	0.368	1.04
59	0.272	7.93	2.42	0.869	0.686	2.13
60	0.466	8.05	2.78	0.560	0.662	1.49
61	0.257	4.86	1.90	0.149	0.579	0.424
62	4.15	26.1	9.36	1.42	0.482	3.96
63	1.77	9.38	2.64	1.50	1.00	3.33
64	1.43	6.07	2.42	1.14	0.503	2.04
65	3.30	16.5	9.38	1.29	0.431	2.36
66	1.79	4.94	3.64	1.03	0.566	2.00
67	2.24	13.4	6.60	1.11	0.453	1.67

Appendix IV. Forage Trace Elements Composition(mg/kg DM)

Spl.No	Co	Fe	Mn	Zn	Mo	Cu	Cd	Pb
1	0.658	460	69.7	16.6	0.88	43.0	0.118	2.63
2	0.624	152	294	7.88	1.83	-	0.486	0.62
3	0.223	293	74.3	14.2	2.35	4.21	0.040	0.66
4	0.204	1660	139	32.9	23.7	8.61	0.183	0.61
5	0.226	30	118	2.20	0.37	9.04	0.244	1.35
6	0.637	491	64.7	20.1	3.29	5.44	0.267	1.27
7	0.887	255	126	13.6	3.26	14.9	0.239	1.33
8	0.202	266	39.4	36.5	4.81	15.7	0.182	0.61
9	0.431	400	47.0	17.5	7.18	5.55	0.077	0.02
10	0.617	887	125	38.8	7.41	3.61	0.182	0.61
11	0.437	682	37.3	15.4	0.66	2.55	0.118	0.65
12	0.450	287	139	14.5	15.3	6.82	0.283	0.67
13	0.405	377	50.7	25.0	3.46	10.7	0.109	0.61
14	0.813	300	62.5	18.3	10.2	7.43	0.146	1.22
15	0.505	357	58.2	24.0	6.27	9.17	0.181	0.76
16	1.62	208	51.6	18.2	15.0	23.2	0.436	1.21
17	0.604	237	80.7	38.2	5.56	12.6	0.145	0.60
18	0.402	103	109	11.2	22.8	16.7	0.217	1.20
19	0.405	627	200	5.55	3.81	2.13	0.364	0.02
20	0.808	157	78.3	26.0	3.67	7.48	0.218	1.21
21	0.202	277	37.2	8.37	1.32	20.9	0.073	1.21
22	0.202	207	74.5	13.2	2.43	10.2	0.073	0.02
23	0.203	257	22.9	7.15	2.50	7.80	0.146	1.21
24	0.404	391	53.6	24.3	2.52	5.41	0.218	1.21
25	0.403	424	48.5	40.0	4.84	6.29	0.217	1.21

Spl.No	Co	Fe	Mn	Zn	Mo	Cu	Cd	Pb
26	0.404	403	47.5	27.2	14.2	2.77	0.073	1.21
27	0.404	830	67.3	14.3	8.87	5.95	0.145	1.21
28	0.445	341	60.2	23.9	14.7	34.1	0.400	2.66
29	0.405	459	89.3	10.1	1.74	4.32	0.145	1.21
30	0.809	888	40.1	16.9	4.49	24.3	0.145	2.42
31	0.842	822	111	46.0	2.33	8.22	0.302	1.26
32	0.835	245	70.0	28.3	3.26	11.5	0.225	0.02
33	0.406	598	88.7	12.7	3.52	15.7	0.146	0.02
34	0.836	237	185	9.19	2.97	12.3	0.075	1.25
35	0.881	404	128	8.90	0.73	8.16	0.198	0.66
36	0.791	266	91.2	7.47	5.05	2.88	0.213	0.02
37	0.203	837	68.7	10.5	1.56	1.85	0.073	0.02
38	0.812	250	144	4.66	17.4	2.68	0.146	0.02
39	0.813	147	43.2	7.74	3.71	3.07	0.146	0.02
40	0.406	1361	33.4	7.97	1.86	8.21	0.146	0.02
41	0.809	943	123	22.3	0.93	3.72	0.073	0.02
42	0.807	1698	106	12.2	4.06	3.34	0.072	0.02
43	0.810	1077	134	40.1	4.76	11.8	0.437	0.02
44	0.411	822	80.5	22.5	1.96	4.61	0.074	0.02
45	0.401	449	87.7	16.6	7.05	19.1	0.072	0.96
46	0.813	1048	34.3	15.9	5.84	8.16	0.219	0.02
47	0.791	517	107	31.2	5.86	6.83	0.355	1.18
48	0.202	237	84.5	22.1	7.50	9.15	0.145	1.21
49	0.810	386	84.1	18.2	4.20	11.0	0.073	1.21
50	0.404	1439	82.7	9.12	2.26	24.9	0.145	2.42
51	0.810	781	158	25.2	2.76	10.8	0.218	1.21
52	1.21	572	107	12.5	5.14	3.83	0.218	0.02
53	2.03	530	98.4	11.3	13.2	8.57	0.292	1.22

Spl.No	Co	Fe	Mn	Zn	Mo	Cu	Cd	Pb
54	0.809	434	103	4.61	7.74	27.1	0.218	1.21
55	0.809	424	71.3	7.08	2.95	5.50	0.073	0.02
56	0.403	278	75.3	7.88	2.89	8.22	0.004	0.02
57	0.809	368	48.8	8.26	8.54	9.22	0.291	0.02
58	0.202	881	60.2	13.7	3.21	2.32	0.145	0.02
59	0.809	747	102	14.9	5.30	8.30	0.218	1.21
60	0.809	283	73.5	13.3	4.57	2.86	0.145	1.21
61	1.61	551	53.8	5.48	2.57	1.79	0.218	1.20
62	0.818	1132	163	55.8	2.73	20.7	0.515	1.2
63	0.809	442	70.3	27.4	1.00	10.9	0.218	0.02
64	0.405	265	44.2	13.2	2.76	3.13	0.073	0.02
65	1.61	479	113	20.7	14.5	12.8	0.433	1.21
66	0.203	521	64.8	16.5	2.62	4.60	0.004	0.02
67	1.21	267	165	21.2	23.3	15.8	0.290	0.02