

**ASSESSMENT OF LEVELS OF SELECTED NUTRIENTS IN
THE SOIL FOR WHEAT GROWING IN NAROK NORTH
SUB-COUNTY, KENYA**

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**Assesment of levels of selected nutrients in the soil for wheat growing in
Narok north sub-county, Kenya**

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DECLARATION

This thesis is my original work and has not been presented for a degree in any other University.

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DEDICATION

I would like to dedicate this work to my beloved parents Benson and Rosemary Mesoppirr, my beloved husband Roimen, my dear son Ritei, my lovely daughter Teyian and my beloved siblings Senewa, Sankei, Mantai and Reney. Love you all so much.

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LIST OF ABBREVIATIONS AND ACRONYMS

B	Butt
EPZA	Export Processing Zones Authority
FAAS	Flame Atomic Absorption Spectrophotometer
FAO	Food and Agriculture Organization
G	Gas
JKUAT	Jomo Kenyatta University of Agriculture and Technology
KPMG	Kenya Partnership Membership Group
KNBS	Kenya National Bureau of Statistics
MMU	Maasai Mara University
MR	Methyl red
NADH	Nicotinamide Adenine Dinucleotide
NDP	National Development Plan
OECD	Organization for Economic Co-operation and Development
SIM	Sulfur Indole Motility
SL	Slant
TSI	Triple Sugar Iron
TOM	Total Organic Matter
UV-VIS	Ultraviolet- Visible
VP	Voges pasteur

ABSTRACT

Wheat (*Triticum aestivum*) is the most important cereal after maize in Kenya. Its production is lower than its consumption in Kenya and the shortfall is met by imports. The current study analyzed macronutrients, micronutrients, pH, total organic matter and determined diversity of bacteria in the soil wheat growing areas of Narok north sub-county. Stratified random sampling method was used and soil samples collected from Ololulunga, Ntulele and Mau. The standards were prepared using chemicals and reagents of analytical grade. Atomic Absorption spectrophotometer was used to analyze Mg, Ca, Cu, Zn and Fe, UV-VIS spectrophotometer was used in determination of nitrates, sulfates and phosphates and Flame photometry was used to analyze K. Bacteria were isolated and both morphological and biochemical test were done on them by use of different selective media. The data collected was analyzed using student t-test, MSTAT-C and SPSS. Ca and Zn had the strongest positive correlation of 0.642 followed by Mg and Ca with a positive correlation of 0.513 while the rest of the nutrients had very low correlations before planting. Ca and Mg showed a positive correlation of 0.413 after planting whereas phosphates and calcium, phosphates and zinc showed a negative correlation of -0.454 and -0.455 respectively. The bacteria isolated in this study were identified using the Bergey's manual and were found to be *Pseudomonas spp*, *Micrococcus spp*, *Mycobacterium spp*, *Corynebacterium spp* and *Bacillus spp*. It was found that in most of the sampled farms, K, Ca, Mg, Cu and Zn were deficient but Fe was sufficient. All the farms sampled were found to be nitrates sufficient but most farms were deficient in sulfates and phosphates therefore addition of these fertilizers is highly recommended.

CHAPTER ONE

INTRODUCTION AND LITERATURE REVIEW

1.1 Background of the study

1.1.1 Wheat

Wheat is a cereal grass of the Gramineae (Poaceae) family and of the genus *Triticum*. Its edible grain is the world's largest cereal-grass crop (M'mbonyi *et al.*, 2010). It is grown in most parts of the world, from near-arctic to near-equatorial latitudes. It is the most important crop among the cereals in terms of acreage planted and is followed in importance by corn, barley and sorghum (FAO, 2009). Wheat production is widespread throughout the world and supplies much of the world's dietary protein and about 20% of the calories consumed by humans (OECD-FAO, 2009, FAO, 2006). The European countries achieve highest yields, while the countries in Asia tend to obtain yields slightly below the world average (FAO, 2009). The United States, the EU-27, Canada, Australia and Argentina have been traditionally the most important wheat exporters in the world (FAO, 2009), but recently Kazakhstan, the Russian Federation and Ukraine (often referred to as the Black Sea grain exporters) have become the leading wheat exporters and will likely hold their dominant position in wheat trade in the foreseeable future (OECD- FAO, 2009).

There are two main cultivated wheat species in the world: *T. aestivum* or bread wheat and *T. turgidum var. durum* or durum wheat. The bread wheat constitutes roughly 95% of the cultivated crop and is used for making bread, cookies and pastries. Durum wheat on the other hand contributes the remaining 5% and is used for making pasta and other semolina products (Dubcovsky & Dvorak, 2007).

1.1.2 Wheat growing in Africa

Wheat grown in South Africa, Ethiopia, Sudan and Kenya occupy 2.5 million ha and makes up 94 percent of the total area of wheat planted in East and Southern Africa. Tanzania, Zimbabwe and Zambia collectively plant 105 000 ha, with a minor amount being grown in Angola, Mozambique and Somalia (CIMMYT, 1996). Consumption varies from 61 kg/caput in South Africa to 5 kg/caput in Tanzania. Yield average for the region in the 1993-1995 period was 1.6 tonnes/ha, with high yields of 4.9 tonnes/ha for Zimbabwe and 3.1 tonnes/ha for Zambia and a very low yield of 0.4 tonnes/ha for Somalia (CIMMYT, 1996).

Most of the wheat in East Africa is grown at high elevations of above 1,500 m but along the equator, the elevation for wheat is 3,000 m or more. Wheat is generally produced under rain fed conditions, except in the lowlands of Somalia, Zambia and Zimbabwe where irrigation is practiced. In the highlands, the average rainfall is between 600 and 700 mm and usually falls from June to September. Soils are generally low in nitrogen (N) and phosphorus (P) and some minor elements. Wheat is usually planted from the months of May to July and harvested in September or October. Common wheat (bread wheat) is grown in most of East Africa, except in Ethiopia where durum wheat is grown on 60 to 70 percent of the area (FAO, 2009). Kenya's economy largely depends on the agricultural sector, which accounted for 24% of the GDP in 2011 (KPMG, 2012). The agriculture sector recorded a lower growth of 1.5% in 2011 compared to 6.4% in 2010. This was attributed to the decline of production of all major crops except rice, cotton, pyrethrum and sisal. Wheat production in 2010 was 199,700 tonnes and decreased to 105,900 tonnes in the year 2011 which is a negative (-47%) change (KNBS, 2012).

Wheat is the second most important cereal grain in Kenya, after maize (Nyangito *et. al*, 2002; EPZA, 2005; NDP, 2003). The crop is grown largely for commercial purposes on a large scale. Kenya is self-sufficient in the hard variety of wheat which is high in protein and suitable for yeast bread, but is a net importer of the softer variety which low

in gluten and suitable for tender baked goods e.g. biscuits (M'mbonyi *et al.*, (2010). Wheat growing areas in Kenya include the scenic Rift Valley regions of Uasin Gishu, Narok, Elgeyo-Marakwet, Londiani, Molo, Nakuru and Timau areas. These areas have altitudes ranging between 1200m and 1,500m above sea level, with annual rainfall varying between 800 mm and 2,000 mm, with up to 2,500 mm on higher grounds. The area under wheat production in Kenya increased from 144,000 ha in 2002 to 150,000 ha in 2003 (EPZA, 2005).

Cereal grains are the most important dietary source of micronutrients in many developing countries. Micronutrient concentrations and bioavailability in cereal grain is generally low. Increasing the micronutrient concentration of cereal grains has been identified as a way of addressing human micronutrient deficiencies (Manthey *et al.*, 1994; Muminjanov *et al.*, 2007; Pahlavan-Rad & Pessarakli, 2009).

1.2 Previous studies

1.2.1 Plant nutrients

Plant nutrients fall under two categories: macro nutrients and micro nutrients.

Macro nutrients are those nutrients that are needed in relatively large amounts and they include: nitrogen, calcium, phosphorous, potassium, sulfur and magnesium (Shanyn & Bradley, 1999). Micro nutrients are those nutrients that plants require in small amounts. They include; zinc, copper, manganese, iron and boron (Shanyn & Bradley, 1999).

Plant nutrients are essential for producing sufficient and healthy food for the world's rapidly growing population. Plant nutrients are therefore a vital component of any system of sustainable agriculture (FAO, 1998). Iron (Fe), zinc (Zn), copper (Cu), and manganese (Mn) are essential micronutrients for plants and humans (Kaya *et al.*, 1999; Asad & Rafique, 2000; Hao *et al.*, 2007). A deficiency of just one of these nutrients can greatly reduce plant yield and even cause plant death. Micronutrient deficiency,

especially Fe and Zn deficiency, is widespread in humans (Graham *et al.*, 1999; Stoltzfus, 2001; FAO, 2002; Liu *et al.*, 2006; Hao *et al.*, 2007).

Crop management strategies are an important complement to ongoing breeding programs. Studies have shown that Fe, Zn, Cu, and Mn concentrations in rice or wheat grain can be increased by proper irrigation management, Nitrogen fertilization, and late planting (Hao *et al.*, 2007; Pearson *et al.*, 2008). The growth and yield of a plant (wheat included) is determined by the availability of some specific mineral nutrients that are absolutely essential for the completion of their life cycle (Marshner, 1995).

Application of fertilizers containing these essential nutrients is very important for intensive agriculture. Fertilizers are mineral or organic substances, natural or manufactured, that are applied to the soil, irrigation water or hydroponic medium, to supply plants with nutrients (FAO, 1998). Essential nutrients are nutrients required by plants for the completion of their life cycle, must be directly involved in plant metabolism and must not be replaceable by another element (Barack, 1999).

1.2.1.1 Nitrogen

Nitrogen is an essential constituent of plant proteins (chlorophyll, growth hormones, enzymes and comprises a significant portion of the protoplasm. Nitrogen in the soil occurs in different forms, both inorganic and organic. Organic nitrogen mostly is from plant and microbial remains. Inorganic soil nitrogen is composed of ammonium, nitrate and nitrite forms. The form of nitrogen taken up by plants is mainly the ammonium-N and the nitrate- N (Salisbury & Ross, 1992). Nitrate N is the major form taken up by plants and the critical level for nitrates is 15mg/Kg as stated by Dennis and John (2003).

Early nitrogen application promotes tillering and leaf growth, whereas late application prolongs leaf duration and expansion (Spiertz & Hole, 1984). The amount of nitrogen applied also affects the pattern of nitrogen uptake by crops (Sylvester *et al.*, 1990).

Nitrogen deficient plants will mature early and crop quality and yield are often reduced (Jones, 1998). On the other hand plants with excess nitrogen turn deep in green color and have delayed maturity. Due to nitrogen being involved in vegetative growth, excess nitrogen results in tall plants with weak stems, possibly causing lodging to occur. New growth will be succulent and plant transpiration high (low water use efficiency) (Jacobsen and Jasper, 1991). Wheat plants with nitrogen deficiency have reduced plant height and produce fewer tillers than healthy plants, and they also turn yellow in color (Gene *et al.*, 2002).

1.2.1.2 Phosphorus

Phosphorus (P) is a naturally occurring element in the environment that can be found in all living organisms as well as in water and soils. It is an essential component for many physiological processes related to proper energy utilization in both plants and animals. (Mike *et al.*, 2011). Phosphorus is an essential nutrient both as a part of several key plant structure compounds and as a catalyst in the conversion of numerous key biochemical reactions in plants (Bill, 2011). Soil P exists in inorganic and organic forms. Each form is a continuum of many P compounds, existing in equilibrium with each other and ranging from solution P (taken up by plants) to very stable or unavailable compounds (the most typical). In most soils, 50% to 75% of the P is inorganic (Swasiager, 2011).

Plants uptake phosphorous from soil mostly in the orthophosphate form. Native soil P levels are often low enough to limit crop production. Both inorganic P fertilizers (treated rock phosphate) and organic P sources (animal manures) are equally adept at supplying the orthophosphate ion and correcting P deficiencies in soil. Most of the P in animal manure is in an organic form and must be converted to plant-available forms via soil biological activity, a process known as mineralization. The net effect of this characteristic is that P derived from animal manure may act more like a slow-release fertilizer than commercial inorganic fertilizers, which are more soluble and readily available to plants. (Mike *et al.*, 2011). Phosphorus contained in plant material is

recycled to the soil when the crops rot away in the farm. Mild phosphorus deficiency in wheat can be characterized by stunted growth with no distinct leaf symptoms, but in severe cases phosphorus deficient plants become purple or brown (Gene *et al.*, 2002).

Wheat requires a large amount of readily available P, when the root system is feeding primarily from the upper soil surface. To compensate for the higher P requirements of wheat, it is suggested that 1.5 times the amount of expected P removal be applied prior to seeding these crops (Fernandez & Hoefl, 2012). Availability of phosphorus (P) for plant utilization is not a function of its concentration in the soil, but rather on the rate of its release from the soil surface into the soil solution. Phosphorus is considered the most unavailable and inaccessible of all mineral nutrients (Holford, 1997). For wheat production 40-45lb/A (20-28 mg/kg) concentration of P in the soil is adequate (Fernandez & Hoefl, 2012).

1.2.1.3 Potassium

Potassium (K) is one of essential nutrients required for plant growth and reproduction (Shanyn & Bradley, 1999). It is also important for photosynthesis, it adds stalk and stem stiffness, it makes the crop resistance to drought and diseases and gives plumpness to grain seed (Tucker, 1999). It is taken up by plants in its ionic form (K^+) (Shanyn & Bradley, 1999). The deficiency of potassium in wheat is characterized by sharp difference in plant size, length, smaller kernels and condition of roots. Only in advanced stages does withering or burn of leaf tips and margins occur. (Mosaic, 2011). For most crops a K concentration in the soil of around 360-400lb/A (180-200 mg/Kg) is required but for wheat production a minimum concentration of as low as 100lb/A (50 mg/Kg) in the soil is adequate (Farnandez & Hoefl, 2012).

1.2.1.4 Calcium

Calcium is a component of plant cell walls and regulates cell wall construction (McCauley *et al.*, 2009). Calcium is absorbed from the soil in form of divalent Ca^{2+} . Insufficient calcium can cause young leaves to become distorted and turn abnormally dark green. Leaf tips often become dry or brittle and will eventually wither and die. Stems are weak and germination is poor (McCauley *et al.*, 2009). A concentration of at least 800lb (200 mg/Kg) of Ca in the soil is adequate for wheat production (Farnandez & Hoef, 2012).

1.2.1.5 Sulfur

Sulfur is an essential constituent of certain amino acids and proteins. It is absorbed from the soil as divalent sulphate anions (SO_4^{2-}) (Saliburry & Ross, 1992). Sulfur deficiency results in the inhibition of protein and chlorophyll synthesis. Sulfur deficiency symptoms can be difficult to diagnose as effects can resemble symptoms of nitrogen and molybdenum deficiencies (McCauley *et al.*, 2009). When the concentration of sulfur in the soil is 0-12lb/A (0-6 mg/Kg), it is considered to be very low whereas 12-22lb/A (6-11 mg/Kg) is considered adequate (Farnandez & Hoef, 2012).

1.2.1.6 Magnesium

Magnesium is the central molecule in chlorophyll and is an important cofactor for the production of adenosine triphosphate (ATP). It is absorbed from the soil as divalent Mg^{2+} (Saliburry & Ross, 1992). Magnesium deficiency in wheat leads to distinct mottling as yellowish-green patches will occur on the leaves (McCauley *et al.*, 2009). A concentration of 150-200lb/A (75-100 mg/Kg) of Mg in the soil is adequate for crop production (Farnandez & Hoef, 2012).

1.2.1.7 Zinc

Zinc is needed by plants for growth hormone production and is particularly important for internodes elongation. Zinc has intermediate mobility in the plant and symptoms will initially show up in middle leaves (McCauley *et al.*, 2009). Zinc deficiency in soils has been recognized as a worldwide nutritional constraint (Cakmak *et al.*, 1998), and is probably the most widespread micronutrient deficiency in cereals (Graham *et al.*, 1992). Low soil zinc is attributed to a number of soil and environmental factors including low soil organic matter, high soil pH, calcareousness, water logging and arid climate (Mortvedt *et al.*, 1991; Tandon, 1995). Zinc deficiency in plants reduces not only the grain yield, but also the nutritional quality (Erenoglu *et al.*, 1999; Nan & Cheng, 2001). Zinc deficiency can lead to unbalanced nutritional status of plants (Graham *et al.*, 1987). Low solubility of zinc in soils rather than low total amount of zinc is the major reason for the widespread occurrence of zinc deficiency problem in crop plants (Cakmak, 2008). It is evident that for obtaining increased yield of wheat, zinc status of soil should be improved using zinc fertilizers (Shaheen *et al.*, 2007).

Ghulam *et al.*, (2009) found out that application of Zn alongside with NPK increased significantly improved growth and yield parameter of wheat. Their research further indicated that application of Zn increased its uptake by wheat crop and also resulted in a built up of Zn in the upper 15 cm layer to be available for the next crop. Yilmaz *et al.*, (1997) concluded that overuse of P-fertilizer resulted in even lower levels of Zn in wheat grain and human diets. A Zn concentration of 0.5 mg/Kg is adequate for the production of wheat (Farnandez & Hoefft, 2012).

1.2.1.8 Copper

Cu is an essential micro nutrient for plant growth at low concentrations, but excessive amount is phytotoxic (Michaud *et al.*, 2007). Copper is needed for chlorophyll production, respiration, and protein synthesis (McCauley *et al.*, 2009). Plants absorb

copper in ion form (Shanyn & Bradley, 1999). Copper deficient plants display chlorosis in younger leaves, stunted growth, delayed maturity (excessively late tillering in grain crops), lodging, and, in some cases, melanosis (brown discoloration). In cereals, grain production and fill is often poor, and under severe deficiency, grain heads may not even form (McCauley *et al.*, 2009). Copper deficient plants are prone to increased disease, specifically ergot (a fungus causing reduced yield and grain quality); (Solberg *et al.*, 1999). Buszewski *et al.*, (2000) determined the soil copper in Torun, Poland and found it to be 9.69mg/kg on average in spring and 5.03mg/kg in autumn. A Cu concentration of 1 mg/Kg is adequate for crop production (Farnandez & Hoefl, 2012).

1.2.1.9 Iron

Iron plays an important role in plant respiratory and photosynthetic reactions (McCauley *et al.*, 2009). Most iron deficiency occurs on calcareous, high pH soils and also in soils low in organic matter especially where land leveling has removed the upper organic rich soils and exposed calcareous subsoil (Clain & Jacobsen, 2009). Fe deficiency reduces chlorophyll production and is characterized by interveinal chlorosis with a sharp distinction between veins and chlorotic areas in young leaves. As the deficiency develops, the entire leaf will become whitish-yellow and progress to necrosis (McCauley *et al.*, 2009). Slow plant growth also occurs. When viewed from a distance, Fe deficient fields exhibit irregularly shaped yellow areas, especially where the subsoil is exposed at the surface (Follett & Westfall, 1992). Fernandez & Hoefl, 2012, stated a concentration of 4 mg/Kg and above to be adequate.

1.2.2 Soil pH

Soil pH is a measure of hydronium ion (H_3O^+ or more commonly, H^+) activity in a soil suspension. In very acidic soils, all the major plant nutrients including nitrogen (N),

phosphorous (P), potassium (K) and also the trace element molybdenum (Mo) may be unavailable, or only available in insufficient quantities (Gazey, 2009). The major impacts of acidity occur in the sub-surface soil. When soils acidify, aluminium in the soil becomes soluble. In this form, aluminium retards root growth, restricting access to nutrients and water deeper in the soil profile. Poor crop and pasture growth, yield reduction and smaller grain size occur as a result of inadequate water and nutrients (Gazey, 2009).

Nitrogen sources (fertilizers, manures, legumes) which contain or form ammonium ions increase soil acidity unless the plant directly absorbs the ammonium ions. The greater the nitrogen fertilization rate with these sources, the greater the soil acidification. As ammonium ions are converted to nitrates in the soil (nitrification), H^+ ions are released (Cliff, 2011). The main cause of soil acidity is the inefficient use of nitrogen fertilizers, particularly ammonium-based fertilizers. Ammonia is readily converted to nitrate in the soil, which if not taken up by plant roots, can leach away from the root zone, leaving the soil acidic (Gazey, 2009). For many plants a pH of 5.5-6.5 typically allows the various mineral nutrients to be absorbed at adequate levels; and not at levels too high that toxicity can result. Soil pH affects the availability of nutrients and how the nutrients react with each other. At a low pH, beneficial elements such as molybdenum (Mo), phosphorus (P), magnesium (Mg) and calcium (Ca) become less available to plants. Other elements such as aluminium (Al), iron (Fe) and manganese (Mn) may become more available and Al and Mn may reach levels that are toxic to plants. The changes in the availability of nutrients cause the majority of effects on plant growth attributed to acid soils. Wheat crop does best when the pH is between 5.0 and 6.5 (Lake, 2000).

1.2.3 Soil bacteria

Soil is a complex and dynamic biogeochemical system comprising tens of thousands to millions of bacterial species (Tanu & Hoque, 2013). Bacteria in the soil are very important in that soil would not be fertile without them and organic matter such as straw or leaves would accumulate within a short time (Kummerer, 2004). Environmental Stress like agriculture may reduce this bacterial diversity (Tanu & Hoque, 2013). Extensive use of agricultural chemicals of different chemical families in crop production lead to degradation of beneficial bacteria hence their physiological activities which are very important to the soil are adversely affected (Wani *et al.*, 2005; Srinivas *et al.*, 2008; Ahemed & Khan, 2010).

Soil microorganisms exist in large numbers in the soil as long as there is a carbon source for energy (Hoorman & Islam, 2010). Soil teams with microscopic life (bacteria, fungi, algae, protozoa and viruses) as well as macroscopic life such as earthworms, nematodes, mites, and insects, and also the root systems of plants. The numbers and kinds of microorganisms present in soil depend on many environmental factors such as amount and type of nutrients available, available moisture, degree of aeration, pH and temperature (Prescott *et al.*, 1999).

Fungi population numbers are smaller but they dominate the soil biomass when the soil is not disturbed. Bacteria, actinomycetes, and protozoa are hardy and can tolerate more soil disturbance than fungal populations so they dominate in tilled soils, while fungal and nematode populations tend to dominate in untilled soils (Hoorman & Islam, 2010). Soil bacteria and fungi play pivotal roles in various biochemical cycles and are responsible for the recycling of organic compounds (Wall & Virginia, 1999). Soil microorganisms also influence above ground ecosystems by contributing to plant nutrition, plant health, soil structure and soil fertility (O'Donnell *et al.*, 2001). Microorganism are the most important source of enzymes production each with a

specific role required by nature to break down compounds during biodegradation process (Pandey, 2000).

1.2.4 Soil sampling

Soil sampling and testing provide an inventory of nutrients in the soil (Jones and Jacobsen, 2001). Soil testing plays an important role in crop production and nutrients management (Reid, 2008). Soil testing for nitrogen (N), phosphorus (P), potassium (K), and sulfur (S) is done to help professionals determine crop nutrient needs and monitor previous management practices. The other macronutrients (calcium and magnesium) and micronutrients (boron, chlorine, copper, iron, manganese, molybdenum, nickel and zinc) are sometimes analyzed to diagnose nutrient deficiencies (Jones & Jacobsen, 2001). Sampling depth and timing are critical components of a well-designed sampling plan (Jones & Jacobsen, 2001).

There are various methods of soil sampling which include: haphazard sampling, judgment sampling and probability sampling (Carter & Gregorich, 2006).

Haphazard also known as accessibility or convenience sampling involves a series of non-reproducible, idiosyncratic decisions by the sampler and no systematic attempt is made to ensure that samples taken are representative of the population being sampled. This type of sampling is antithetical to scientific sampling designs (Carter & Gregorich, 2006). Judgment sampling also known as purposive sampling, involves the selection of sampling points based on knowledge held by the researcher. This method of sampling can result in accurate estimates of population parameters such as means and totals but cannot provide a measure of the accuracy of these estimates (Gilbert, 1987). Moreover the reliability of the estimate is only as good as the judgment of the researcher. The third method is probability sampling which selects sampling points at random locations using a range of specific sample layouts, and the probability of sample point selection can be calculated for each design. This allows an estimate to be made of the accuracy of the

parameter estimates, unlike judgment sampling. This allows a range of statistical analyses based on the estimates of variability about the mean to be used, and is by far the most common type of sampling in soil science (Carter & Gregorich, 2006).

There are two main types of sampling designs and these are systematic and random. Random sampling is further divided into two: simple random and stratified random. In simple random sampling all samples of the specified size are equally likely to be the one chosen for sampling. In stratified random sampling, points are assigned to predefined groups or strata and a simple random sample chosen from each stratum. The probability of being selected can be weighted proportionally to the stratum size or the fraction of points sampled can vary from class to class in disproportionate sampling. Disproportionate sampling would be used if the degree of variability is believed to vary greatly between classes, in which case a higher number of samples should be drawn from the highly variable classes to ensure the same degree of accuracy in the statistical estimates.

Stratified sampling (correctly applied) is likely to give a better result than simple random sampling, but four main requirements should be met before it is chosen (Williams 1984):

- i. Population must be stratified in advance of the sampling.
- ii. Classes must be exhaustive and mutually exclusive (i.e., all elements of the population must fall into exactly one class).
- iii. Classes must differ in the attribute or property under study; otherwise there is no gain in precision over simple random sampling.
- iv. Selection of items to represent each class (i.e., the sample drawn from each class) must be random.

The most commonly used sampling design for many field studies is systematic sampling using either transects or grids. Systematic sampling designs are often criticized by statisticians but the ease with which they can be used and the efficiency with which they

gather information makes them popular in the field of earth sciences. Ideally the initial point of the transect or grid and its orientation should be randomly selected. The major caution in the use of systematic sampling with a constant spacing is that the objects to be sampled must not be arranged in an orderly manner which might correspond to the spacing along the transect or the grid (Carter & Gregorich, 2006).

In random sampling individual samples are collected from locations that are randomly distributed across the representative portion of the field. A zigzag sampling pattern is often used for field sampling. The sampler should avoid sampling typical areas such as eroded knolls, depressions, saline areas, fence lines, old roadways and yards, water channels, manure piles, and field edges. Typically, all samples are combined and a composite sample is taken for analysis. Composite sampling is comparatively inexpensive since only one sample from each field or subsection of a field is taken for analysis. However, this design provides no assessment of field variability, and relies on the ability of the farm operator to identify portions of the field that may have inherently different nutrient levels. Soil-testing laboratory guidelines consistently suggest that on average 20 samples be collected for each field or subsection of a field regardless of the actual area involved (Carter & Gregorich, 2006).

A necessary and important step in the planning stages of a project is to determine the number of samples required to achieve some prespecified accuracy for the estimated mean. One approach is to use prior knowledge about the coefficient of variation (CV) of the property under study to estimate sample numbers required to achieve a certain prespecified relative error. The relative error (dr) is defined as

$$dr = (\text{sample mean} - \text{population mean}) / \text{population mean}$$

The sample numbers required to achieve a specified relative error at a selected confidence level can be estimated from Table 1.1. For example, at a confidence level of 0.95 and a relative error of 0.25, 17 samples are required if the CV is 50% and 45

samples are required if the relative error is 0.25 at a confidence level of 0.90 and CV is 100% (Pennock *et al*, 2006).

Table 1.1 : Sample sizes Required for Estimating the True Mean

Confidence level	Relative Error, dr	Coefficient of Variation (CV)%					
		10	20	40	50	100	150
0.8							
	0.1	2	7	27	42	165	370
	0.25			6	7	27	60
	0.05				2	7	15
	1					2	4
0.9	0.1	2	12	45	70	271	609
	0.25			9	12	45	92
	0.05				2	13	26
	1					2	8
0.95	0.1	4	17	63	97	385	865
	0.25			12	17	62	139
	0.05				4	16	35
	1					9	16

-

(Source: Gilbert, 1987)

1.2.5 Review of analytical techniques

UV-Visible spectrophotometry is one of the most frequently employed technique in pharmaceutical analysis. It involves measuring the amount of ultraviolet or visible radiation absorbed by a substance in solution (Bahera, *et al.*, 2012). UV-visible spectrophotometer uses light over the ultraviolet range (185 - 400 nm) and visible range (400 - 700 nm) of electromagnetic radiation spectrum (Filip *et al.*, 2012). UV-Vis spectrophotometer measures the intensity of light passing through a sample (I), and compares it to the intensity of light before it passes through the sample (I_0) expressed in absorbance (A) or transmittance (T). The ratio is called the reflectance, and is usually expressed as a percentage (%R). A beam of light from a visible and or UV light source (colored red) is separated into its component wavelengths by a prism or diffraction grating (Filip *et al.*, 2012).

Atomic Absorption Spectrophotometry (AAS) is a technique for measuring quantities of chemical elements present in environmental samples by measuring the absorbed radiation by the chemical element of interest (Garcia & Baez, 2013; Filip, 2012). This is done by reading the spectra produced when the sample is excited by radiation. Atomic absorption methods measure the amount of energy in the form of photons of light that are absorbed by the sample. A detector measures the wavelengths of light transmitted by the sample, and compares them to the wavelengths which originally passed through the sample. A signal processor then integrates the changes in wavelength absorbed, which appear in the readout as peaks of energy absorption at discrete wavelengths. The energy required for an electron to leave an atom is known as ionization energy and is specific to each chemical element. When an electron moves from one energy level to another within the atom, a photon is emitted with energy E (Garcia & Baez, 2013). Atoms of an element emit a characteristic spectral line. Every atom has its own distinct pattern of wavelengths at which it will absorb energy, due to the unique configuration of electrons

in its outer shell. This enables the qualitative analysis of a sample. The concentration is calculated based on the Beer-Lambert law. Absorbance is directly proportional to the concentration of the analyte absorbed for the existing set of conditions. The concentration is usually determined from a calibration curve, obtained using standards of known concentration (Garcia & Baez, 2013).

1.3 Statement of the problem

Narok north sub-county is a wheat growing area but farmers have experienced decreased wheat yields over the years. This has been mainly attributed to climate change and also lack of the knowledge on the requirements of wheat in terms of macronutrients, micronutrients, soil pH and even the organic matter content of the soil. Despite the use of fertilizers during planting and even when the crop has grown, farmers continue to experience low yields. Application of these fertilizers has become a routine without the knowledge of the nutrients already available in the soil or the knowledge of the nutrients lacking. Hence this research set out to determine the levels of various macronutrients, micronutrients, soil pH, soil bacteria and organic matter content in soil sampled at wheat farms in Narok north.

1.4 Justification

Wheat is one of the major crops that are central to achieving development in agriculture and is ranked second most important cereal crop after maize in Kenya (Mahagayu *et al.*, 2007). Consumption stands approximately at 700,000 tonnes while production is only 350,000 per year (NDP, 2003; Monrof, *et al.*, 2013) the short fall being met by 50% imports. This problem may be due to lack of knowledge on the correct requirement for wheat production in terms of nutrients. This study determined the levels of macro and micro nutrients in the soil and compare them with the levels that are required for a good yield, therefore giving information to the agriculture sector and farmers of Narok north

sub county on what may be the cause of the poor yields despite the continuous use of fertilizer which is primarily meant to maximize yields.

1.5 Research Hypothesis

The soils in the wheat growing farms in Narok north sub-county are deficient in nutrients.

1.6 Objective of the study

1.6.1 General objective

The main objective of this study was to determine the levels of nutrients in the soil in the wheat planting areas of Narok north sub-county.

1.6.2 Specific objectives

1. To determine the pH and the total organic matter content of the soil in the wheat growing areas in Narok north sub-county.
2. To determine diversity of aerobic bacteria in the soil in Narok north sub-county before and after planting of wheat.
3. To determine the levels of macronutrients (N, P, K, Ca, Mg and S) and micronutrients (Cu, Fe, and Zn) in the soil before and after planting of wheat in Narok north sub-county.

CHAPTER TWO

2.0 MATERIALS AND METHODS

2.1. Study Area

The area of study was in Narok north sub-county located in the southern part of the Rift valley of Kenya and borders north of Tanzania. Its geographical co-ordinates are $1^{\circ} 5' 0''$ south and $35^{\circ} 52' 0''$ East with an altitude of 1827 (5,997 feet) above sea level.

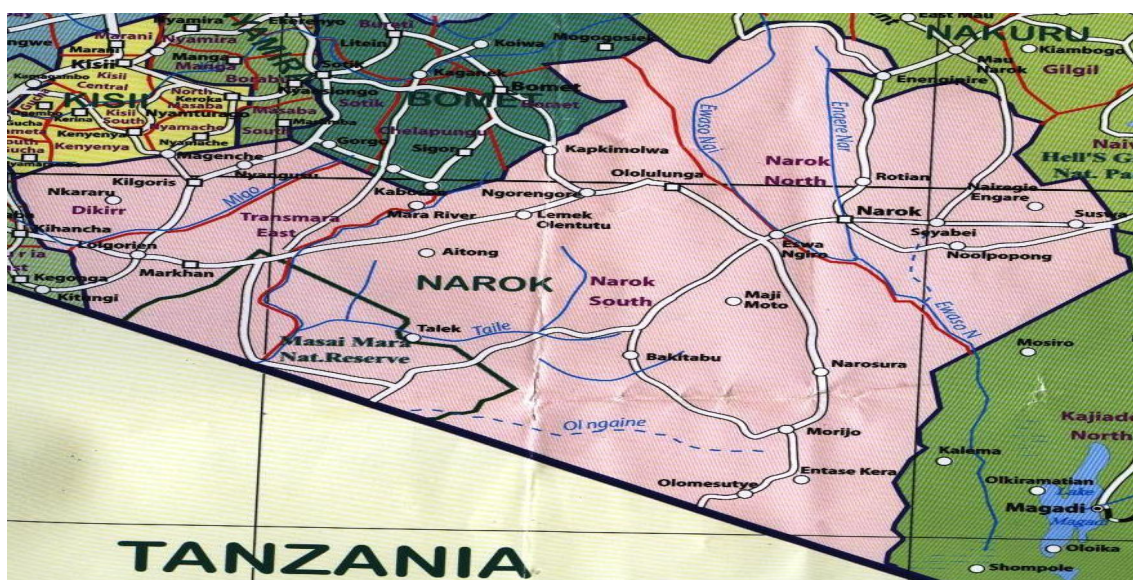


Figure 2.1 Map showing location of Narok North sub-county (flicker.com/photo, 2013)

2.2 Sample collection

Soil samples were collected during the months February March and April, 2013 in a stratified way but randomly (Carter and Gregorich, 2006) from various wheat farms in Narok north sub-county before the planting period and after planting. The soil samples were collected at the depth of 0 to 15cm (Carter & Gregorich, 2006; Mahler & Tindall,

1990). Once in the wheat farms to be sampled several sub samples depending on the size of the farm were collected in a zigzag manner and mixed thoroughly and a composite sample picked from the mixture and put in plastic bags. (Mahler & Tindall, 1990). A total of 45 samples were collected from the entire area as indicated in Table 1.1 with 15 samples from each of the three farming areas in Narok north namely Ololulunga, Ntulele and Mau. These samples were transported to Maasai Mara University and JKUAT laboratories where they were air dried and stored in plastic bags ready for analysis. The control sample was picked from an area where farming has not been done since 1960's as per the knowledge of the local people living in the area. Sample 1 to 15 were collected from Ololulunga, 16 to 30 and 31 to 45 were sampled from Ntulele and Mau respectively.

2.3 Apparatus and instrumentation

Apparatus used include mortar and pestle, volumetric flasks, filter papers, beakers, plastic sample bottles, universal bottles, Petri dishes, crucibles, pipettes, mechanical shakers and hot plate with stirrer. The instruments used include pH meter (Hanna P211) Analytical weighing balance, Microscope (Optika B350), Flame photometer (AFP 100) for analysis of K, an oven for drying (FN 400), atomic absorption spectrophotometer (model PG- 990) for analysis of Fe, Zn, Mg, Ca and Cu, UV-VIS spectrophotometer (Shimadzu 1240) for analysis of sulfates, phosphates and nitrates and an incubator (Labtech LIB-030M) for bacteria culturing and autoclave (All American Model 25X) for sterilizing of media and apparatus used for analysis of bacteria.

All glassware which included beakers, pipettes, measuring cylinders, volumetric flasks and conical flasks were thoroughly cleaned with tap water and detergent. They were rinsed with water and twice with 4% nitric acid solution. They were then rinsed with water and finally with de-ionized water. The glassware were then dried in the oven at 105°C.

2.4 Chemicals and Reagents

The reagents which were used were of analytical grade (AR) and included sulfuric acid, nitric acid, orthophosphoric acid, perchloric acid, phenol, nitro phenol indicator, hydrochloric acid, potassium dichromate, disulphonic acid, ammonium acetate and ammonium hydroxide. Preparations of these reagents and chemical were done using distilled de-ionized water.

2.5 Preparation of standards

All the chemicals and reagents used in preparation of standards were of analytical grade.

A copper stock standard solution (1000ppm Cu^{2+}) was prepared by dissolving 3.989g of $\text{CuSO}_4 \cdot 5\text{H}_2\text{O}$ in de-ionized water and made up to the mark in 1000mL volumetric flask and different standard solutions were prepared from it through serial dilution. A zinc stock solution (1000ppm Zn^{2+}) was prepared by dissolving 4.697g of $\text{Zn}(\text{NO}_3)_2 \cdot 6\text{H}_2\text{O}$ and made up to the mark in a 250mL volumetric flask with de-ionized water. Working standards were prepared from it through serial dilution. Iron stock solution (1000ppm Fe^{2+}) was prepared by dissolving 5.054g of $\text{Fe}(\text{NO}_3)_2 \cdot 7\text{H}_2\text{O}$ in de-ionized water and made up to the mark in 1000mL volumetric flask. Different standards were made from it through serial dilution.

Magnesium stock solution (1000ppm Mg^{2+}) was prepared by dissolving 8.366g of $\text{MgCl}_2 \cdot 6\text{H}_2\text{O}$ in de-ionized water and made up to the mark in 1000mL volumetric flask. Different concentrations of standards were made from it through serial dilution. Calcium stock solution (1000 ppm Ca^{2+}) was prepared by dissolving 7.0 g of $\text{Ca}(\text{NO}_3)_2 \cdot 4\text{H}_2\text{O}$ in de-ionized in and made up to the mark in 1000mL volumetric flask. Different concentrations of standards were made from it through serial dilution.

About 2.5g of potassium dihydrogen phosphate (KH_2PO_4) were dried in the oven at 105°C for 1 hour, cooled in a desiccator, and stored in a tightly stoppered bottle. A mass of 0.4393g of dried KH_2PO_4 were dissolved in deionized water and topped up to 1000mL. A series of standard solutions were made from this stock solution through serial dilution. A volume of 5mL of vanadomolybdate was added to each working standards made.

About 3g of KCl were dried in the oven at 120°C for 2 hours and cooled in a desiccator. A mass of 1.907g of the dried KCl were dissolved in deionized water and topped up to 1000mL to get 1000ppm K^+ stock solution. A series of standards were prepared from the stock solution through serial dilution. A mass of 0.5434g of potassium sulfate were dissolved in distilled deionised water and topped up to 1000mL in a volumetric flask to make 100ppm sulfate. A series of standards were made from this stock solution through serial dilution. The working standards were conditioned using 5mL the conditioning reagent (30mL HCl, 100mL ethanol, 75g NaCl and 50mL glycerol) and 5mL of BaCl_2 .

The nutrient agar (28g/L) was used, which was weighed and dissolved in distilled water by warming on a hot plate and autoclaved under pressure for fifteen minutes at 121°C . Finally, the media were dispensed in sterile petri dishes (Cappucino and Sherman, 2002).

2.6 Experimental procedure

2.6.1 pH measurement

The pH meter was first calibrated using pH buffer 4 and pH buffer 9 before the pHs of the samples were taken. A scoop of 5g of soil was taken into paper cup and 5 mL of distilled de-ionized water was added and stirred vigorously for 5seconds and let to stand for 10 minutes. The pH electrode was placed in the soil slurry and the pH was read immediately.

2.6.2 Determination of total organic matter (TOM)

A mass of 0.1g of air dried soil was placed in conical flasks and 10 mL of 1 M potassium dichromate was added and the contents swirled. Then 15 mL of sulfuric acid was added and swirled again for two minutes and then allowed to stand for 30 minutes before 150 mL of water was added followed by Orthophosphoric acid. The contents were the titrated with 0.5 M ferrous ammonium sulphate solution till the colour changes from blue to green. A blank was run simultaneously without the soil (Walkley- Black, 1934).

2.6.3 Digestion of materials for metal analysis (Cu, Zn, Fe, Mg and Ca)

Digestion of soil was carried out as follows: soil samples of 0.1g were put in a beaker and 5ml of tri-acid mixture (concentrated HNO₃, HClO₄ and H₂SO₄ in the ratio 3:1:1) were added. The mixture was heated on a hot plate at 105°C in the hood until white fumes were observed (Lindsay and Norvell, 1978). The digested soil samples were then filtered using a Watmann filter paper No. 42 into a 100 mL volumetric flask and topped up to the mark with distilled water and two drops of HNO₃ were added for preservation. The digested sample was transferred into plastic bottles ready for analysis using AAS.

2.6.4 Digestion of materials for phosphorous analysis

A mass of 1g of each of the soil sample was weighed and placed in conical flasks, 50 mL of the diacid (HCl and HNO₃ in the ratio of 3:1) was added and shaken for every 30 minutes in the mechanical shaker, then left standing for 6 hours to allow complete digestion. The contents were then filtered using a porous filter paper no 42. 10 mL of the filtered sample was then placed in a boiling tube and 3 drops of the nitro-phenol indicator was added. The colour sample turned yellow by addition of 6M ammonia solution. It was then decolorized using 1M HNO₃, then 5mL of vanadomolybdate. The volume is made to 50 mL in a volumetric flasks using distilled water (Olsen *et al.*,

1954). The concentration of the samples in mg/Kg was read after 30 minutes at 400 nm in a UV-VIS spectrophotometer. The phosphates concentration in the sample aliquots was read with the help of the standard curve obtained by running standards.

2.6.5 Digestion of material for potassium analysis

A mass of 5g of air dried soil were weighed into a 50 mL centrifuge tube and 33 mL of ammonium acetate added and were shaken on a shaker. The mixture was centrifuged until a supernatant liquid is clear. The extract was put in 100 mL volumetric flask through a filter paper and topped up to the mark using 1M ammonium acetate solution. The extracts were analyzed using a flame photometer at wavelength of 767 nm (Peech *et al.*, 1947)

2.6.6 Digestion of material for analysis of Sulfates

A mass of 1g of the air dried soil sample was weighed and placed in plastic bottles, 100 mL of distilled water was then added and the contents shaken for 1 hour in a mechanical shaker. A volume of 50 mL of the sample was taken into 250 volumetric flask and topped up to the mark. The sample was further diluted 50 times (1mL in 50 mL of distilled water). A volume of 5 mL of the conditioning agent (30 mL HCL, 100 mL ethanol, 75g NaCl and 50mL glycerol) was added followed by 5ml BaCl₂ solution. All the samples turned milky in colour. The intensity of the milky colour was read at 420 nm using a UV-VIS.

2.6.7 Digestion of material for analysis of nitrates

A digestion mixture was prepared by digestion using a mixture of 25g of phenol in 250mL concentrated sulphuric acid. A mass of 1g of sample was weighed and placed in a glass conical flask and then 50ml of the digestion mixture was added. The contents were left to stand for 6 hours. Then 25ml of the digest was placed in crucible and

evaporated to dryness on a hotplate. A volume of 3ml of phenol disulphonic acid was added and swirled gently and left to stand for 10minutes after which 15ml of distilled water was added and stirred with a glass rod. And on cooling 3 drops of para nitro-phenol indicator is added. Ammonia solution was added until intense yellow colour was achieved. The sample volume was then topped to 100mL and left to stand for 30 minutes and the intensity of the yellow colour read at 420nm in a colorimeter.

2.6.8. Culturing and Isolation of bacterial isolates

The samples were aseptically ground in a motor and pestle in potassium dihydrogen phosphate buffer (pH 6.8). An aliquot (1 mL) was taken and placed in the same buffer (9 mL) for serial dilution. This procedure was repeated to form a fivefold serial dilution. The serially diluted aliquot (100µl) were then inoculated on nutrient agar. The cultures were then placed in an incubator at 30⁰C for 24hours to allow for endophyte growth. Individual colonies were picked and streaked on fresh media for purification to generate pure cultures. The pure cultures were then used to perform morphological and biochemical characterization (Cappucino and Sherman, 2002).

2.6.9. Morphological characterization of bacteria Isolates

This was done to determine the cell shape of the bacterial cells, in each case the classical gram staining method were used (Bathlomew, 1962). Smears of bacterial isolates were prepared and heat fixed, after which they were flooded with crystal violet (Sigma Aldrich, Steinheim, Germany) and left to stand for a minute. This were gently washed with tap water and then flooded with gram iodine (Sigma Aldrich, Steinheim, Germany) which were washed with tap water after a minute. Decolorisation with 95% ethanol (Schorlab S.L. Spain) were done and the counter stain (Safranin) applied for forty five seconds. Smears were then washed gently with tap water, and blot dried for observation under oil immersion.

2.6.10. Biochemical characterization of bacteria isolates

These tests were done to determine the metabolic properties of different bacterial strains that were isolated. These biochemical tests were done as follows;

2.6.10.1 Urease test

The pure isolates were inoculated in urea broth and incubated for 24 hours at 30⁰C and the changes noted. The urease test was used to determine the ability of an organism to split urea into two units of ammonia by the enzyme urease produced by the isolates. The formation of the two units of ammonia results into increased salinity which were detected using a pH indicator, phenol red in Christensen's urea which under acidic conditions (pH 6.8) is yellow and rose pink in alkaline conditions (pH 8.4). Positive reaction was indicated by development of a deep pink color (Cappuccino and Sherman, 2002).

2.6.10.2. Oxidase test

Oxidase positive bacteria possess cytochrome oxidase or indophenols oxidase (an iron containing haemoprotein). They both catalyze the transport of electrons from donor compounds (NADH) to electron acceptors (usually oxygen). The test reagent N,N,N',N'-tetra-methyl-p-phenylenediamine dihydrochloride acts as an artificial electron acceptor for the enzyme oxidase. The oxidized reagent forms the colored compound indophenol blue. In this case, impregnated oxidase test strip method were used. A 24 hour old culture were scrapped using the oxidase discs and a blue color is expected to develop after 10 seconds to indicate a positive result for oxidase test (Cappuccino and Sherman, 2002).

2.6.10.3 Gelatin liquefaction test

Nutrient broth supplemented with 12% gelatin were used to demonstrate hydrolytic activity of gelatinase. The presence of gelatinase enzyme breaks down gelatin, which liquefies the media. After incubation, the cultures that remain liquefied when placed in the refrigerator at 4⁰C for 30 minutes were considered positive for gelatin hydrolysis (Cappuccino and Sherman, 2002).

2.6.10.4. Indole and hydrogen sulfide production

Sulfur indole motility (SIM) agar media were used to demonstrate the production of tryptophanase and cysteine desulfurase by the isolates. Tryptophanase utilizes the amino acid tryptophan as a carbon and energy source producing indole, pyruvic acid and ammonia. The indole produced were detected by addition of Kovac's reagent (p-dimethylamino-benzaldehyde) (Harold, 2002). Positive results for indole were indicated by production of a cherry red layer. Cysteine desulfurase breaks down sulfur containing amino acids producing pyruvate, ammonia and hydrogen sulfide. Iron in the medium reacts with hydrogen sulfide producing the characteristic black precipitate which is a positive test for hydrogen sulfide production by the isolates (Cappuccino and Sherman, 2002). This indole test was further confirmed using trypton water by incubating the culture in trypton water.

2.6.10.5. Catalase Test

A small drop of normal saline was placed on a clean glass slide. A 24 hour old culture was then placed on the glass slide with a sterilized inoculating loop to make a smear. With a Pasteur pipette a drop of hydrogen peroxide was placed over the test smear. The appearance of gas bubbles indicated a positive result (Cappucino and Sherman, 2002).

2.6.10.6. Methyl Red-Vogues Pasteuer (MR-VP)

Methyl red test was done using a 48 hour old culture incubated in MR-VP broth at 37⁰C. Five drops of methyl red was then added to the culture and a positive result was indicated by a red colour and a negative result was indicated by a yellow appearance of the media.

VP test was done using 1 mL of a 48 hour old culture in a test tube at room temperature by adding 0.6 mL of alpha-naphthol and 0.2 mL of 40% potassium hydroxide. The results were read after 4hours. A positive result was indicated by an eosin pink color (Cappucino and Sherman, 2002).

2.6.10.7. Citrate utilization

Organisms that are able to use citrate as the sole source for metabolism and growth are able to grow on Simmons Citrate agar (Sigma Aldrich, Steinheim, Germany). The bacteria metabolize the citrate forming alkaline conditions in the medium. This is shown by the pH indicator in Simmons citrate agar, bromothymol blue which is green in acidic conditions and royal blue in alkaline conditions (Jason and Woodland, 2004). A 24 hour old culture was incubated in citrate agar slanted in a tube. A positive result was indicated by the green media turning blue.

2.6.10.8 Triple Sugar Iron (TSI)

Triple Sugar Iron is a medium that can determine the ability of an organism to utilize specific carbohydrates incorporated in a basal growth medium. TSI contains three sugars in varying concentrations: glucose (1X), lactose (10X) and sucrose (10X). It also contains the pH indicator phenol red. If sugar fermentation occurs, glucose will be initially used and the butt of the tube will be acidic (yellow). After glucose utilization the organism may continue to ferment the remaining sugars hence the entire tube will be acidic. Certain bacteria are unable to utilize any sugars and will break down the peptone present. Peptone utilization causes an alkaline shift in the media that causes the color

change from orange to red. Blackened medium is caused by hydrogen sulfide production, which changes ferrous sulfate to ferrous sulfide. Splitting of the medium or presences of bubbles in the bottom of the tube indicates production of gas (Jason and Woodland, 2004). A 24 hour old, pure culture was inoculated in a both slant and butt in a tube containing TSI medium and incubated for 24 hours and the changes in the medium was noted (Cappucino and Sherman, 2002).

2.7 Data Analysis

The tests were done three times and results obtained expressed as mean and standard deviation. The statistical data during research were stored and recorded in research log book. Comparison of statistical differences was done using student T-test, MSTAT-C and Correlation analysis, SPSS was used for clustering.

CHAPTER THREE

3.1 RESULTS AND DISCUSSION

3.1.1 Soil pH

The pH of the soil samples collected were determined and the results tabulated as shown in Table 3.1. Samples 1 to 15 were sampled from Ololulunga, 16 to 30 from Ntulele and 31 to 45 from Mau. The pH ranged between 5.15 and 7.19 before planting and between 5.44 and 7.22 after planting in all the sampled areas. The highest acidic soils before planting were samples 6 and 37 both with a pH of 5.15 and least acidic during the same season was sample 13 with a pH of around neutral of 7.19. After planting, the most acidic was sample 8 with a pH of 5.44 and least acidic were samples 13 and 29 with a pH of 7.22. All the samples 6, 13 and 8 were from Ololulunga area and sample 29 and 37 were from Ntulele and Mau region respectively. The average pH of all the sampled areas were 6.321 before planting and 6.168 after planting for Ololulunga area, 6.441 before planting and 6.399 after planting for Ntulele area and for Mau region the average pH were 6.470 and 6.443 before and after planting respectively. Ololulunga area was found to have the most acidic soils (6.321) before planting as compared to both Mau and Ntulele with an average of 6.441 and 6.470 respectively.

After planting Ololulunga area had the most acidic soils with an average pH of 6.168 followed by Ntulele area with a pH of 6.399 and the least acidic was Mau with a pH of 6.443. The CV% revealed that pH had a low variation of 6.80% in Ololulunga, 3.56% in Ntulele and 4.81% in Mau and this may be attributed to the bedrock being the same in the sampled areas and the application of the nutrients did not affect the soil pH maybe due to the fact that the mulching acts as buffer.

The general trend in the pH is that it decreased slightly after planting as compared to before planting with 60% of the samples showing a decrease. This can be attributed to

addition of limestone during planting. The soil samples from all the sampled areas were found to be slightly acidic during the two seasons with a mean pH of 6.413 ± 0.373 before planting and 6.336 ± 0.359 after planting. Most plants suffer when soil pH is below 4.8 (Espinoza *et al.*, 2001). Wheat grows best when the soil pH is between a pH 6.0 and 7.0 (Vitosh, 1998). In that case it can be concluded that all soils from all the sampled areas had a pH which is suitable for plant growth (Espinoza *et al.*, 2001). According to Vitosh, 1998, 82% of the samples in this study before planting had a pH suitable for wheat growth with only 9% with a pH of below 6 and 9% above pH 7. Yost *et al.*, (1997) generalized adequate pH to be between 5.8 and 6.2.

All the samples before planting except two (sample 6 and 37) had a pH above 5.8 but only three samples fall within the range reported by Yost *et al.*, (1997) of between 5.8 and 6.2. After planting only 13 samples fall within that range. Daryl (2004), generalized the soil pHs as follows; below 4.5 (very low), 4.5-5.3 (low), 5.3-6.0 (medium), 6.0-7.5 (High), more than 7.5 (very high). A medium pH indicates a need for limestone in the near future, but soil acidity is likely not causing yield reductions at the time of test. Soils rated high have a soil pH optimum for crop growth and limestone is not needed in the following two or three years (Daryl, 2004). This indicates that the sampled soils ranged between medium and high with most of them being high. It can therefore be concluded that the pH of the sampled areas are within the range for optimum wheat growth. A study done by Javed *et al.*, (2011) found that the average pH of the soils was 7.3 which is higher than all the averages for the two seasons found in this study, that is, 6.321 and 6.168 for Ololulunga, 6.441 and 6.399 for Ntulele and 6.470 and 6.430 for Mau. These results also differ greatly from those reported by Hamilton *et al.*, (1993) of an average pH of 7.8. Akporhonor *et al.*, (2005) in a study on nutrients uptake found an average pH of the soil to be 6.05 which is slightly lower than the average pH found in this study. The pH decreased slightly after planting as compared to before planting but for both seasons the pH was found to be suitable for wheat growing for all the farms sampled. The mean difference between pH before planting and pH after planting for the whole area was not significant ($P > 0.05$)

Table 3.1 pH of soil samples from all the sampled areas before and after planting

Sample	pH Before planting	pH After planting	Sample	pH Before planting	pH After planting
1	5.90±0.017	6.33±0.007	24	6.34±0.003	6.20±0.002
2	6.54±0.001	6.33±0.006	25	6.34±0.016	6.12±0.004
3	6.64±0.003	6.20±0.001	26	6.89±0.002	6.61±0.004
4	6.77±0.011	6.48±0.005	27	6.49±0.001	6.72±0.016
5	6.10±0.003	6.43±0.004	28	6.53±0.015	6.13±0.012
6	5.15±0.001	6.39±0.013	29	6.28±0.011	7.22±0.009
7	6.59±0.004	5.49±0.002	30	6.33±0.000	6.08±0.000
8	5.79±0.001	5.44±0.017	31	6.76±0.023	6.55±0.001
9	6.33±0.002	6.05±0.006	32	6.53±0.002	6.78±0.009
10	6.43±0.001	6.16±0.001	33	6.66±0.000	6.69±0.010
11	6.58±0.020	5.62±0.005	34	6.36±0.007	6.35±0.002
12	6.19±0.009	6.43±0.001	35	6.62±0.008	6.55±0.000
13	7.19±0.013	7.22±0.007	36	6.27±0.002	6.46±0.001
14	6.28±0.000	5.52±0.021	37	5.15±0.017	6.48±0.012
15	6.33±0.007	6.43±0.018	38	6.52±0.009	6.47±0.016
16	6.45±0.011	6.53±0.009	39	6.57±0.011	6.60±0.018
17	6.64±0.017	6.35±0.004	40	6.90±0.001	6.68±0.016
18	6.72±0.001	6.52±0.002	41	6.86±0.009	6.14±0.000
19	6.33±0.009	6.34±0.008	42	6.45±0.011	6.22±0.001
20	6.36±0.011	6.16±0.011	43	6.49±0.000	6.24±0.009
21	6.21±0.017	6.29±0.001	44	6.43±0.001	6.21±0.006
22	6.36±0.000	6.37±0.009	45	6.48±0.009	6.23±0.004
23	6.45±0.009	6.32±0.011	Control	6.59±0.009	-

3.1.2 Total organic matter

The total organic matter in the soil was determined from Ololulunga, Ntulele and Mau areas and results tabulated in Table 3.2. The organic matter ranged from 5.101 to 11.284 before planting and from 6.801 to 12.051 after planting in all the sampled areas. The sample with the highest organic matter before planting was samples 25 and least was sample 34. After planting, sample 33 had the highest organic matter and the least was sample 27. The average organic matter for all the sampled areas was 8.574 before planting and 10.006 after planting for Ololulunga area, 8.996 before planting and 10.285 after planting for Ntulele area and for Mau region the average organic matter was 7.739 and 9.986 before and after planting respectively. Ntulele area was found to have the most organic matter (8.996) before planting as compared to both Ololulunga and Mau with an average of 8.574 and 7.739 respectively. After planting Ntulele area had the most organic matter with an average of 10.285 followed by Ololulunga area with a mean of 10.006 and Mau had the least organic matter of 9.986. The mean difference between the organic matter before and after planting for the whole area was significant ($P < 0.05$) indicating an increase in organic matter after planting.

Generally the organic matter for all the three areas increased after planting and these may be due to plant remains from previous seasons completely decaying after the tilling of the land. The range in this study is far much higher than that found in soils of Varanasi in a study done by Jyotsana *et al.*, (2013). Vijayakumar *et al.*, (2011) in their study found a range of 0.50 to 15.40% organic matter. This range agrees to some extent with the range found in this study. Organic matter contributes to improved soil properties but on many soils, suitable soil properties occur at relatively low levels of organic matter of between 2 to 4 percent (Manitoba, 2013). This therefore indicates that all the samples in this study had organic matter content above the recommended level.

TABLE 3.2 Percent (%) organic matter of soil from all the sampled areas

Sample	Before planting	After planting	Sample	Before planting	After planting
1	8.193	10.202	24	8.966	10.048
2	9.120	10.048	25	11.284	10.975
3	9.738	10.821	26	6.801	9.429
4	9.275	10.975	27	4.792	7.729
5	10.048	9.893	28	8.038	10.821
6	8.502	9.429	29	10.048	10.511
7	7.729	9.120	30	10.357	11.284
8	7.884	9.584	31	10.666	11.439
9	8.193	10.048	32	7.420	9.120
10	10.511	11.593	33	10.821	12.057
11	7.265	8.966	34	5.101	8.656
12	9.275	10.666	35	6.338	8.347
13	8.966	9.893	36	9.584	11.130
14	6.029	8.656	37	9.120	10.511
15	7.884	10.202	38	9.584	10.821
16	9.275	11.130	39	8.656	10.511
17	7.729	9.275	40	6.956	8.966
18	8.656	9.429	41	8.193	9.275
19	10.511	11.130	42	8.038	9.738
20	8.347	9.893	43	7.111	9.120
21	10.048	11.284	44	5.565	11.903
22	9.429	9.893	45	5.565	10.202
23	8.193	10.202	Control	7.260	-

3.1.3 Levels of Calcium, Magnesium and Potassium

The concentrations of Ca, Mg and K in the samples from the whole were tabulated in the Table 3.3. The concentration of K in the soil before planting was found to range between (21.800 mg/Kg and 66.867 mg/Kg) and (14.867 to 54.010 mg/Kg) after planting. Before planting sample 41 which was from the Mau region had the highest concentration of 66.867 ± 0.635 and sample 42 also from Mau had the least concentration of 21.800 ± 0.818 . After planting sample 34 from Mau region had the highest concentration of 54.010 ± 0.503 and sample 5 from Ololulunga area had the least concentration of 14.867 ± 0.643 . The average concentration of K before planting was found to be 42.486 ± 10.007 and 37.366 ± 9.376 after planting. Ololulunga area had the highest average concentration before planting of 45.622 ± 9.288 followed by Ntulele with a mean of 42.546 ± 9.288 and least being Mau region with mean of 37.613 ± 10.146 . After planting Ntulele had the highest concentration of 42.344 ± 7.728 followed by Mau region with a mean of 38.918 ± 6.365 and least was Ololulunga with a mean of 36.797 ± 10.399 .

The control sample had a concentration of 50.100 ± 0.503 which is higher than all the means from the three different areas with only four samples (40, 2, 19, 34) having a concentration above that of the control. K exhibited low levels of variability in all the sampled areas with a CV of 13.95% in Ololulunga, 15.25% in Ntulele and 28.08% in Mau and this will be attributed to depletion of mineral by plants from the bedrock concentration. It will also be due the fact that the added K had little impact on the overall concentration change for the two seasons due the large amount available K from the bedrock. Potassium is most available through wide range of pH (Hoeft *et al.*, 2000). K is bound more weakly to the soil, so can leach out of the surface soil especially at low pH (Clain and Jacobsen, 2001).

According to Fulton (2001) soils with a K concentration of less than 75 mg/Kg is considered to be very low, 75-150 mg/Kg low, 150- 250 mg/Kg medium, 250-800 mg/Kg high and 800 mg/Kg very high. All the sampled areas before and after planting had a concentration of below 75 mg/Kg which indicate a potassium deficiency in the soil, but for wheat production a minimum concentration of as low as 100lb/A (50 mg/Kg) in the soil is adequate (Farnandez and Hoefl, 2012). This therefore indicates that 10 samples before planting had sufficient potassium concentration and the rest had a concentration slightly below 50 mg/Kg. A level of 125 mg/Kg of K in the top 6 inches of soils determined by ammonium acetate is generally considered as the critical level (Rigas and Norm, 2000) this indicate that all sampled areas in this study had a concentration below the critical level. Majority of prairie soils of Canada as tested by Rigas and Norm (2000) were found to have a concentration of 300 mg/Kg in excess in the top 6 inches. These soils therefore have a concentration way above those determined in this study. Greg and Lindsey (2012) determined the critical level of K concentration to be between 100-130 mg/Kg. All the sampled areas in this research were found to be below this level reported by Greg and Lindsay (2012). Bijay *et al.*, (2004), found exchangeable K to range between 78 to 273 mg/Kg in the Indo-Gangetic plains of India. This range is way above the range found in this study. Yadvinder and Bijay, (2001) stated that a concentration less than 55 mg/Kg are rated as low in available K and the critical levels are between 39 to 156mg/Kg. This agrees to some extent to the results obtained in this study. Geoff *et al.*, (2013) after doing 76 experiments found the critical range of K in loam soils to be 44 to 74 mg/Kg with an average of 59 mg/Kg. These results agree with those found in this study which ranged between 21.8 to 66.867 mg/Kg before planting and 14.867 to 54.010 mg/Kg.

According to Iowa State University interpretation, soil test K optimum concentration is 90 to 130 mg/Kg (Clover *et al.*, 2007). This is way above the range found in this study indicating that all the sampled areas in this study are K deficient. Saifullah *et al.*, (2002), in a field experiment in Bhattian found the level of K before planting to be 86 mg/Kg. This concentration is much higher than the concentrations found this study. This

concentration is far much higher than those found in this study and the critical level is 80 mg/Kg. Rahman *et al.*, (2013) found the concentration of K in the soils of south Surma and Jalalpur to be 16.8 mg/Kg and 13.7 mg/Kg which is much lower than those found in this study. The mean difference between potassium concentration in the soil before and after planting in the whole area was significant ($P < 0.05$) indicating a decrease in potassium concentration after planting.

The concentration of Ca before planting was found to range between (0.320 to 4.542 mg/Kg) and (0.265 to 5.855 mg/Kg) after planting. The soil sample with the highest Ca concentration before planting was sample 1 (4.542 ± 0.053) which is from Ololulunga area and the lowest concentration was sample 7 (0.320 ± 0.029) which was also sampled from Ololulunga area. After planting the highest Ca concentration was sample 33 (5.855 ± 0.056) from Mau area and the least was sample 20 (0.265 ± 0.029) which was sampled from Ntulele area. Mau region had the highest average concentration before planting of 2.254 ± 0.922 , followed by Ntulele with a mean of 1.894 ± 1.003 and lastly Ololulunga with an average concentration of 1.733 ± 1.348 . After planting the same order was maintained with a mean of 3.091 ± 1.394 , 1.999 ± 0.864 and 1.566 ± 0.856 respectively. The average concentration of Ca in the soil before planting was found to be 1.972 ± 1.089 and 2.249 ± 1.25 after planting. The control sample had a concentration of 3.154 ± 0.089 which is higher than all the means from the three different areas with only 15.56% of all sampled areas before planting and the same percentage after planting having a concentration above that of the control. Ca exhibited a high level of variability of 59.62% in Ololulunga and 40% in Mau area but only a small variation of 24% in Ntulele. This can be due to the different application rates of Ca based fertilizers by farmers in the different areas. It can also be attributed to the difference in bedrock.

The soil samples were found to be slightly acidic during the two seasons with a mean pH of 6.413 ± 0.373 before planting and 6.336 ± 0.359 after planting with a range of 5.15 to 7.19 before planting and 5.44 to 7.22 after planting. This range according to Hoefl *et al.*, (2000) is not suitable for the availability of Ca and Mg. The availability of calcium

and magnesium is best when the pH is between 7.0 and 8.5 (Hoeft *et al.*, 2000). Buszweski *et al.*, (2000) in the soil of Torun found the concentration of Ca to be 309.42 mg/Kg during autumn and 122.66 mg/Kg during spring. Rahman *et al.*, (2013) found the concentration of Ca in the soils of south Surma and Jalalpur to be 68.8 mg/Kg and 82.5 mg/Kg which are all higher than those found in this study. Ca concentration found in the soils of wheat farms in Uasin Gishu district ranged between 179-738 mg/Kg and the critical level to be 400 mg/Kg. The samples analyzed from this study had a far much lower concentration as compared to the critical level for Ca therefore Ca based fertilizers should be added in order for farmers in Narok north to experience good yields. The mean difference between Ca concentration before and after planting was not significant ($P>0.05$).

The concentration of Mg before planting was found to range between (0.634 to 1.977 mg/Kg) and (0.544 to 1.728 mg/Kg) after planting. The highest concentration before planting was found in sample 3 (1.977 ± 0.038) and the lowest in sample 7 (0.634 ± 0.004) which were both from Ololulunga area. After planting sample 15 had the highest concentration (1.728 ± 0.067) and sample 4 had the lowest (0.544 ± 0.042). Ntulele had the highest concentration before planting of 1.403 ± 0.240 followed by Mau region with a mean of 1.296 ± 0.246 and lastly Ololulunga with a mean of 1.251 ± 0.373 . After planting Ololulunga had the highest concentration of 1.266 ± 0.374 followed by Ntulele with a mean of 1.133 ± 0.292 and finally Mau region with a mean of 1.081 ± 0.246 . The average concentration of Mg before planting was 1.274 ± 0.328 and 1.140 ± 0.298 after planting. The control had a concentration of 1.712 ± 0.008 which was higher than all the averages from all the sampled areas. 11% of the samples before planting were higher than the control and 7% after planting. Mg exhibited low levels of variability in all the sampled areas with a CV range of 22.05-27.8%. This might be due to the bedrock Mg concentration or the farmer applied the fertilizer in the recommended rates, hence little variation from farm to farm.

Mg concentration before and after planting had a significant mean difference ($P < 0.05$). After planting there was a mean increase in Mg concentration. Low pH has many adverse effects, including toxicities as well as low amounts of Ca and Mg (Yost and Uchida, 2000). Buszweski *et al.*, (2000) in the soil of Torun found the concentration of Mg to be 35.64 mg/Kg during autumn and 18.16 mg/Kg during spring. Rahman *et al.*, (2013) found the concentration of Mg in the soils of south Surma and Jalalpur to be 18.8 mg/Kg and 20.2 mg/Kg which are higher than those found in this study. This concentration is far much higher than those found in this study and the critical levels are reported to be 120 mg/Kg. the results found in this study are far much below the critical level hence Mg based fertilizers are highly required in order to experience high yields in the area.

Table 3.3 Concentrations of Ca, Mg and K in mg/Kg

S	Ca		Mg		K	
	Before planting	After planting	Before planting	After planting	Before planting	After planting
1	4.542±0.053	1.527±0.021	1.242±0.005	1.480±0.045	46.400±0.200	43.133±0.208
2	4.196±0.035	2.711±0.058	1.751±0.002	1.484±0.048	45.333±0.404	51.800±0.265
3	2.623±0.049	1.939±0.028	1.977±0.038	1.376±0.044	45.833±0.321	48.933±0.153
4	2.072±0.021	1.809±0.063	1.295±0.002	0.544±0.042	64.8±0.818	47.500±0.000
5	1.149±0.028	5.101±0.037	1.043±0.002	1.717±0.02	35.167±0.115	14.867±0.643
6	0.334±0.053	0.889±0.042	0.685±0.002	0.791±0.083	33.333±0.252	22.400±3.857
7	0.320±0.029	0.426±0.014	0.634±0.004	0.996±0.032	39.3±0.721	30.900±0.300
8	0.893±0.021	0.468±0.087	1.069±0.009	1.090±0.028	46.3±0.265	28.033±0.473
9	1.055±0.016	2.169±0.029	1.416±0.002	1.569±0.056	53.367±0.611	44.600±0.529
10	1.407±0.042	1.883±0.014	1.005±0.010	1.210±0.007	54.533±0.306	38.833±0.115
11	1.703±0.028	1.129±0.021	1.695±0.011	1.393±0.048	47.000±0.265	39.167±0.577
12	1.444±0.029	0.713±0.068	1.143±0.009	0.790±0.0322	42.533±0.252	30.433±0.451
13	0.815±0.042	1.989±0.029	1.023±0.008	1.092±0.044	47.667±0.462	35.967±1.365
14	1.023±0.014	1.392±0.058	1.395±0.010	1.528±0.046	54.567±0.115	43.767±0.058
15	2.780±0.029	3.256±0.014	1.393±0.015	1.728±0.067	28.200±0.265	31.633±0.153
16	1.102±0.045	1.800±0.028	0.756±0.061	1.143±0.032	53.867±0.115	43.300±0.200
17	1.148±0.028	1.541±0.042	1.033±0.027	1.133±0.029	52.267±0.808	39.500±0.265
18	2.198±0.035	2.059±0.035	1.783±0.009	0.957±0.019	44.233±0.153	38.200±0.100

S	Ca		Mg		K	
	Before planting	After planting	Before planting	After planting	Before planting	After planting
19	0.991±0.029	2.207±0.049	1.006±0.010	1.061±0.060	47.233±1.069	52.067±2.874
20	0.625±0.029	0.265±0.029	0.879±0.03	0.756±0.049	43.633±0.503	34.933±0.153
21	1.244±0.050	2.322±0.049	1.664±0.041	1.314±0.019	39.967±0.115	41.067±0.321
22	1.448±0.016	1.430±0.042	1.055±0.005	0.972±0.082	47.900±0.265	39.900±0.625
23	1.504±0.035	1.272±0.042	0.985±0.008	0.925±0.028	43.100±0.200	43.267±0.153
24	2.336±0.021	2.447±0.040	1.558±0.014	1.044±0.021	42.000±0.872	46.900±1.947
25	4.140±0.056	3.136±0.021	1.357±0.048	1.290±0.048	38.200±0.200	31.133±0.058
26	3.659±0.037	3.108±0.029	1.409±0.023	1.566±0.053	32.567±0.451	39.567±0.643
27	2.845±0.049	3.543±0.049	1.762±0.045	0.871±0.028	30.800±0.100	25.533±0.116
28	1.809±0.029	2.063±0.050	1.641±0.041	0.665±0.037	38.533±0.503	20.033±0.321
29	1.323±0.029	1.818±0.066	1.103±0.026	1.233±0.011	52.067±2.875	33.700±0.173
30	2.035±0.037	0.977±0.063	1.143±0.008	1.171±0.034	28.800±0.100	36.200±0.265
31	3.663±0.029	2.183±0.029	1.754±0.045	0.986±0.029	36.600±0.100	32.467±0.231
32	1.014±0.142	4.870±0.076	1.340±0.011	1.139±0.046	32.733±0.231	23.000±0.400
33	3.599±0.042	5.855±0.056	0.973±0.010	1.360±0.081	35.733±0.404	38.667±0.321
34	1.606±0.037	0.704±0.014	1.515±0.033	0.819±0.053	30.533±0.379	54.533±0.551
35	1.772±0.028	3.164±0.035	1.033±0.081	0.777±0.067	27.400±0.458	30.467±0.462
36	2.697±0.035	2.216±0.050	1.009±0.172	0.816±0.052	36.800±0.100	24.667±0.306
37	2.239±0.021	2.193±0.035	1.302±0.031	0.906±0.027	28.933±0.473	27.033±0.252
38	2.836±0.021	4.801±0.068	1.371±0.030	1.476±0.036	52.100±0.265	49.667±0.451
39	3.538±0.042	3.945±0.021	1.668±0.041	1.312±0.040	59.600±0.361	34.867±0.757
40	2.803±0.035	2.933±0.042	1.124±0.020	1.197±0.041	43.233±0.306	51.100±0.361
41	2.503±0.042	2.184±0.049	1.677±0.033	0.913±0.037	66.867±0.635	27.933±0.208
42	1.263±0.021	4.195±0.035	1.120±0.041	1.392±0.022	21.800±0.818	40.033±0.058
43	1.189±0.077	3.192±0.063	0.870±0.013	1.086±0.039	41.733±0.321	41.000±2.427
44	1.198±0.029	2.059±0.053	1.117±0.032	1.282±0.038	35.967±0.404	49.167±0.651
45	1.864±0.021	1.865±0.016	1.568±0.040	0.751±0.025	41.533±0.153	39.6±0.100
C	3.155±0.089	-	1.712±0.008	-	50.1±0.503	-

KEYS- Sample C- Control

3.1.4 Levels of Iron, Zinc and Copper

The concentration of Fe, Zn and Cu was measured from Ololulunga, Ntulele and Mau areas and the results tabulated in Table 3.4. The concentration of Cu before planting was found to range from (0.011 to 0.047) and (0.011 to 0.090) after planting. The highest concentration before planting was found in sample 5 (0.047 ± 0.006) which was from Ololulunga area and lowest concentration of 0.011 ± 0.000 was found in several samples. After planting the highest concentration was found in sample 26 (0.095 ± 0.002) and lowest concentration (0.011 ± 0.000) was found in several samples. Before planting, Mau had the highest average concentration of 0.0311 ± 0.026 followed by Ololulunga (0.023 ± 0.022) and finally Ntulele with 0.022 ± 0.009 . The same order was maintained after planting with Mau region having 0.028 ± 0.021 , Ntulele 0.027 ± 0.022 and Ololulunga 0.020 ± 0.009 . The average concentration of Cu before planting from all the sampled areas was 0.023 ± 0.01 and 0.025 ± 0.018 after planting. Cu exhibited high levels of variability for all the sampled areas with a CV of between 51.2% and 63.43%. This variation could be due to different application of pesticides which is Cu based in these areas.

In a study done by Nafuma *et al*, 2001, in the soils of Uasin Gishu found the concentration of Cu in the soil to range between 2-7 mg/Kg which is sufficient considering the critical level to be 1 mg/Kg. Javed *et al.*, (2011), found the concentration of Cu in the soil to be 6.90 ± 0.04 and Sharma *et al.*, (2003) found a range of 0.5 to 3.9 mg/Kg Cu in the soils of Rajasthan. These concentration are higher than all those found in this study. In a study done by Kumar and Babel, 2011 on the soils of Jhunjhunu Tehsil, the concentration of Cu ranged between 0.17 to 3.32 mg/Kg with a mean of 1.19 mg/Kg. This range is far much higher than those found in this study. Farshid, (2011) found the concentration of Cu in the soils of Abadeh Tashk, Iran to be 1.4 mg/Kg which is much higher than the concentrations found in this study. Buszweski *et al.*, (2000) in the soil of Torun found the concentration of Cu to be 9.69 mg/Kg during autumn and 5.03 mg/Kg during spring these concentrations are higher than those found in this study.

Sidhu and Sharma, (2010) reported that Cu concentration in the Trans-Gangetic plains ranged from 0.22 to 4.72 mg/Kg, 0.06 to 4.32 mg/Kg in the upper Gangetic plains, 0.09 to 7.80 in the middle Gangetic plains and 0.21 to 4.38 mg/Kg in the lower Gangetic plains. These concentrations were found to be higher than those found in this study even though some areas were found to be comparable. James and Topper, (1993) classified the concentration of Cu as 0-0.2 mg/Kg to be low and above 0.2 mg/Kg as high. This indicates that all the sampled areas are Cu deficient. The mean difference of Cu concentration before and after planting is not significantly different ($P > 0.05$).

The concentration of Fe before was found to range from 14.976 to 72.174 and 17.976 to 150.436 mg/Kg after planting. The highest concentration before planting was found in sample 15 and lowest in sample 7 both from Ololulunga area. After planting the lowest concentration was found in sample 1 from Ololulunga and highest in sample 20 from Ntulele area. Ntulele had the highest mean concentration of 49.533 ± 14.199 before planting followed by Mau (46.540 ± 12.657) and lastly Ololulunga with 37.294 ± 15.196 . After planting Ntulele had the highest concentration of 52.255 ± 29.762 , followed by Mau (38.147 ± 9.982) and finally Ololulunga with 30.294 ± 8.457 . The mean concentration of Fe was found to be 44.456 ± 14.940 before planting and 40.232 ± 20.512 after planting. The control had a mean of 21.296 ± 0.561 with only two samples having a concentration below the control before planting and three samples after planting.

Fe exhibited moderate levels of variability with Mau having the least CV of 24.52%, followed by Ololulunga with 37.95% and lastly Ntulele with 47.72%. This can be due to the fact that different farmers applied fertilizers at different rates and it could also be due to difference in the bedrock concentration of Fe. The concentration of Fe before and after planting did not have a significant mean difference ($P > 0.05$).

Iron is most available in acidic soils of pH not more than 6.5. Javed *et al.*, (2011) found the concentration of Fe in the soil to be 39.000 ± 0.040 . This concentration agrees with

an average concentration found in this study. In a study done by Kumar and Babel, 2011 on the soils of Jhunjhunu Tehsil, the concentration of Fe ranged between 1.22 to 5.87 mg/Kg with a mean of 3.30 mg/Kg. This range is far much lower than those found in this study. Sharma *et al.*, (2003) found a range of 1.0 to 6.6 mg/Kg Fe in the soils of Rajasthan which is also much lower than that found in this study. Sidhu and Sharma (2010) reported that Fe concentration in the Trans-Gingatic plains ranged from 1.05 to 97.9 mg/Kg, 3.48 to 90.20 mg/Kg in the upper Gangetic plains, 9.22 to 256.70 in the middle Gangetic plains and 3.60 to 182.5 mg/Kg in the lower Gingatic plains. These concentrations were found to agree to a greater extent with those found in this study. All the sampled areas in this study had adequate Fe concentration according to the classification done by James and Topper 1993, who classified Fe concentration in the soil as 0-3.0 mg/Kg as low, 3.1-5.0 mg/Kg as marginal and above 5.0 mg/Kg as adequate. Farshid (2011), found the concentration of Fe in the soils of Abdek Tashk, Iran to be 6.2 mg/Kg which is far lower compared to the ones found in this study. Buszweski *et al.*, (2000), found the concentration of Fe to be 234.88 mg/Kg during autumn and 206.99 mg/Kg during spring in the soil of Torun. These concentrations were much higher than those found in this study. In a study done by Nafuma *et al.*, (2001) in the soils of Uasin Gishu found the concentration of Fe in the soil to range between 25-50 mg/Kg which is sufficient considering the critical level to be 10 mg/Kg. Rahman *et al.*, (2013) found the concentration of Fe in the soils of south Surma and Jalalpur to be 178.4 mg/Kg and 144.5 mg/Kg which are higher than those found in this study.

The concentration of Zn before planting was found to range from (0.082 to 0.389) and (0.073 to 0.456 mg/Kg) after planting. The highest concentration was found in sample 2 from Ololulunga with a concentration of 0.389 ± 0.008 before planting and sample 43 (0.456 ± 0.015) after planting. The lowest concentration before planting was found in sample 13 and 18 with a concentration of 0.082 ± 0.006 and sample 1 after planting with concentration of 0.073 ± 0.004 mg/Kg. Before planting Mau region had the highest concentration of 0.201 ± 0.047 followed by Ntulele with 0.186 ± 0.039 mg/Kg and finally Ololulunga with 0.176 ± 0.093 mg/Kg. After planting the same order was

maintained with concentrations of 0.226 ± 0.081 , 0.191 ± 0.072 and 0.117 ± 0.032 mg/Kg respectively. The mean concentration of Zn before planting was found to be 0.187 ± 0.064 and 0.178 ± 0.078 mg/Kg after planting. The control had a concentration of 0.168 ± 0.007 mg/Kg with 56 % of the samples having a concentration above the control before planting and 49% after planting. Zn exhibited moderate levels of variability with a CV of 25.91% for Ntulele, 51.74% for Ololulunga and 35.05% for Mau. This can be due to the fact that different farmers applied fertilizers at different rates and it could be due to difference in the bedrock concentration of Zn.

Zn concentration of 0.5 mg/Kg is adequate for the production of wheat (Farnandez and Hoefft, 2012). This indicates that all the sampled areas had a Zn concentration below that reported by Farnandez and Hoefft, (2012). Javed *et al.*, (2011) found that the concentration of Zn to be 5.00 ± 0.02 . This concentration is higher than all those found in this study. In a study done by Kumar and Babel, 2011 on the soils of Jhunjhunu Tehsil, the concentration of Zn ranged between 0.12 to 1.30 mg/Kg with a mean of 0.51 mg/Kg. This range agrees to some extent with the results found in this study with 20% of the samples after planting falling below this range and only 11% of the samples before planting. Sharma *et al.*, (2003) found a range of 0.1 to 1.7 mg/Kg in the soils of Rajasthan and those results agrees to some extent with the range found in this study. Sidhu and Sharma, 2010 reported that Zn concentration in the Trans-Gingatic plains ranged from 0.11 to 5.08 mg/Kg, 0.04 to 2.53 mg/Kg in the upper Gangetic plains, 0.17 to 8.60 in the middle Gangetic plains and 0.04 to 3.46 mg/Kg in the lower Gingatic plains. These concentrations were found to agree to a greater extent with those found in this study. The critical Zn levels in the soil are between 0.6 and 2.0 mg/Kg (Farshid, 2011). This indicate that all the sampled areas in this study are Zn deficient. Lindsay and Norvell, (1978), stated the critical level of Zn in the soil to be 1.0 mg/Kg and Page *et al.*, 1983, classified the concentration of Zn in the soil as 0.0-0.5 mg/Kg as very low, 0.6-1.0 mg/Kg as low, 1.3-2.0 mg/Kg as medium and above 3.0 mg/Kg as high.

In a study done by Nafuma *et al.*, (2001) in the soils of Uasin Gishu found the concentration of Zn in the soil range between 4-6 mg/Kg which is sufficient considering the critical level to be 5 mg/Kg. According to the classification done by Page *et al.*, (1983) all the sampled areas are very low in Zn concentration. Topper *et al.*, (1993), classified 0 - 0.8 mg/Kg Zn concentration as low therefore all the sampled areas in this study are low in Zn concentration. Buszweski *et al.*, (2000) found the concentration of Zn to be 19.88 mg/Kg during autumn and 13.52 mg/Kg during spring in the soil of Torun and these concentrations are higher than those found in this study. Rahman *et al.*, 2013 found the concentration of Zn in the soils of south Surma and Jalalpur to be 0.75 mg/Kg and 0.66 mg/Kg which are higher than those found in this study. The mean difference in Zn concentration before and after planting was not significant ($P>0.05$).

Table 3.4 Concentration of Fe, Zn and Cu in mg/Kg

S	Fe		Zn		Cu	
	Before planting	After planting	Before planting	After planting	Before planting	After planting
1	50.948±0.743	17.731±0.743	0.338±0.005	0.073±0.004	0.011±0.000	0.015±0.006
2	58.725±0.743	34.744±1.012	0.389±0.008	0.134±0.004	0.022±0.001	0.033±0.001
3	37.985±0.972	38.957±0.972	0.238±0.010	0.076±0.005	0.032±0.000	0.033±0.001
4	43.170±0.281	33.286±0.972	0.160±0.003	0.142±0.006	0.033±0.000	0.011±0.000
5	40.253±2.192	19.999±0.486	0.156±0.005	0.165±0.003	0.047±0.006	0.023±0.000
6	34.096±1.286	31.989±1.123	0.107±0.006	0.101±0.004	0.026±0.006	0.011±0.000
7	14.976±0.561	30.694±0.486	0.142±0.005	0.145±0.007	0.015±0.006	0.018±0.006
8	27.615±2.013	27.453±1.707	0.082±0.008	0.101±0.004	0.022±0.000	0.011±0.000
9	19.351±1.012	40.091±0.281	0.281±0.008	0.090±0.005	0.033±0.000	0.026±0.006
10	23.564±0.743	28.587±0.743	0.107±0.006	0.174±0.006	0.026±0.006	0.011±0.000
11	34.258±1.012	21.944±0.486	0.157±0.004	0.108±0.005	0.033±0.000	0.011±0.000
12	35.717±0.743	18.055±0.486	0.143±0.005	0.087±0.006	0.011±0.000	0.026±0.006
13	24.050±0.561	46.410±1.563	0.082±0.006	0.144±0.022	0.011±0.000	0.011±0.000
14	42.522±0.743	27.939±1.965	0.137±0.003	0.116±0.005	0.011±0.000	0.032±0.000

S	Ca		Mg		K	
	Before planting	After planting	Before planting	After planting	Before planting	After planting
15	72.174±1.123	36.527±0.486	0.123±0.003	0.098±0.006	0.011±0.000	0.033±0.000
16	36.365±0.281	53.216±0.561	0.154±0.004	0.162±0.012	0.011±0.000	0.022±0.000
17	70.391±0.561	47.221±0.486	0.194±0.004	0.167±0.005	0.022±0.000	0.011±0.000
18	62.938±1.223	37.499±0.486	0.208±0.007	0.196±0.003	0.022±0.000	0.011±0.000
19	28.911±0.561	36.203±1.223	0.132±0.009	0.118±0.003	0.011±0.000	0.033±0.000
20	33.772±0.281	150.436±0.743	0.098±0.005	0.185±0.009	0.029±0.006	0.011±0.000
21	67.475±2.024	52.082±0.486	0.241±0.008	0.358±0.011	0.011±0.000	0.033±0.000
22	49.975±1.485	33.610±0.486	0.195±0.003	0.123±0.011	0.012±0.000	0.025±0.006
23	45.438±0.743	35.878±0.561	0.226±0.006	0.196±0.005	0.026±0.006	0.012±0.002
24	64.072±0.743	58.401±1.458	0.204±0.006	0.350±0.004	0.033±0.000	0.018±0.006
25	54.512±0.972	71.039±0.972	0.207±0.006	0.105±0.020	0.022±0.000	0.022±0.000
26	45.600±0.281	36.041±0.486	0.168±0.007	0.209±0.004	0.036±0.006	0.095±0.016
27	51.596±0.486	40.577±0.743	0.179±0.004	0.192±0.005	0.033±0.000	0.011±0.000
28	39.767±0.743	26.644±0.742	0.239±0.010	0.181±0.004	0.026±0.006	0.051±0.006
29	64.072±0.743	62.452±0.281	0.154±0.006	0.179±0.003	0.022±0.000	0.033±0.000
30	28.101±1.223	42.522±0.281	0.186±0.003	0.147±0.023	0.011±0.000	0.022±0.000
31	52.730±2.245	55.646±0.743	0.239±0.004	0.207±0.009	0.026±0.006	0.094±0.001
32	56.619±0.743	37.985±1.753	0.156±0.004	0.210±0.005	0.011±0.000	0.011±0.000
33	56.295±0.743	34.096±1.753	0.233±0.005	0.311±0.008	0.022±0.000	0.052±0.006
34	38.795±0.281	37.175±2.398	0.159±0.008	0.231±0.009	0.023±0.000	0.033±0.000
35	50.137±0.486	33.124±0.486	0.163±0.003	0.190±0.011	0.015±0.006	0.018±0.006
36	48.679±3.188	60.021±1.707	0.285±0.014	0.236±0.005	0.029±0.006	0.022±0.000
37	68.123±0.486	46.734±0.842	0.202±0.003	0.164±0.010	0.011±0.000	0.015±0.006
38	32.152±0.972	31.828±2.398	0.190±0.008	0.140±0.004	0.040±0.006	0.022±0.000
39	42.340±0.486	37.499±1.286	0.270±0.006	0.198±0.009	0.033±0.000	0.018±0.006
40	23.402±0.842	31.990±0.281	0.189±0.009	0.140±0.007	0.011±0.000	0.033±0.000
41	47.059±0.743	25.995±0.743	0.211±0.004	0.215±0.009	0.018±0.006	0.018±0.006
42	26.805±1.286	41.874±0.486	0.123±0.024	0.311±0.013	0.011±0.000	0.023±0.000
43	40.416±0.486	39.767±0.743	0.1428±0.003	0.456±0.015	0.036±0.006	0.018±0.006
44	62.128±0.743	36.041±0.972	0.249±0.017	0.208±0.007	0.033±0.000	0.033±0.000
45	52.406±0.743	22.430±2.119	0.200±0.005	0.174±0.019	0.033±0.000	0.011±0.000
C	21.299±0.561	-	0.168±0.0078	-	0.139±0.012	-

KEY:: S- Sample C- Control

3.1.5 Levels of Nitrate, Phosphate, and Sulfate in the wheat growing soils

The concentration of nitrates, phosphates and sulfates was determined from Ololulunga, Ntulele and Mau and the results tabulated as shown in table 3.5. The concentration of nitrates before planting was found to range between (11.108 to 32.441 mg/Kg) and (21.363 to 45.741 mg/Kg) after planting. The soil sample with the highest nitrates concentration before planting was sample 10 (32.441 ± 0.052) which is from Ololulunga area and the lowest concentration was sample 7 (11.108 ± 0.002) which was also sampled from Ololulunga area. After planting the highest nitrates concentration was sample 27 (45.741 ± 0.004) from Mau area and the least was sample 30 (21.363 ± 0.008) which was sampled from Ntulele area. Mau region had the highest average concentration before planting of 18.947 ± 2.700 , followed by Ololulunga with a mean of 18.421 ± 4.969 and lastly Ntulele with an average concentration of 18.010 ± 2.389 . After planting Ntulele had the highest mean concentration of 28.308 ± 5.922 , followed by Mau with a mean of 26.605 ± 3.169 and lastly Ololulunga with 26.170 ± 4.297 . The average concentration of nitrates in the soil before planting was found to be 18.459 ± 3.484 and 27.027 ± 4.593 after planting. The control sample had a concentration of 35.147 ± 0.078 which is higher than all the means from the three different areas with only one sample after planting and none before planting having a concentration above that of the control. Nitrates exhibited very low levels of variability of between 6.71% and 14.23% for all sampled areas.

Heckman 2003, categorized soil test levels of NO_3^{2-} to be deficient if the concentration is below 20 mg/Kg and sufficient if the concentration is between 20 and 24 mg/Kg but fertilizers can be added. A concentration above 30 mg/Kg is considered sufficient and fertilization not required but concentration above 50 mg/Kg is excessive (Heckman 2003). According to this categorization, most of the samples before planting were N deficient but after planting, they were all sufficient. This is most likely due to addition of

nitrogen based fertilizers during planting. Vanek *et al.*, 2003 found the concentration of NO_3^{2-} in the soil from farms to be between 9 mg/Kg and 14 mg/Kg. This range agrees with the range in this study before planting and way below those after planting. Before and after planting all the sampled areas had a nitrate concentration above the critical levels as stated by Dennis and John, (2003) of 15 mg/Kg. The mean difference in nitrate concentration before and after planting was significant ($P < 0.05$). There was lower nitrate concentration before planting than there was after planting in the whole area.

The concentration of phosphates before planting was found to range from 0.352 to 3.247 mg/Kg and 2.092 to 9.503 mg/Kg after planting. The soil sample with the highest phosphates concentration before planting was sample 44 (3.247 ± 0.023 mg/Kg) which is from Mau area and the lowest concentration was sample 32 (0.352 ± 0.001 mg/Kg) which was also sampled from Mau area. After planting the highest phosphates concentration was sample 3 (9.508 ± 0.006 mg/Kg) from Ololulunga area and the least was sample 40 (2.092 ± 0.008 mg/Kg) which was sampled from Mau area. Ololulunga region had the highest average concentration before planting of 1.153 ± 0.823 , followed by Mau with a mean of 0.903 ± 0.789 and lastly Ntulele with an average concentration of 0.730 ± 0.206 . After planting, Ololulunga had the highest phosphate mean concentration of 4.857 ± 1.923 , followed by Mau with a mean of 3.086 ± 0.615 and lastly Ntulele with 2.922 ± 0.551 . The average concentration of phosphates in the soil before planting was found to be 0.928 ± 0.006 and 3.622 ± 1.475 after planting. The control sample had a concentration of 1.594 ± 0.000 which is higher than all the means before planting but lower than all the means after planting from the three different areas. All the samples after planting had concentration above the control. Only three samples before planting had a concentration above the control.

Phosphates variability ranged between 21.91 and 40.88% with Ololulunga having the highest and Ntulele having the least. Phosphates varied from farm to farm maybe due to the variable farm tillage methods and amounts of fertilizer applied. Courteney *et al.*, (2013) classified the concentrations of P in the soil as: below 8 mg/Kg as low, 8-16

mg/Kg as medium and above 16 mg/Kg as high. According to this classification all the sampled areas before and even after planting in this study are deficient in P with only one sample in the medium range. Micheni *et al.*, (2001) in the soils of Eastern Kenya found the concentration P to be 13.9 mg/Kg which they classified as low. This concentration is far much higher than those found in this study and this low concentration can be associated with soil P mining through farming activities. P concentration found in the soils of wheat farms in Uasin Gishu district ranged between 7-11 mg/Kg (Nafuma *et al.*, 2001). This concentration is far much higher than those found in this study. The critical values for P is 20 mg/Kg (Nafuma *et al.*, 2001). All the sampled areas in this study are therefore P deficient. Rahman *et al.*, (2013) found the concentration of P in the soils of south Surma and Jalalpur to be 6.6 mg/Kg and 5.9 mg/Kg which are higher than those found in this study. Before and after planting all the sampled areas had a phosphate concentration below the critical levels as stated by Dennis and John, (2003) of 25 mg/Kg. The mean difference in phosphate concentration before and after planting was significant ($P < 0.05$) There was lower phosphate concentration before planting than there was after planting.

The concentration of sulphate before planting was found range from 2.003 to 6.678 mg/Kg and 4.111 to 8.650 mg/Kg after planting. The soil sample with the highest sulfates concentration before planting was sample 2 (6.678 ± 0.000) which is from Ololulunga area and the lowest concentration was sample 45 (2.003 ± 0.000) which was sampled from Mau area. After planting the highest sulphates concentration was sample 20 (8.65 ± 0.000) from Ntulele area and the least was sample 5 (4.111 ± 0.000) which was sampled from Ololulunga area. Ololulunga region had the highest average concentration before planting of 5.199 ± 0.805 , followed by Ntulele with a mean of 4.682 ± 0.872 and lastly Mau with an average concentration of 3.755 ± 0.881 . After planting, Ntulele had the highest mean concentration of 6.712 ± 1.011 , followed by Mau with a mean of 6.461 ± 0.615 and lastly Ololulunga with 6.432 ± 0.966 . The average concentration of sulfates in the soil before planting was found to be 4.545 ± 1.030 and 6.497 ± 1.040 after planting. The control sample had a concentration of 2.939 ± 0.053

which is lower than all the means before planting and after planting from the three different areas with only one sample having a concentration below the control. Sulfates exhibited low levels of variability ranging between 8.54% and 16.54% with Ololulunga having the least and Mau having the highest. The mean difference in sulphates concentration before and after planting was significant ($P < 0.05$). There was lower sulphates concentration before planting than there was after planting.

Table 3.5 Concentration of Nitrate, Phosphate and Sulphate in mg/Kg

S	Nitrate		Phosphate		Sulphate	
	Before planting	After planting	Before planting	After planting	Before planting	After planting
1	16.928±0.002	22.348±0.001	0.429±0.007	4.938±0.032	6.393±0.006	7.361±0.015
2	17.350±0.005	26.110±0.000	0.839±0.001	3.421±0.018	6.678±0.000	7.453±0.051
3	14.657±0.001	23.204±0.004	3.001±0.006	9.508±0.006	4.913±0.003	5.221±0.011
4	19.024±0.023	24.417±0.004	1.138±0.004	4.010±0.008	6.007±0.012	6.017±0.000
5	16.733±0.003	26.424±0.006	2.077±0.006	2.958±0.001	3.661±0.016	4.111±0.000
6	14.853±0.010	21.646±0.005	0.930±0.016	2.357±0.105	5.040±0.059	6.944±0.000
7	11.108±0.002	22.555±0.006	1.035±0.001	5.917±0.008	5.005±0.000	6.301±0.000
8	20.955±0.039	26.309±0.002	0.368±0.005	4.008±0.006	5.538±0.011	8.028±0.000
9	24.088±0.008	33.314±0.001	2.642±0.028	6.526±0.006	5.157±0.029	6.051±0.000
10	32.441±0.052	36.759±0.006	1.058±0.010	5.028±0.006	5.293±0.000	6.700±0.019
11	16.111±0.003	23.459±0.017	0.627±0.016	3.128±0.006	5.480±0.011	7.272±0.000
12	17.065±0.012	25.566±0.006	1.686±0.011	4.017±0.005	4.408±0.013	6.515±0.000
13	15.189±0.001	24.423±0.004	0.429±0.007	7.073±0.001	5.388±0.021	6.366±0.058
14	19.039±0.005	25.034±0.003	0.484±0.010	3.530±0.008	4.994±0.003	6.299±0.001
15	20.777±0.011	30.973±0.005	0.547±0.002	6.432±0.009	4.022±0.001	5.842±0.000
16	16.927±0.001	25.111±0.001	0.685±0.011	2.147±0.007	6.213±0.000	7.195±0.027
17	17.720±0.001	22.683±0.003	0.387±0.001	2.461±0.005	5.544±0.014	7.109±1.088
18	15.757±0.001	28.038±2.823	0.675±0.006	3.019±0.007	3.616±0.010	6.988±0.000
19	15.899±0.009	29.315±2.750	0.911±0.005	2.913±0.013	4.130±0.006	7.776±0.000
20	19.060±0.001	27.096±0.005	0.819±0.005	2.736±0.014	4.042±0.010	8.565±0.000
21	19.868±0.003	23.445±0.006	0.822±0.005	2.473±0.000	5.111±0.001	7.509±0.016
22	18.441±0.001	32.415±0.009	0.585±0.012	3.129±0.006	3.021±0.000	4.407±0.007
23	16.424±0.003	26.561±0.010	1.086±0.001	3.968±0.030	4.411±0.016	6.190±0.003
24	20.025±0.001	34.425±0.005	1.040±0.001	2.949±0.030	5.016±0.006	6.017±0.000
25	17.498±0.001	25.631±0.006	0.401±0.001	4.019±0.001	5.017±0.000	7.323±0.000

S	Nitrate		Phosphate		Sulphate	
	Before planting	After planting	Before planting	After planting	Before planting	After planting
26	16.414±0.001	29.177±0.001	0.697±0.004	2.425±0.006	5.551±0.000	6.100±0.000
27	24.333±0.001	45.741±0.004	0.634±0.001	2.518±0.008	4.331±0.000	6.296±0.013
28	19.992±0.003	27.541±0.007	0.879±0.006	2.632±0.022	5.771±0.068	6.773±0.004
29	14.853±0.008	26.082±0.010	0.538±0.003	2.920±0.021	4.259±0.003	5.453±0.016
30	16.936±0.007	21.363±0.008	0.793±0.001	3.518±0.005	4.200±0.000	6.976±0.000
31	17.899±0.003	24.186±0.013	0.395±0.006	2.889±0.015	4.704±0.0042	7.735±0.005
32	17.967±0.004	24.096±0.001	0.352±0.001	3.012±0.012	4.821±0.000	6.361±0.009
33	20.411±0.006	34.324±0.004	0.367±0.002	2.924±0.003	3.839±0.008	4.141±0.011
34	19.032±0.001	28.200±0.000	0.737±0.002	2.532±0.006	3.309±0.009	7.849±0.019
35	17.889±0.001	28.717±0.010	0.366±0.001	3.293±0.007	5.181±0.017	7.309±0.009
36	13.806±0.004	25.527±0.008	0.378±0.002	3.032±0.006	3.955±0.000	5.362±0.048
37	17.095±0.004	27.091±0.000	0.735±0.001	4.045±0.024	4.352±0.022	6.140±0.001
38	16.317±0.003	23.414±0.003	0.985±0.012	4.291±0.010	4.132±0.001	6.722±0.001
39	21.654±0.001	25.126±0.001	2.133±0.007	2.951±0.006	3.804±0.007	5.117±0.002
40	19.990±0.017	24.516±0.038	0.663±0.004	2.092±0.001	3.864±0.034	7.115±0.001
41	20.347±0.009	27.858±0.003	0.499±0.004	2.808±0.004	3.170±0.000	5.963±0.000
42	25.033±0.001	31.688±0.041	0.843±0.009	2.519±0.006	2.231±0.000	5.241±0.000
43	21.360±0.001	26.818±0.004	0.863±0.006	2.636±0.011	3.202±0.001	7.408±0.000
44	19.050±0.001	23.573±0.011	3.247±0.023	3.247±0.005	3.744±0.000	7.680±0.003
45	16.347±0.002	23.944±0.006	0.981±0.011	4.022±0.006	2.003±0.000	5.075±0.001
C	35.147±0.078	-	1.594±0.000	-	2.940±0.053	-

KEY: S- Sample C- Control

3.1.6 Microbial assays

The morphological analysis revealed that most of the isolates before planting were gram positive cocci bacteria (Plate 3.1) with only isolate 43 being gram positive rod. After

planting all the isolates were gram positive rod shaped bacteria (Plate 3.2) except isolate from sample 2 which was gram negative rod (Plate 3.3) and isolate from sample 42 which was gram positive cocci. The change in cell wall characteristic and the shape of the bacteria between the two seasons may be due to the addition of fertilizers, pesticides and herbicides suppressing the growth of some bacteria and supporting others. In this study the gram positive bacteria constituted the majority of the species in the soil and this perfectly agrees with the study done by Amna and Fozia, (2011) in the soils of Pakistan and that done by Suchi *et al.*, (2013) in Kaziranga National Park Assam, India. After the planting season, the results obtained were in agreement with the study done by Tanu and Hoque, (2013) in the soils collected from various area in Bangladesh, in which they found the majority of the bacteria being gram positive rod shaped.



Plate 3.1 Gram positive cocci

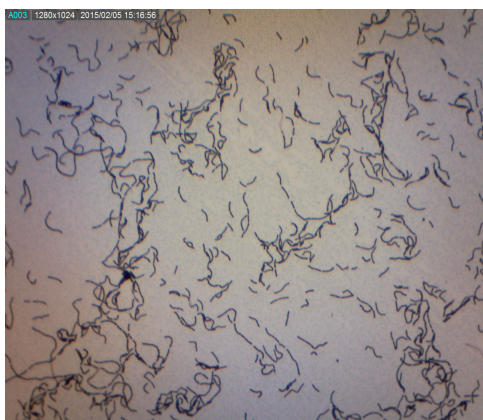


Plate 3.2 Gram positive rod

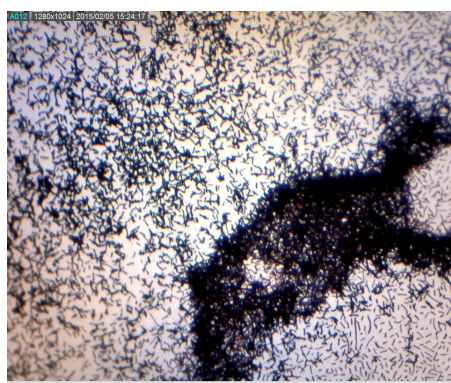


Plate 3.3 Gram negative rods



Plate 3.4 Gram positive rods

In this study all the isolates before and after planting gave a negative results for the indole biochemical test (Plate 3.5) thus the tryptophan did not break down into indole. This indole test was further confirmed by a test carried out using trypton water which gave negative result for all the isolates (Plate 3.6). These results are in agreement with the research done by Tanu and Hoque, (2013), Jitendra *et al.*, (2014), Amna and Fozia, (2011) where they also found negative results for all the isolates for the indole test to be negative. Saliu, (2009) in a study done on a cassava dumping site in Nigeria isolated a bacteria which was indole negative, this is in agreement with the results found in this study. Joshi *et al.*, (2007), Anam and Zakia, (2012) found indole positive isolates which is contrary to the result in this study.

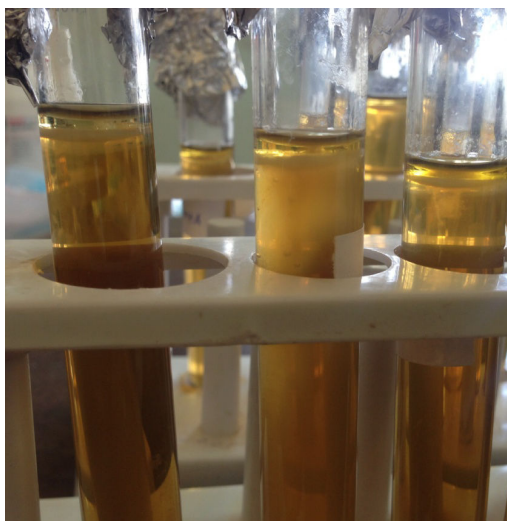


Plate 3.5 Sulphur Indole Test (all negative)



Plate 3.6 Trypton water test

Isolates from almost all the samples before planting with an exception of isolates from samples 10, 43 and 45 were urease negative (Plate 3.7) an indication that they are unable to hydrolyze urea to simpler forms of nitrates which can be readily absorbed by plants to promote growth. A positive result (Plate 3.7) was shown by isolate from samples 10, 43 and 45 implying an important aspect in growth and development of wheat as the bacteria

has shown a potential to convert urea to simpler forms which are readily absorbed by plants. This study is in agreement with the study done by Saliu, (2009) and Jitendra *et al.*, (2014) where they isolated bacteria that are urease negative. Several workers have found out that most of the isolates were urease negative (Amna and Fozia, 2011, Anam and Zakia, 2012) which is in agreement with the current study. Most of the results in this study do not agree with the results found by Joshi *et al.*, (2007) where they found urease positive isolates.



Plate 3.7 Urease test showing positive (pink) and negative (yellow)

Before planting all the isolated bacteria in the samples tested positive in the oxidase test (Plate 3.8) indicating the presence of respiratory enzyme cytochrome oxidase but after planting eleven isolates tested positive and the remaining four (9, 41, 44 and 45) tested negative (Plate 3.8). The results in this study are in agreement with the results found by Tanu and Hoque, (2013) in the soils of Bangladesh where nine of their isolates were oxidase positive and six were oxidase negative. Saliu, (2009) also isolated an oxidase negative bacteria which agrees with some of the results found in this study. Lubanze *et al.*, (2014) in a study done in Malelwane farm North west South Africa found most of their isolates to be oxidase positive and a few oxidase negative bacteria and also Amna and Fozia (2011) found similar results which agrees perfectly with the results in this

study. Anam and Zakia, (2012) found all of their isolates being oxidase negative which does not agree with the results found in this study.



Plate 3.8 Oxidase discs showing positive (blue) and a negative result (white)

In this study all the samples before and after planting showed a positive result by turning the citrate agar from green to royal blue (Plate 3.10 and 3.11) an indication that the isolated bacteria are able to use citrate as their sole source of metabolism and growth. Several workers found most of their isolates to be citrate positive (Tanu and Hoque, 2013 and Lubanze *et al.*, 2014), this agrees with the results found in this study. This study is also in agreement with the study done by Saliu, (2009) and Joshi *et al.*, (2007) where they isolated citrate positive bacteria. The results in this study do not agree with the results found by Amna and Fozia, (2011), Anam and Zakia, (2012) and Jitendra *et al.*, (2014) where they found most of the isolates to be citrate negative.



Plate 3.9 Slanted citrate agar before inoculation



Plate 3. 10 Samples showing positive citrate



Plate 3.11 Several samples showing positive results

Before planting and after planting all the isolates in this study did not show any signs of production of hydrogen sulfide which is in agreement with the results found by Lubanza *et al.*, (2014), Joshi *et al.*, (2007), Amna and Fozia, (2011), Anam and Zakia, (2012). Only isolate from sample 45 before and after planting showed signs of a gas production. Before planting all the isolates except isolate from sample 45 showed negative results for all the TSI tests which agree with the results found by Jitendra *et al.*, (2014) where they isolated bacteria that tested negative for all the TSI tests. After planting, isolates from seven samples (9, 16, 18, 21, 30, 44 and 45) showed a yellow colour change on the

slant with only isolates from two samples- (30 and 45) turning yellow at the bottom (Plate 3.13) and this agrees with the study done by Suchi *et al.*, (2013) where they isolated pure culture of bacteria which was able to ferment glucose, lactose and sucrose but did not produce hydrogen sulfide.



Plate 3.12 TSI media
before inoculation



Plate 3.13 TSI results (both
positive and negative)

All the isolates before and after planting were found to be catalase positive since they produced bubbles when put in hydrogen peroxide indicating the presence of enzyme catalase which catalyses the breakdown of hydrogen peroxide to release free oxygen which is an important aspect required by bacteria to reproduce avoiding cellular toxicity (Cappuccino & Sherman, 2002). These results are in agreement with those found by Suchi *et al.*, (2013) where they isolated catalase positive bacteria in a study done in Kiziranga National Park, India. Anam and Zakia, (2012), Jitendra *et al.*, (2014) and Lubanza *et al.*, (2014) in studies done in Molelwane Farm north West Province of South Africa and found all the isolates to be catalase positive, this is in agreement with this study where all the isolates were also positive. The results also agree with those found by Joshi *et al.*, (2007) where they found catalase positive bacteria. The results found in

this study do not agree with those found by Amna and Fozia, (2011) who found most of their isolates to be catalase negative.

All the isolates before planting, tested negative for gelatinase which is in agreement with the results found by Jitendra *et al.*, (2014). After planting isolates from only 3 samples (16, 18, and 21) tested positive. These results agree with those found by Amna and Fozia, (2011) in the soils of Lahore and Changa Manga where they found most of their isolates being gelatinase negative. Tanu and Hoque, (2013) in a study done on the soils collected from various areas in Bangladesh found all their isolates to be gelatinase positive this disagree with the results found in this study. Saliu, (2009) in a study done in a cassava dumpsite in Nigeria and Joshi *et al.*, (2007) were able to isolate gelatinase positive bacteria which does not agree with the result found in this study.

Before and after planting all the samples tested negative for methyl red. This is in agreement with the study done by Munees and Mohammad, (2011) in a study done in the soils of Uttar, India and Anam and Zakia, 2012 which they isolated bacteria which tested negative for Methyl red. Tanu and Hoque, 2013, Saliu, 2009, Joshi *et al.*, (2007), Jitendra *et al.*, (2014) and Lubanza *et al.*, (2014) also found most of their isolates to be methyl red negative. Ogot *et al.*, (2013) in a study done on the soil collected from JKUAT farm found positive results for the bacteria isolated which disagree with the results found in this study.

Only isolate from sample 30 after planting and isolate from sample 45 before planting tested positive for Voges Proskauer. These results are in agreement with the results found by Tanu and Hoque, (2013), Ogot *et al.*, (2013), Joshi *et al.*, (2007), Lubanza *et al.*, (2014), Amna and Fozia, (2011), Jitendra *et al.*, (2014) and Saliu, (2009) where they found their isolates to be Voges Proskauer negative. Anam and Zakia, (2012), isolated bacteria from various source with most of them were Voges Proskauer negative with a few positive and this was is in agreement with this study.

The bacteria isolated in this study were identified with the help of the Bergey's manual of determinative bacteriology and found to be majorly *micrococcus spp* before planting and *Bacillus spp*, *Pseudomonas spp*, *Mycobacterium spp* and *Corynebacterium spp* after planting. This is in agreement with a study done by Tanu and Hoque, 2013; Laila *et al.*, 2011 and Sargervanshi *et al.*, 2012 where they found both *Bacillus spp* and *Pseudomonas spp* to be very common in agricultural soils. Teli *et al.*, 2013 also found *Micrococcus spp* and *Pseudomonas spp* abundant in garage soil. Najia *et al.*, 2012 isolated *Pseudomonas spp* in agricultural soil treated with organophosphates.

The abundance of the *Bacillus spp* in the samples of soil after planting was possibly due to their spore formation which increases their resistance against stress like pesticides, herbicides and fertilizers. This is supported by studies done by Sagardoy and Salerno, 1983; Malik *et al.*, 2002. In the current study, the presence of *Pseudomonas spp* was found to exist even after application of pesticides and herbicides and this can be correlated with the results obtained by Dasai *et al.*, 2008 who found the presence of *Pseudomonas spp* in stress conditions. Members of the *Bacillus* genus generally have the ability to disintegrate proteins. They contain protease enzyme which play an important role in the nitrogen cycle which contribute to the fertility of the soil (Belma *et al.*, 2002). This property is very important for wheat farming because nitrogen is highly required alongside other nutrients. Most of the *Pseudomonas spp* contain strains that can suppress plant diseases (McSpadden, 2007) hence their suitability for the growth of wheat. This property is very important in wheat farming because wheat is very prone to various plant diseases hence the presence of *Pseudomonas spp* help in suppressing the diseases therefore farmers might not be required to use pesticides to in their wheat farm.

Table 3.6 Summary of the morphological and biochemical tests carried out on representative samples before planting

Sam ple	Shape	Gram	TSI				Citrat e	Urea	MR	VP	Gela tin	Trypton	SIM motility	Indo le	Catal ase	Oxid ase
			B	SL	G	H ₂ S										
2	Cocci	+	-	-	-	-	+	-	-	-	-	-	+	-	+	+
3	Cocci	+	-	-	-	-	+	-	-	-	-	-	+	-	+	+
4	Cocci	+	-	-	-	-	+	-	-	-	-	-	+	-	+	+
9	Cocci	+	-	-	-	-	+	-	-	-	-	-	-	-	+	+
10	Cocci	+	-	-	-	-	+	+	-	-	-	-	-	-	+	+
16	Cocci	+	-	-	-	-	+	-	-	-	-	-	-	-	+	+
18	Cocci	+	-	-	-	-	+	-	-	-	-	-	-	-	+	+
21	Cocci	+	-	-	-	-	+	-	-	-	-	-	-	-	+	+
23	Cocci	+	-	-	-	-	+	-	-	-	-	-	-	-	+	+
30	Cocci	+	-	-	-	-	+	-	-	-	-	-	+	-	+	+
41	Cocci	+	-	-	-	-	+	-	-	-	-	-	+	-	+	+
42	Cocci	+	-	-	-	-	+	-	-	-	-	-	-	-	+	+
43	Rod	+	-	-	-	-	+	+	-	-	-	-	-	-	+	+
44	Cocci	+	-	-	-	-	+	-	-	-	-	-	-	-	+	+
45	Cocci	+	+	+	+	-	+	+	-	+	-	-	+	-	+	+

Table 3.7 Summary of the morphological and biochemical tests carried out on representative samples after planting

Sam ple	Gram	Shape	TSI				Citrat e	Ure a	MR	VP	Gela tin	Tryp ton	SIM motility	Indo le	Catal ase	Oxid ase
			B	SL	G	H ₂ S										
2	-	Rod	-	-	-	-	+	+	-	-	-	-	+	-	+	+
3	+	Rod	-	-	-	-	+	+	-	-	-	-	-	-	+	+
4	+	Rod	-	-	-	-	+	+	-	-	-	-	-	-	+	+
9	+	Rod	-	+	-	-	+	-	-	-	-	-	-	-	+	-
10	+	Rod	-	-	-	-	+	-	-	-	-	-	-	-	+	+
16	+	Rod	-	+	-	-	+	-	-	-	+	-	-	-	+	+
18	+	Rod	-	+	-	-	+	-	-	-	+	-	-	-	+	+
21	+	Rod	-	+	-	-	+	-	-	-	+	-	-	-	+	+
23	+	Rod	-	-	-	-	+	-	-	-	-	-	-	-	+	+
30	+	Rod	+	+	+	-	+	+	-	+	-	-	+	-	+	+
41	+	Rod	-	-	-	-	+	+	-	-	-	-	-	-	+	-
42	+	Cocci	-	-	-	-	+	-	-	-	-	-	+	-	+	+
43	+	Rod	-	-	-	-	+	+	-	-	-	-	-	-	+	+
44	+	Rod	-	-	-	-	+	-	-	-	-	-	-	-	+	-
45	+	Rod	-	+	-	-	+	-	-	-	-	-	-	-	+	-

3.1.7 Correlation analysis

Calcium and zinc had the strongest correlation (0.642) as shown in Table 3.8. Magnesium and calcium had a positive correlation (0.513). Those with lower correlation were iron and calcium (0.356), zinc and iron (0.371), iron and magnesium (0.344) and copper and phosphates (0.452). The pH showed no correlation before planting with all the nutrients analyzed.

After planting there was very little correlation among all the nutrients except Ca and Mg which had a low positive correlation (0.413) as shown in Table 3.9 indicating a possible similar source of input for these nutrients. Those with a negative correlation were PO_4^{2-} with Ca (-0.454) and PO_4^{2-} and Zn (-0.455). The pH showed no correlation with all the nutrients except Ca which showed a positive correlation of 0.429 indicating possibility of availability of Ca as pH increased.

TABLE 3.8 Correlation analysis of the nutrients in the soil before planting

	K	Mg	Cu	Ca	Zn	Fe	NO_3^{2-}	SO_4^{2-}	PO_4^{2-}	pH
K	1									
Mg	0.089	1								
Cu	0.132	0.219	1							
Ca	-0.031	0.513	0.123	1						
Zn	0.096	0.465	0.138	0.642	1					
Fe	-0.159	0.344	-0.024	0.356	0.371	1				
NO_3	0.077	0.114	0.064	0.022	-0.093	-0.098	1			
SO_4^{2-}	0.24	0.021	-0.079	0.127	0.17	-0.025	-0.103	1		
PO_4^{2-}	0.124	0.143	0.452	-0.13	0.228	-0.025	0.076	-0.103	1	
pH	0.264	0.248	0.099	0.19	0.057	-0.112	0.048	-0.034	-0.029	1

TABLE 3.9 Correlation analysis of the nutrients in the soil after planting

	K	Mg	Cu	Ca	Zn	Fe	NO ₃ ²⁻	SO ₄ ²⁻	PO ₄ ²⁻	pH
K	1									
Mg	0.203	1								
Cu	0.088	0.239	1							
Ca	-0.122	0.413	0.182	1						
Zn	0.039	-0.119	0.12	0.357	1					
Fe	-0.018	-0.185	-0.012	-0.154	0.19	1				
NO ₃ ²⁻	-0.053	-0.009	0.023	0.3	0.232	0.03	1			
SO ₄ ²⁻	0.232	-0.193	-0.014	-0.455	-0.06	0.288	-0.253	1		
PO ₄ ²⁻	0.119	0.242	-0.134	-0.454	-0.455	-0.128	-0.079	-0.191	1	
pH	-0.056	-0.107	0.149	0.429	0.157	0.126	0.133	-0.244	-0.121	1

3.1.8 Dendograms

The dendogram (Figure 3.14) showing Ca, Mg and K showed that the sampled farms cannot be grouped into definite clusters. This can be a clear indication that the farmers in these areas applied the fertilizers in the different ways. The dendogram (Figure 3.15) shows concentrations of nitrates, phosphates and sulfates in the sampled farms. The farms can majorly be grouped into three groups with same trend of concentration, with a one group having up to 13 farms and this might be an indication that the farmers in these areas applied the fertilizers in the same way or they applied the same type of fertilizers. Only six farms had a high difference in the nutrients application as compared to the other thirty nine farms.

The dendogram (Figure 3.16) shows concentrations of Zn, Cu and Fe in the sampled farms. The sampled farms can be grouped into two groups with largest group having up to twenty farms and second largest having seventeen. This is a clear indication

that the minerals concentrations are due to the bedrock concentrations. Almost all farms had a very small concentration difference and this can be attributed to bedrock. The dendogram (Figure 3.17) shows concentrations of all the parameters analyzed in the farms. The sampled farms could not be grouped into groups except one cluster of 10 farms which seemed to have a close relation in these parameters while others were individually grouped or were in groups of three and two. This might be an indication that the farmers in these areas applied the nutrients and minerals in different ways probably according to their financial capabilities.

The dendogram (Figure 3.18) shows bacteria isolated from the representative farms. The sampled farms could be grouped into two groups with one cluster having 10 farms which seemed to have close relation in the isolates and another cluster had four farms. This shows that bacteria in these farms were very similar in the farms before planting may be due to the similar environmental conditions. The dendogram (Figure 3.19) shows bacteria isolated from the representative farms. The sampled farms could not be grouped into definite groups. The farms were individually grouped or were in groups of threes and twos. This shows that after planting the bacteria in these farms were different from each other maybe due to the application of fertilizers, pesticides and herbicides which might have suppressed the existence of some bacteria and favored others.

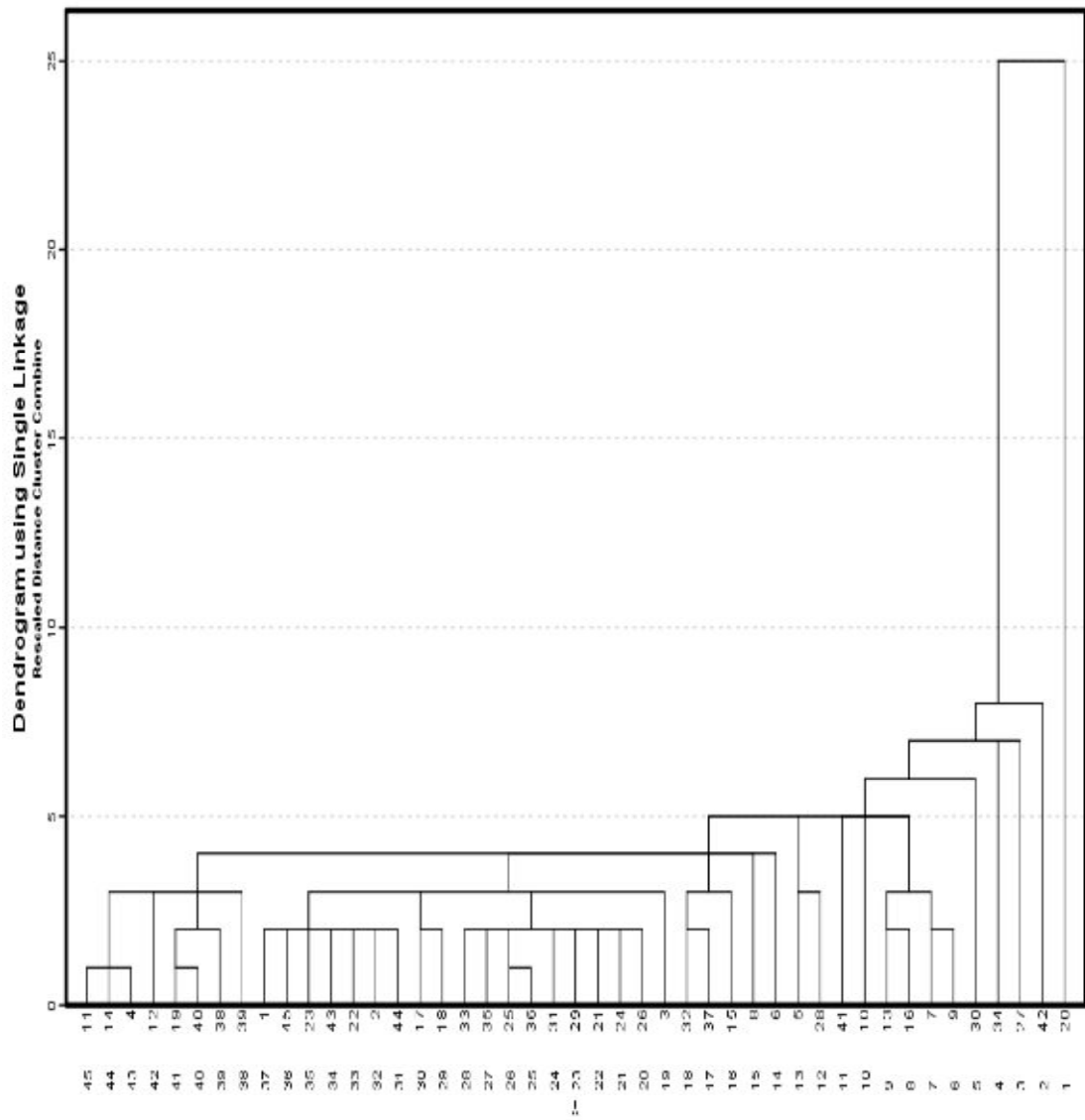


Figure 3.1 A dendrogram showing the clustering of Ca, Mg and K in all the sampled areas

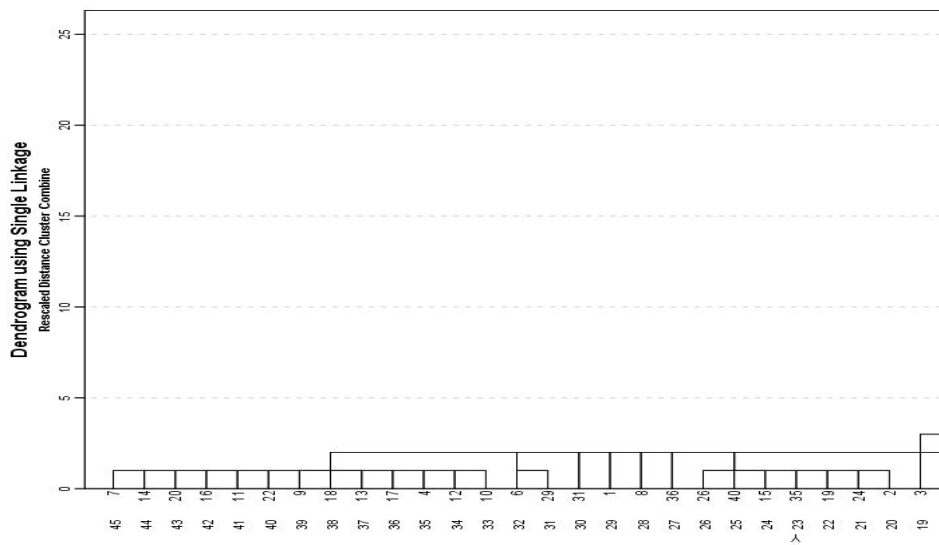


Figure 3.2 A Dendrogram showing the clustering of nitrates, sulfates and phosphates in all the sampled areas

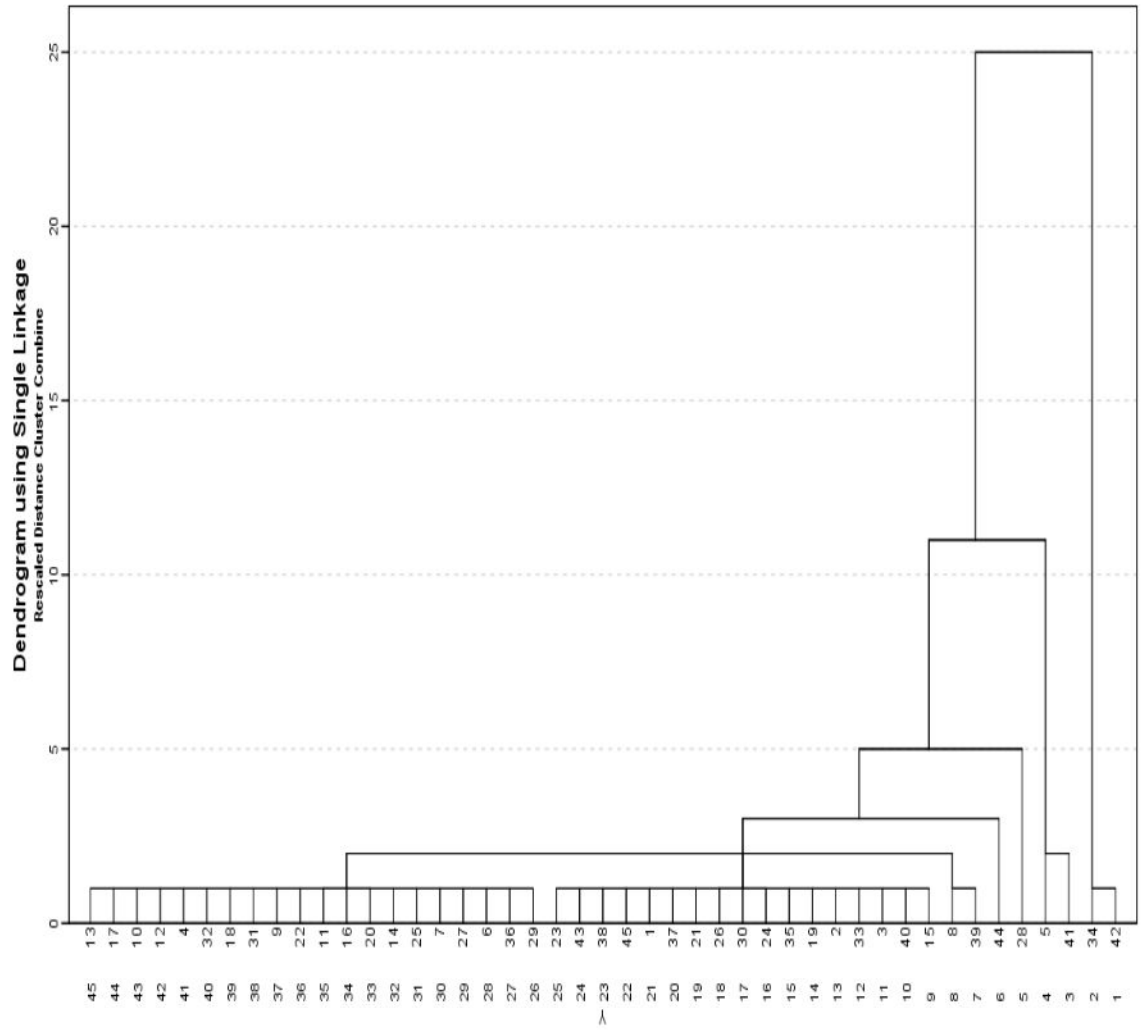


Figure 3.3 A Dendrogram showing the clustering of Zn, Fe and Cu in all the sampled areas

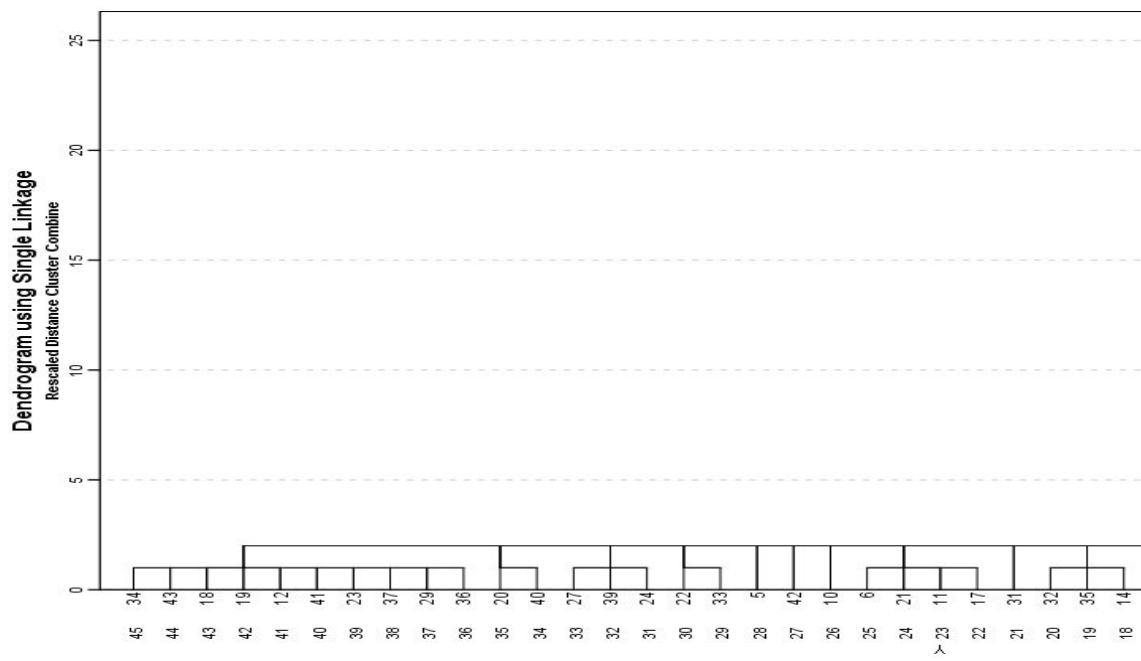


Figure 3.4 A Dendrogram showing the clustering of all parameters analyzed in all the sampled areas

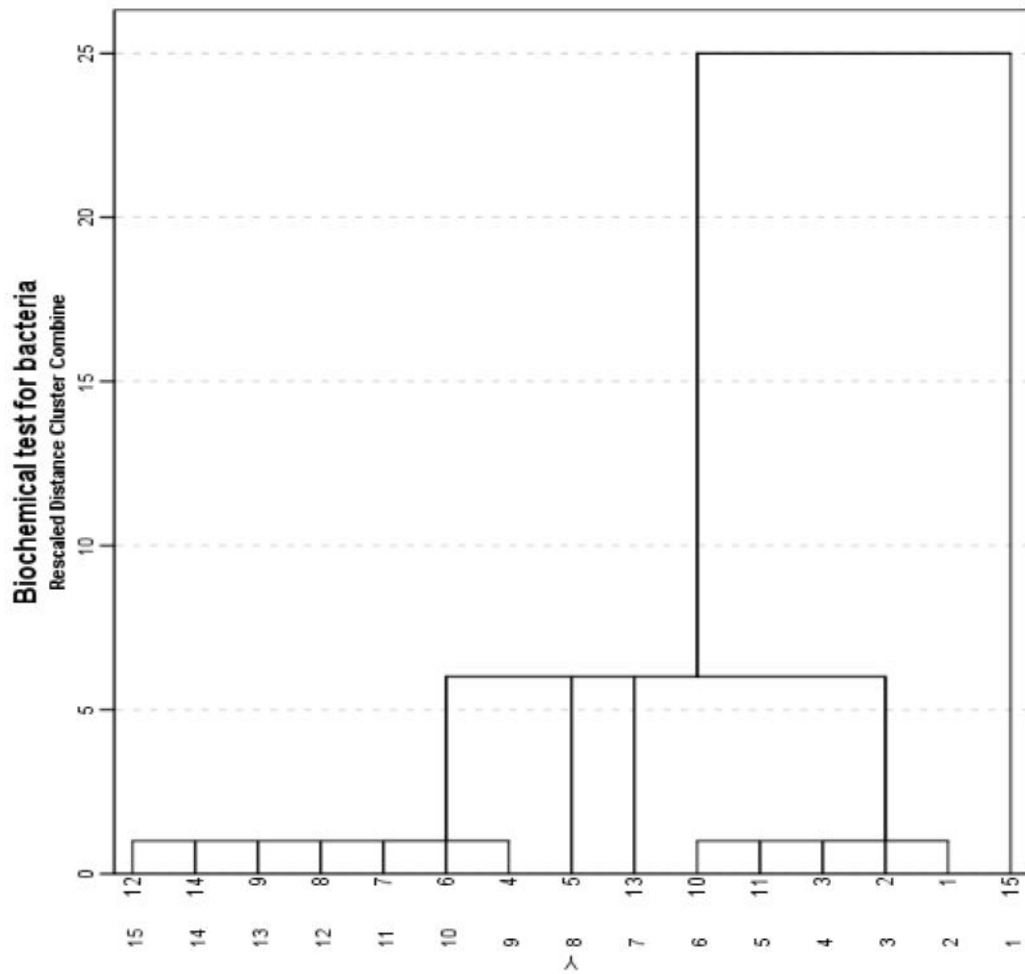


Figure 3.5 A Dendrogram showing the clustering of morphological and biochemical tests done on representative samples before planting

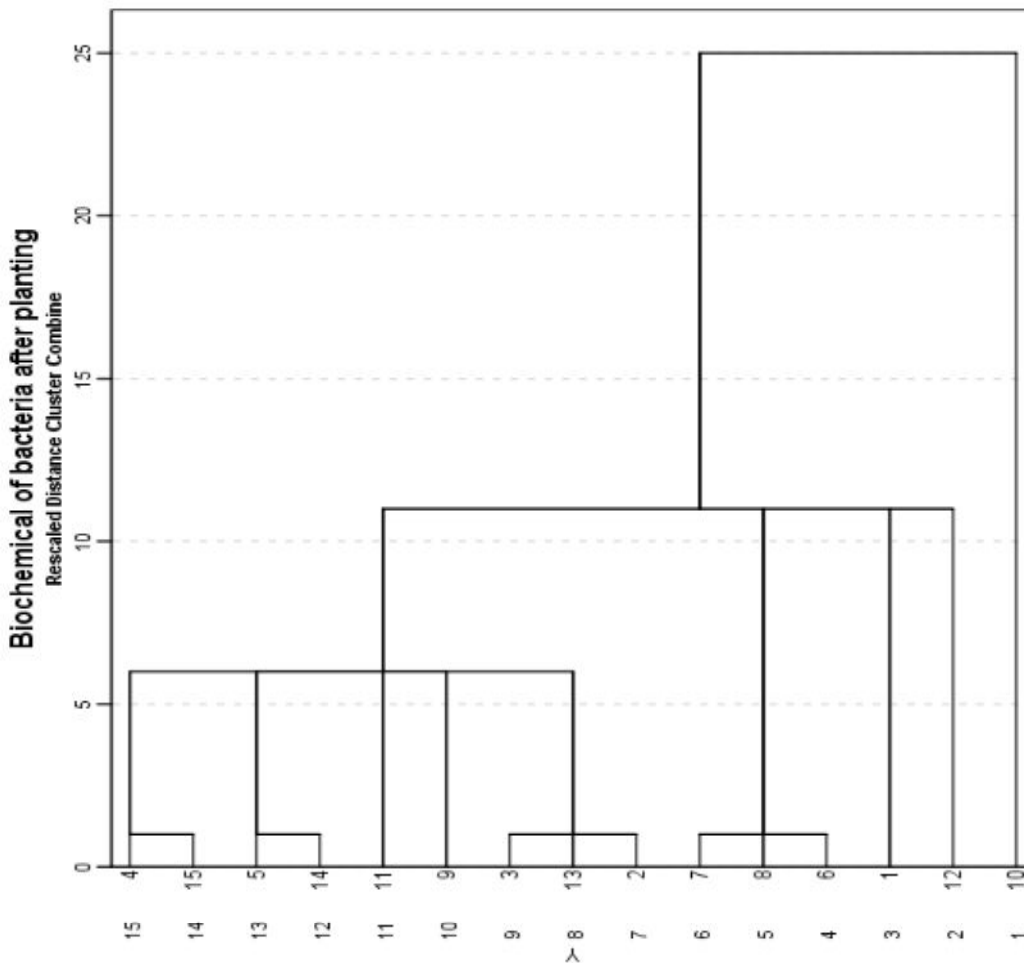


Figure 3.6 A Dendrogram showing the clustering of morphological and biochemical tests done on representative samples after planting

3.2. Conclusion and recommendation

3.2.1 Conclusion

The pH of all sampled areas was slightly acidic for both seasons with a mean pH of 6.413 ± 0.373 before planting and 6.336 ± 0.359 after planting. The pH decreased slightly after planting as compared to before planting. The pH observed in this study was suitable for wheat growing. The mean organic matter was found to have increased from 8.436 before planting to 10.092 after planting. These levels of organic matter were found to be sufficient for wheat growing. Potassium concentration in

most of the sampled farms were lower than the concentration of the control due to depletion of the mineral as tilling continues hence most of the farms were found to be potassium deficient. Calcium and magnesium concentrations in all the sampled farms were found to be also deficient hence addition of potassium, magnesium and calcium based fertilizers is required for the cultivation of wheat in this area.

Copper and zinc concentration in all the farms were found to be deficient hence addition of copper and zinc based fertilizers is highly recommended during the planting season. Iron was found to be sufficient in all the sampled farms hence no addition is required.

All the sampled areas were found to be nitrates sufficient but deficient in both sulfates and phosphates. It can clearly be seen that phosphates and sulfates were added during planting but this addition was not enough to make these nutrients sufficient for the crop hence more addition of phosphate and sulfate based fertilizers is still required in the farms. Comparing the samples with the control concentration, it was deduced that planting has depleted the amount of nitrates in the wheat growing farms and generally, it was observed that almost all farms had different application rates of these nutrients and maybe this depended on the ability of individual farmers financially. All the nutrients analyzed in this study therefore were found to be deficient except iron and nitrates hence addition of these nutrients is highly recommended.

The gram positive cocci bacteria dominated in the samples before planting but after planting the gram positive rod were favored. The change in cell wall characteristic and the shape of the bacteria between the two seasons may be due to the addition of fertilizers, pesticides and herbicides suppressing the growth of some bacteria and supporting others. The bacteria isolated in this study are *Pseudomonas spp*, *Micrococcus spp*, *Mycobacterium spp*, *Corynebacterium spp* and *Bacillus spp* and the abundance of the *Bacillus spp* in the samples of after planting was possibly due to their spore formation which increases their resistance against stress like pesticides, herbicides and fertilizers.

3.2.2 Recommendations

The results found in this study should be given to the Narok north sub-county department of Agriculture so that they can disseminate the information to the farmers.

This study recommends that an investigation should be done to determine the type and amount of fertilizers required per acreage to maximize the yields. A study should be done in a controlled environment and different concentration of nutrients applied to determine the exact levels of nutrients required by wheat for maximum growth in order to set standards in Kenya.

Further research could also be carried to determine the distribution of the nutrients in the different parts of wheat plant to know the nutritive value of the wheat grain from Narok north sub-county in terms of minerals. Nutrients necessary for wheat growth were determined therefore further research should be carried out on other essential elements like boron and molybdenum.

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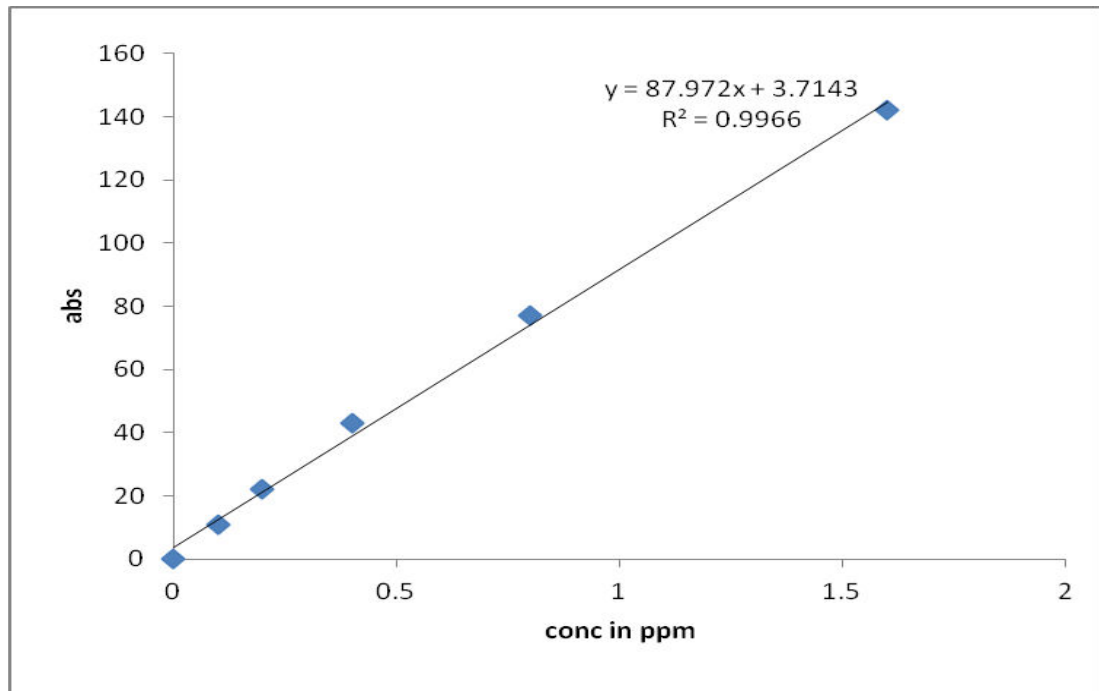
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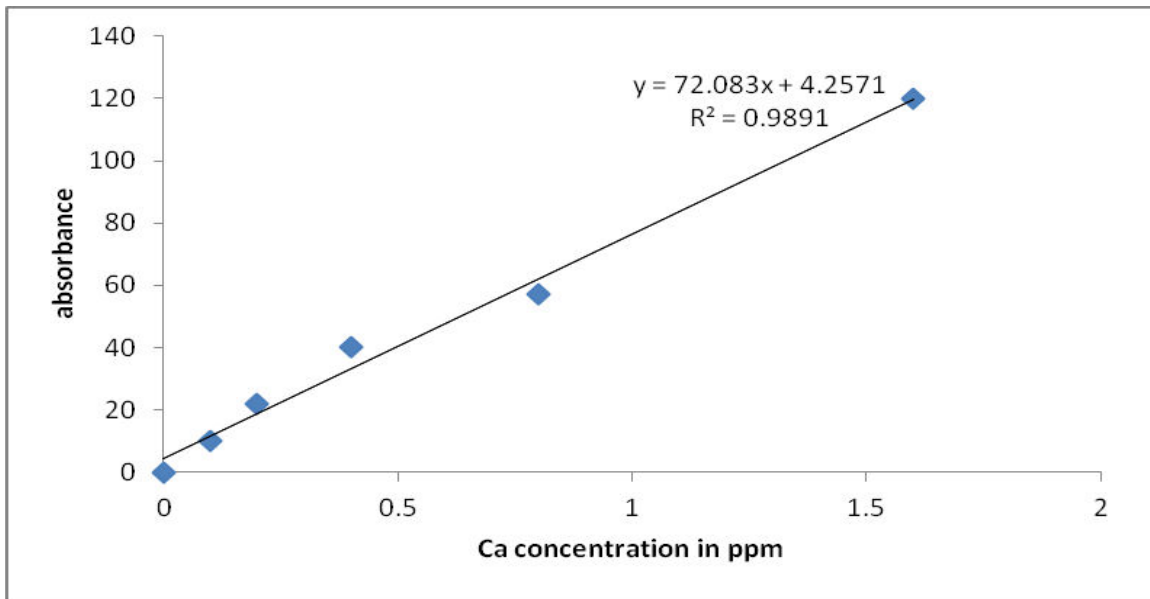
EAT

APPENDICES

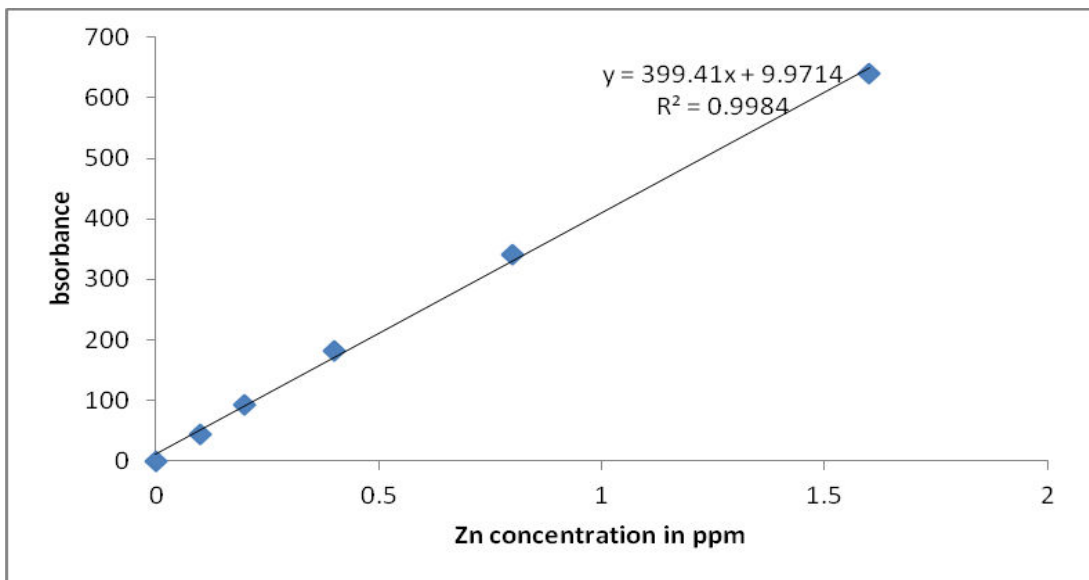
Appendix A1: A Calibration graph of Copper



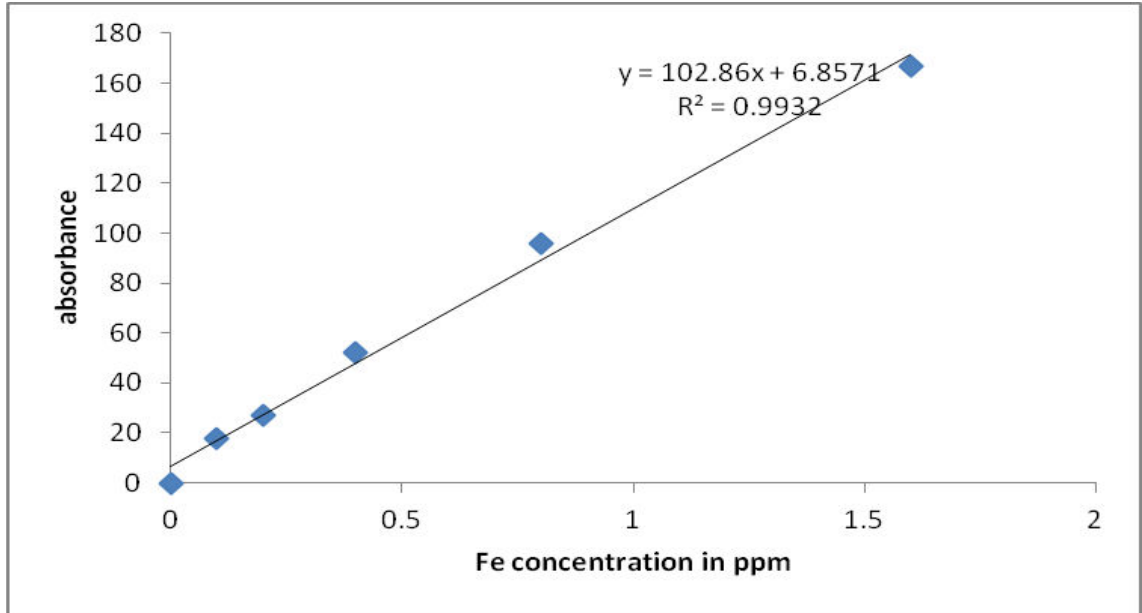
Appendix A2: A Calibration graph of Calcium



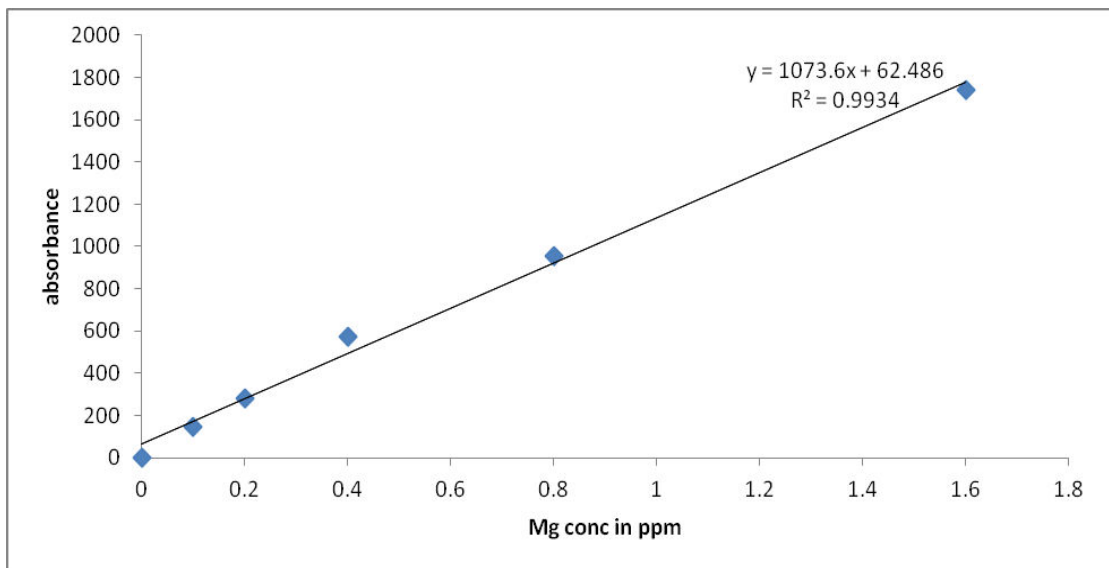
Appendix A3: A Calibration graph of Zinc



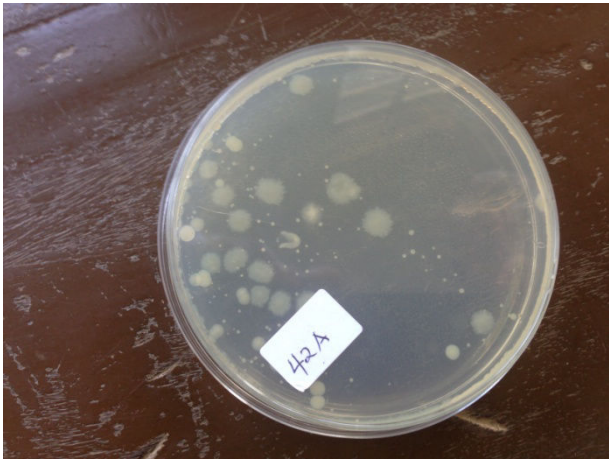
Appendix A4: A Calibration graph of iron



Appendix A5: A Calibration graph of Magnesium



Appendix B1: A petri dish showing bacteria colonies on nutrient agar



Appendix B2. Samples of selective media used for biochemical tests before dispensing into tubes



Appendix C. Publications from research work