NUTRIENTS LIMITING SOYBEAN (*Glycine max L*) PRODUCTION IN ACRISOLS AND FERRALSOLS OF KAKAMEGA AND BUSIA COUNTIES

BY

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DECLARATIONS

Declaration by student

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DEDICATION

To my parents and the entire family for their prayers, support and encouragement throughout my studies. To my supervisors for their dedication and support during my studies despite all the challenges. May God bless you abundantly.

ABSTRACT

Soybean is a good source of protein and calories and therefore important for food security. Soybean yields in western Kenya stand at 0.6 t ha^{-1} $season^{-1}$against a potential of 3 to 3.6 t ha^{-1} $season^{-1}$. This has been attributed to low soil fertility and a lot of work has been carried out on nitrogen (N) and phosphorus (P) nutrition but yields are still low leading to suspicion that other nutrients could also be limiting. To investigate this, two trials were set up. The first was a nutrient omission trial set up in the greenhouse to diagnose nutrients limiting soybean production in five distinct locations; Kakamega (Shikhulu and Khwisero sub –locations), Masaba central, Butere and Butula sub counties. Each of the soils collected from the different sites were subjected to ten nutrient solution treatments; 6 macronutrients (N, P, K, Ca, Mg and S) tested separately, micro-nutrients (B, Mo, Mn, Cu, Zn) in one treatment, a complete (macro and micro-nutrients), a control (distilled water) and a lime treatment. The experiment was laid out in Completely Randomized Design. Omission of nutrients from the nutrient solutions resulted in their low concentrations in plant tissues from all the soils. Potassium for instance had, 0.97, 0.78, 1.24, 1.14 and 1.78% in Masaba central, Khwisero, Shikhulu, Butere and Butula respectively. These concentrations were lower than the sufficient ranges in all the soils except in Butula. Significantly ($P \leq 0.05$) lower shoot dry weights (SDWs) were obtained from omission of K, Mg, P and micro-nutrients in soils from Masaba central and Butula, Mg and P in soils from Khwisero and K, Mg and P in soils from Shikhulu and Butere. Lime application significantly ($P \leq 0.05$) improved soil pH and SDWs in soils from Masaba central and Butere. In conclusion, K, Mg, P and micro-nutrients are limiting in most of the soils. In the second trial, field experiments were carried out in Masaba central and Butula to assess the effect of manure and Sympal (0:23:15 plus 10% CaO, 4% S, 1% and MgO) application as sources of macro-nutrients and micro-nutrients on soybean performance and to assess their effect and lime application on selected soil chemical properties and soybean yields. The treatments included; control, Sympal, manure, Sympal plus manure, Sympal plus micro-nutrients (B and Mo), manure plus micro-nutrients, Sympal plus lime, manure plus lime, Sympal plus micro-nutrients plus lime. The experiment was laid out in completely randomized block design. Sympal and farmyard manure were applied at 30 kg P/ha, lime at 2 t/ha, B at 0.44 kg/ha and Mo at 0.57 kg/ha. Manure significantly improved soil pH (8%) and total N (12.5%) in Masaba central and available P by 28% in Masaba central and 31% in Butula. Sympal, significantly improved soil pH by 8.1% in Butula and available P by 51% in Masaba central and 58.3% in Butula. Liming significantly ($P \leq 0.05$) improved soil pH by 11.7% in Masaba central and 16.4% in Butula and total N in combination with Sympal by 12.5% in Masaba central. Number of active nodules was significantly improved by micro-nutrients in Masaba central and Sympal plus manure in both sites. Manure significantly increased nodule dry weights, 2.0 and 4.03 g in Masaba central and Butula respectively. Micro-nutrients and lime in combination with Sympal and manure significantly improved soybean grain yields in both sites. From the results, fertilizers that supply macro (P, Ca, Mg, and S) and micro (B and Mo) nutrients and lime may be adopted to improve soil pH, available P, total soil N and soybean grain yields.

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CHAPTER ONE

INTRODUCTION

1.1 Background information

Grain legumes represent an important component of agricultural food crops consumed in developing countries and are considered vital for achieving food and nutritional security among the smallholder farmers (Sitou and Maredia, 2011). They are also an important source of income for the poor farmers and vital in improving soil fertility. This is because of their ability to fix the freely available atmospheric nitrogen into the form that can be utilized by plants, thereby reducing the need for mineral nitrogen fertilizers. It is estimated that one third to one half of the total nitrogen added to agricultural land come from legume-rhizobia symbiosis (Alistair *et al.*, 2009)

Soybean stands out as the most popular grain legume in the world. Its popularity is attributed to a number of factors related to its composition and productivity. Soybean is a source of the most consumed edible oil and livestock feeds (Myaka *et al*., 2005). Many other soybean products are directly used for human consumption including soymilk, soya sauce, protein extracts and concentrates. Apart from nutritional qualities, soybean’s high ability to fix nitrogen results to higher yields than other common grain legumes, has relatively few field and storage pests and diseases (Vanlauwe *et al.*, 2003). Cultivation of soybean is rapidly gaining popularity in Africa following high demand from the expanding livestock feed industry (which consumes about 70 - 80% of soybean produced per year) and need to restore soil nitrogen (Mahasi *et al*., 2011, Chianu *et al.*, 2009). Kenya is not an exception with substantial demand of approximately

150,000 mt/yr (Mauyo *et al*., 2010). Despite this huge demand, soybean production in Kenya is estimated at 8,000 mt /yr, with 80% of the volume produced in western Kenya. Productivity in western Kenya remains low with average yields of 600 kg/ha against the potential yield of 3,000 kg /ha (Mahasi *et al.*, 2011; Thuita *et al.*, 2012).

Adaptive research campaigns initiated across western Kenya by an N2Africa project team to assess the responses of soybean to the application of phosphorus (P) and Potassium (K) fertilizers and their combination with inoculants recorded yield increase on only 60% of the sites (Baijukya *et al.,* 2010). The soils not responding to P and K application were grouped as non-responsive. They include the sandy or the highly weathered -nutrient-depleted soils, majority in farms owned by poor farmers. The low soybean grain yields experienced in these soils could be attributed to declining soil fertility (Mburu *et al.*, 2011; Thuita *et al*., 2012) including chemical, physical and biological factors. The soil chemical degradation, specifically nutrient limitations can be caused by; continuous nutrient mining with minimal nutrient replenishment via crop harvests, soil erosion, leaching during heavy rainfall and soil acidity (Verde *et al.,* 2013). The minimal nutrient replenishment can be attributed to minimal fertilizer use in the region due to lack of money to purchase by the farmers. Soil acidity is a major contributor to soil chemical degradation through its associated nutrient deficiencies such as phosphorus, calcium, magnesium, molybdenum and potassium (Kisinyo *et al.*, 2013). This can be the case in western Kenya since most of the soils (about 0.9 million hectares of land) have got the pH values of less than 5.50 (Kanyanjua *et al*., 2002), therefore liming can help in alleviating the problem (Kiplagat *et al*., 2010). There is therefore an over arching need to close soybean yield gap in smallholder farms in western Kenya since the issue of non responsiveness has proven to be more than an unfavourable classification but rather a liability to farmers. It is imperative therefore to investigate and fix the causal factors to non-responsiveness such as nutrient limitations and soil acidity to improve soil productivity by use of organic and inorganic fertilizers.

1.2 Problem statement

Soils in western Kenya are nutrient depleted with the major cause being continuous cultivation with limited nutrient replenishment and widespread soil acidity. As a result, crop yields continue to decline and yield gap continues to widen leading to more food insecurity (Mahasi *et al.*, 2011). Grain legumes, a key component of the farming system of western Kenya are also affected by the declining soil fertility. Although soybean has the ability to manufacture its own nitrogen, in most soils the availability of other nutrients and favourable pH ranges to enhance the biological nitrogen fixation (BNF) process and other plant functions is limiting (Weisany *et al.*, 2013). A lot of effort has been made to improve soybean production including application of mineral fertilizers and rhizobial inoculants but have yielded little success (Giller, 2001). Application of rhizobial inoculants have led to yield increases over the control treatments but to a very small extent, especially when compared to the potential yields (Thuita *et al.*, 2012). In these studies low yields which mainly coincides with poor nodulation and a lack of response to P, K and inoculants indicates that other nutrients may be limiting (Baijukya *et al.*, 2010). Much of the early research on crop nutrition in west Kenya focused on the plant's needs for the three major macro nutrients (NPK). It is now recognized that there is need to emphasize on the deficiency of other macro and micro nutrients and their interactions in order to ensure balanced nutrition and efficient use by the plants (Robert, 2004).

1.3 Justification

Soybean is increasingly becoming an integral part of the farming systems of Kakamega and Busia counties as it offers nutritional and market opportunity to farmers. However, soybean yields remain low because of soil infertility (Majengo *et al*., 2011). For soybean, nodulation and nitrogen fixation are dependent upon adequate supply of essential micro (B, Zn, Mo, Cu, Co and Mn) and macro (P, K, Mg, Ca and S) nutrients under favourable pH ranges (Musandu and Ogendo, 2001). These nutrients are essential for symbiotic interaction and also for the host plant microbial partners. They help in the establishment and functioning of the symbiosis by carrying out their specific physiological and biochemical roles. Although soil infertility in western Kenya is caused by natural causes (e.g. inherently low fertile parent materials in which the soils are formed, extensive weathering and leaching of nutrients due to high rainfall) and soil acidity, the situation is aggravated by nutrient depletion through continuous cultivation with inadequate replenishment. A quick solution to this would be to apply fertilizers but this would need an understanding of which specific nutrients other than N, P and K that are limiting including the extent of limitation (Weisany *et al*., 2013). This information is useful in developing fertilizer formulations/blends for specific soils and crops. There is also need to test the effects of the different fertilizers (organic, inorganic and their combinations) and lime application so as to know how to improve the fertility status of these soils.

1.4 Objectives

1.4.1 General objective

The general objective of the study was to identify nutrients limiting production of grain legumes, specifically soybean and assess the crop response following their application in Acrisols and Ferralsols of western Kenya.

1.4.2 Specific objectives

i. To identify the macro and micro nutrients that are limiting soybean production in Acrisols and Ferralsols of Kakamega and Busia Counties

ii. To establish the effects of farmyard manure and sympal as sources of macro-nutrients (P, K, Mg, Ca and S) and micro-nutrient (B and Mo) application on soil pH, available P, total N and soybean performance in Acrisols and Ferralsols of Kakamega and Busia Counties

iii. To establish the effect of liming on soil pH, available P, total N and yield of soybean in Acrisols and Ferralsols of Kakamega and Busia Counties

iv. Assess the economic benefits of applying sympal, farmyard manure, lime and micro-nutrients for soybean production in degraded soils of Kakamega and Busia Counties

1.5 Research hypotheses

i. Nutrients other than N, P and K limit soybean production in Acrisols and Ferralsols of Kakamega and Busia Counties

ii. Application of farm yard manure and sympal as sources of macro (P, K, Mg, Ca and S) and micro-nutrients (B and Mo) will enhance soil pH, P and N availability and improve soybean performance in Acrisols and Ferralsols of Kakamega and Busia Counties

iii. Application of lime will enhance P and N availability, soil pH and yield of soybean in Acrisols and Ferralsols of Kakamega and Busia Counties

iv. The economic management practice to the farmers will be the one that gives optimum economic returns with minimum cost of inputs.

CHAPTER TWO

LITERATURE REVIEW

2.1 Importance of soybean

Soybean is an ideal crop for improving nutrition, food security, sustainable crop and livestock production systems (Mahasi *et al*., 2011). Compared to other legumes soybean has a high commercial value and contains most amino acids required by the human body (Myaka *et al*., 2005). Soybean has the highest concentration of protein (40%) compared to other legumes which contain about 20% protein, thus enhancing human nutrition (Mauyo *et al*., 2010).

When rotated with cereals it can contribute to yield increases of cereals by up to 25% (Sanginga, 2003; Mahasi *et al*., 2011). This is because the bacterium (*Rhizobium japonicum*) harboured in the root nodules enables the crop to fix nitrogen contributing to improved soil fertility (Mathu *et al*., 2012). It is estimated that soybean fixes up to 200 kg N ha^{-1} year^{-1} under optimal field conditions (Cheminingwa *et al*., 2007). Apart from improving soil fertility through biological nitrogen fixation (BNF), soybean has the ability to reverse soil fertility decline through its relatively low N harvest index. This therefore offers a quick way of improving soil fertility especially in densely populated areas such as western Kenya (Vanlauwe *et al.*, 2003).

Soybean has the potential of reducing *Striga hermonthica* parasitism when intercropped with cereal crops such as maize and sorghum (Carsky *et al.*, 2000). This is very important especially in western Kenya where striga is causing a lot of yield losses. Soybean also presents the farmers with the much needed alternative cash income source, thus reducing poverty. At national level, soybeans help in contributing to improvement of the agricultural sector which is one of the main pillars of Kenya's economy towards achieving the vision 2030 goals (Chianu *et al*., 2008).

2.2 Factors affecting soybean production

Soybean is indeed among other grain legumes whose performance and final yield is affected by a wide range of factors. Successful soybean yield in the field depends on the interaction of;

$(G_L \times G_R) \times E \times M$

Where: G_L = Soybean genotype; G_R = rhizobial strain; E = environment (Climate (temperature x rainfall x day length), Soils (nutrient limitations, acidity and toxicities) M = management (agronomy – inoculation, seeding rates, plant density, weeding) - (Diseases and pests are also a function of G x E x M) (Giller *et al.,* 2011). These factors are explained below;

2.2.1 Genotypic variation

Wide genetic variability for nodulation and N_2-fixation exists in many legumes (Giller, 2001). Genetic variation in soybeans as well as compatibility of rhizobium-plant cultivars can greatly affect the efficiency of symbiosis established. The ability to fix substantial amount of nitrogen and subsequent grain yield is often associated with large total biomass production due to indeterminate ability and with long duration. This variability often is associated with plant ability to adapt to environmental stress such as soil moisture stress, diseases and low soil fertility (Yadegeri *et al*., 2008). Investing in breeding varieties for adaptation to adverse environmental stress (notably soil acidity and P limitation) conditions and also to the high yield potential varieties will give rapid success in improving BNF and soybean yields.

2.2.2 Rhizobial strains

The relationship between the legume and the rhizobium strain should be considered for effective nodulation to occur. This relationship can be very specific needing farmers to inoculate the legume seeds with the appropriate bacterial strain (e.g. some soybean and chick pea) or non specific (promiscuous) allowing farmers to grow legumes that nodulate with a wide range of both fast and slow growing rhizobia (Wietske, 2012). The population density of indigenous rhizobia is also a major factor determining competition for nodule occupancy and response to inoculation. Use of massive inoculation rates can overcome competition from indigenous non effective strains. But such a delivery system is not yet practical or economical. Experiments from multi- site standardized field inoculation trials with several legumes have revealed that 59% of the variation in inoculation responses could be accounted to the number of the indigenous rhizobia (Harold and Fudi, 1992).

2.2.3 Environment

2.2.3.1 Environmental stresses

Rhizobia symbiosis is quite sensitive to environmental stresses such as high temperatures and soil dryness leading to low nitrogen fixation efficiency (Hungria and Vargas, 2000). Soil physicochemical constraints and climatic conditions all significantly influence the N availability to achieve increased crop yield (Adriano, 2000).

2.2.3.2 Soils

Soil acidity

Soil acidity is the prevalence of the hydrogen and aluminium cations in the soil solution and is mainly reflected in the soil pH levels generally below 5. This is widespread in highly weathered and leached soils of the tropics mainly the Acrisols, Ferralsols and Nitisols (Okalebo *et al*., 2009). Soil acidification is mainly caused by the leaching of the basic cations (calcium, magnesium, potassium and sodium) over time, continuous cropping, use of ammonia containing fertilizers, atmospheric pollution leading to acid rain, organic matter decomposition and inherent acidity from parent material with little limestone (Kiplagat *et al.*, 2010). Soil acidity can affect plant growth directly or indirectly by affecting the availability of several plant nutrients, increasing levels of some elements' toxic levels and influencing microbial activity or other soil properties (Antonio, 2011). Plant-growth limiting factors in acid soils include N, P, Ca, Mg, Mo and Zn deficiencies and or Al, Mn, Fe and H ion toxicities (Andric *et al*., 2012). Soil pH outside the optimum range for soybean is detrimental to biological nitrogen fixation as it disrupts the communication that needs to occur for root hair infection thereby limiting nodule formation. The level of soil pH also affects the amount of nitrogen fixed, for instance, a pH level of 4.4 leads to reduction in nitrogen fixed by up to 30% (Abendroth and Elmore, 2006). Acidic soils are also low in calcium and therefore it limits soybean root growth thus affecting its efficiency in exploration of other important plant nutrients (Vonvilay *et al.,* 2009). The survival of soil micro-organisms in the soil is also affected by the soil acidity due to chemical imbalances in the soil. However, rhizobia strains vary in their level of tolerance to aluminium toxicity that occurs on acidic soils. Among the *Bradyrhizobium spp, Bradyrhizobium japonicum* strains are more susceptible to Al toxicity than other strains (Majengo *et al.*, 2011).

Liming helps to counteract soil acidity by raising the pH thus making aluminium, iron and manganese less soluble and prevent them from reaching levels that are toxic to plants (Kiplagat *et al*, 2010). Lime contains Ca and/Mg which are very important in displacing the H^+, Fe^{3+}, and Al^{3+} ions from the soil colloids (Kisinyo *et al.*, 2013). Lime can be applied to the soybean and its effect can still be felt in the subsequent seasons by providing adequate phosphorus to the crop (Vonvilay *et al.*, 2009).

Nutrient limitations

In Sub-Saharan Africa (SSA), continuous cropping without adequate fertilization has led to soil fertility depletion and subsequent low crop yields (Muyayabantu *et al.*, 2012). Soil nutrient mining and soil fertility decline can be evidenced by generally observed negative balances for N, P and K at the farm levels (Vanlauwe *et al.,* 2003). Studies in western Kenya strongly suggest that nitrogen and phosphorus are the two most widespread nutrient limitations to crop growth. Many farmers in western Kenya are aware of the need to apply N and P fertilizers to increase crop yields, but the costs of the fertilizers are prohibitive (Okalebo *et al.*, 2010). Deficiency of other nutrients is another factor which might limit plant growth indirectly by inhibition of the process of N_2 fixation. Mineral nutrients may influence N_2-fixation in legumes and non-legumes at various levels of the symbiotic interactions: infection and nodule development; nodule function, and host plant growth. For instance, legumes dependent on N_2- fixation may also suffer from N deficiency when they do not receive an adequate supply of P, because this element plays an important role of energy transfer in the process of N_2-fixation (Marschner, 1995).

2.2.4 Management

Legumes and particularly soybean yields are generally low (average of 600 kg/ ha nationally in Kenya). This can be attributed to soybeans susceptibility to biotic and abiotic stresses (Mahasi *et al.,* 2011). These stresses may come from all the sides and includes; pests, diseases, insects, weeds and nutrient deficiencies. Thus proper management is needed to help in increasing the yields (Dugje *et al*., 2009). Appropriate crop stand should be established to ensure good performance of soybean. Low crop stands leads to low yields while very high stands can encourage occurrence of pests and diseases, thus leading to the requirement of fungicides and pesticides to control them (Pedersen and Lauer, 2004). Ramakrishna *et al.,* (2000) reported that soybeans are poor competitors of weeds during the early stages of growth, and may consequently suffer heavy yield losses. This is because weed competition has an adverse effect on crop yield and BNF.

Soybean in western Kenya is mainly grown by smallholder farmers (0.1 to 0.2 ha) and therefore they are not able to supply adequate amounts of fertilizers needed (Mburu *et al.*, 2011; Mahasi *et al.*, 2011). Inoculation offers a viable alternative since it is simple and installation costs are very low compared to the chemical fertilizers (Mburu *et al.,* 2011). Farmers should be encouraged to adopt inoculation using commercial rhizobial inoculants in combination with fertilizers that supply essential nutrients such as P, K, S and micronutrients to increase yields (Mugendi *et al*., 2010).

Farmyard manure and other farm derived resources such as crop residues, compost and household waste has commonly been used by the farmers in management of soil fertility. They are very important in the soils since they help in improving their water holding capacity and cation exchange capacity (Otieno *et al.*, 2007). Organic manures provide a good substrate for the growth of micro-organisms and maintain a favourable nutritional balance and soil physical properties (Maheshbabu *et al.*, 2008). It has been found that the use of organic manures in integration with inorganic fertilizers meets the needs of micro-nutrients in soybean (Konthoujam *et al.*, 2013).

2.3 Biological nitrogen fixation by soybean

2.3.1 Mechanism

Nitrogen fixation by soybean occurs when in symbiotic associations with specific bacteria known as *Bradyrhizobium japonicum*. The bacterium has a nitrogenase enzyme which is capable of converting atmospheric nitrogen into amino acids and protein through ammonia (Ali and Kemal, 2011). This process begins by the attachment of the rhizobia to the flavanoids produced by the root cells. The bacteria then attach themselves to the extensions of the epidermal cells known as the root hairs. This is followed by a firmer attachment of the bacteria to the root surface. The host legume then senses the chemicals produced by the bacteria known as the nod factors. This causes the colonized root hairs to curl and form a tubular structure known as the infection thread. When the bacteria reach the root itself, they stimulate cortical cell divisions that lead to the formation of a nodule in which chemical process, that is BNF, takes place (Wagner, 2012). Although the process involves a large number of chemical reactions, it can be summarized by the following equation;

$N_2 + 8\ H_2 + 16\ ATP \rightarrow 2\ NH_3 + 2H_2 + 16\ ADP + 16\ Pi$

The equation indicates that one molecule of nitrogen gas combines with eight molecules of hydrogen also known as protons to form two molecules of ammonia and two molecules of hydrogen gas. The 16 molecules of ATP (= Adenosine Triphosphate, an energy storing molecule) represent the energy required for the biological nitrogen fixation to take place. A detailed account of BNF process is explained in, forages.oregonstate.edu/nfgc/eo/…/nitrogenfixation/definition.

2.3.2 Use of rhizobial inoculants to enhance BNF by soybean

The rhizobium-legume symbiosis plays an important role in agriculture because it offers the ability to convert atmospheric molecular nitrogen into the forms usable by the plant (Rebah *et al.,* 2007). This has led to the adoption of commercial rhizobial inoculants to help in enhancing the process of biological nitrogen fixation and also in land remediation (Mohammadi *et al*., 2012). It has also led to the adoption of the promiscuous soybean varieties bred by the Institute of Tropical Agriculture (IITA) which nodulate freely with the indigeneous *Bradyrhizobium* spp. population (Vanlauwe *et al.*, 2003).

Soybean has been known for its good nitrogen fixing ability and is capable of fixing up to 80% of its nitrogen requirements when it is well nodulated. Studies indicated that legumes have specific requirements for rhizobia (e.g. soybean, chickpea) and thus need inoculation (Thuita *et al*., 2012). This can be achieved using commercially available rhizobia inoculants which have already been tested (e.g. the Biofix inoculants produced in Kenya by MEA ltd). Mburu *et al.*, (2011) and Majengo *et al.,* (2011) reported that use of Biofix rhizobial inoculants (containing rhizobia strain USDA 110) results in increased nodule fresh weights compared to the control treatment. They also reported that application of inoculants in conjunction with P greatly increased soybean yields. Vanlauwe *et al.*, (2003), reported that use of promiscuous soybean varieties that nodulate freely with indigenous bacterial strains increased grain yields over the control treatments in western Kenya. The yield increases of soybean obtained after the use of commercial rhizobial inoculants and promiscuous soybean varieties have not reached the potential yields (Mburu *et al.,* 2011, Majengo *et al.,* 2011, Vanlauwe *et al*., 2003 and Mahasi *et al*., 2011). Some of the common inoculants that have been used include; legume fix made by legume technologies limited in the UK, Histick made by Underwood USA, Rhizoliq – top2 made by Rhizobacter in Argentina and Vault made by Becker Underwood in the USA. These inoculants are not easily available to the farmers as compared to Biofix which is manufactured in Kenya.

2.4 Importance of nutrients in soybean production

2.4.1 Macro-nutrients

Nitrogen is the critical limiting element for growth of most plants due to its unavailability. Adequate supply of nitrogen is beneficial for carbohydrates and protein metabolism, promoting cell division and cell enlargement (Wamba *et al*., 2012). Nitrogen is needed during the first weeks of legume establishment (lag phase) as it helps in the formation of source leaf that provides photosynthates required for nodule growth and activity. Thereafter, increase in levels of combined nitrogen leads to the decline in nitrogenase activity, as an expression of nitrogen fixation, declines drastically. Because of this adverse effect, N fertilization usually is not recommended for soybean (Marschner, 1995; Adriana, 2000).

A good supply of P is usually associated with increased root density and proliferation which aid in extensive exploration and supply of nutrients and water to the growing plant parts, resulting in increased growth and yield traits (Wamba *et al*., 2012). A high phosphorus supply is needed for nodulation. This requirement might be higher than for root or shoot growth. It has been found out that nodule dry weight relative to the shoot and, especially to the root dry weight for soybean increased with increase in phosphorus concentration levels (Marschner, 1995). Inadequate supply of phosphorus to soybean which depends on nitrogen fixation may lead to nitrogen deficiency since phosphorus plays an important role in nitrogen fixation (Wietske, 2012). The element helps in the plants energy transfer system and thus its deficiency retards growth and tillering (Zarrin *et al*., 2006). The element is also important in nodule development and signal transduction and, to P lipids in a large number of bacteriods (Weisany *et al*., 2013).

Potassium is the most abundant inorganic cation in the plants and comprises up to 10% of plant dry weight and is vital for various functions in the plant as photosynthesis, osmoregulation and transpiration (Wamba *et al*., 2012). The element is also important in activation of numerous enzymes and is a major cationic inorganic cellular osmoticum. The growth rate of internodes is affected by K deficiency and some cotyledonous species may form rosettes. A qualitative requirement for K has been demonstrated for some rhizobia. It has been suggested that *R. Trifolii* and *R. meliloti* show restricted growth when K is omitted from a medium (Weisany *et al*., 2013). Calcium on the other hand aids in nodule formation. Nodule formation has a much greater requirement for calcium than root and shoots growth of the host plant. Experiments have shown that after nodule initiation further nodule growth was not affected by a decrease in calcium concentration, indicating that only the first step of infection is highly sensitive to calcium supply (Marschner, 1995). This element also affects the attachment of rhizobia to root hairs, nodulation and nodule development (Weisany *et al.,* 2013, Mohammadi *et al*., 2012).

Magnesium functions in plants are mainly related to its capacity to interact with strongly nucleophilic ligands (e.g. phosphoryl groups) through ionic bonding, and to act as a bridging element and/or form complexes of different stabilities. One of the complexes is that of chlorophyll which has covalent bonds. It is also involved in activation of enzymes, an example of the enzymes is RuBP carboxylase (Marschner, 1995). Sulphur on the other hand is a component of amino acids (cystein and methionine) needed for protein synthesis. Sulphur is also a vital part of the Ferredoxin, an iron-sulphur protein occurring in the chloroplasts. Ferredoxin has a significant role in nitrogen dioxide and sulphate reduction, the assimilation of N by root nodule and free living N-fixing soil bacteria (Hussain *et al.,* 2011). The existence of sulphate ions in the root zone has been found to increase the growth and development of plants under hydroponic systems (Masoud *et al.,* 2012).

2.4.2 Micro-nutrients

Molybdenum is a metal component of nitrogenase and thus all the nitrogen fixing systems have a high specific molybdenum requirement. Molybdenum-induced deficiency in legumes relying on nitrogen fixation is widespread in acid mineral soils of the humid and sub-humid tropics. Several key enzymes of the nitrogenase complex as well as for the electron-carrier ferrodoxin require molybdenum (Marschner, 1995). There are reports that foliar application of Mo on the grain legumes in field conditions increases levels of nitrogen fixation and nodule mass resulting in higher overall N content and seed yield (Weisany *et al* ., 2013).

Boron is required for the normal development of most plants. Lack of boron in the soils lead to alteration in biological nitrogen fixation in soybeans. It has also been shown that in boron-deficient plants, the number of rhizobia infecting the host cells and the numbers of infection threads were reduced and developed morphological aberrations (Weisany *et al.*, 2013).

Zinc, manganese and copper are mainly needed for the host plant growth and development. Manganese participates in a number of cellular activities including the stabilization of the structural proteins, ultrastructure of chloroplasts and photosynthesis (Epstein and Bloom, 2005). Research has shown that manganese can be critical in maximizing N_2 fixation activity of soybean, especially under soil water deficit conditions. It also helps in the reduction of ureides in the leaves of soybean as they help in their catabolism (Mayoral and Sinclair, 2005). Copper plays an important role in respiratory proteins that are required for nitrogen fixation in rhizobia. It also plays a role in protein that is expressed with nifgenes that may affect the efficacy of the bacteriod function (Weisany *et al.*, 2013). Its deficiency has been found to result in development of numerous small nodules typical of those associated with a completely ineffective strain. Zinc application is most important on plants exposed to salinity stress. This is because it causes a noticeable enhancement of photosynthesis (*Pn*), water use efficiency, mesophyll efficiency and quantum yield compared with plants exposed to salinity stress alone (Weisany *et al.*, 2013). Chauhan and Brian, (1995) found out that there is a relation between zinc and *B. japonicum.* They found out that through protein engineering, enzyme S-aminolaevulinic acid which normally has Mg^{2+} as a co-factor can bind Zn^{2+}. This relation was however not found to affect the symbiotic nitrogen fixation.

2.5 Identification of nutrient deficiencies

Plant nutrient deficiencies can be identified from the observation of the visual deficiency symptoms. This method can be used at some instances to determine the amount and type of fertilizer to be used, for example foliar sprays containing (Fe, Zn or Mn) or Mg. In some instances, visual diagnosis is inadequate for making fertilizer recommendation (Marschner, 1995). Soil and plant tissue analysis are important and helps in supplementing results from visual deficiency symptoms. Soil tests help tell which nutrients are available to plants and identify other problems that may cause nutritional problems e.g. acidity and salinity. Plant tissue analysis is important because it shows how much amount of nutrients are available in plants and whether they are too low or high (Shobber and Denny, 2010).

2.6 Double pot technique

The double pot technique is a method used to assess the nutritional stress of plants grown in different soils. This technique was developed by Janssen (1974). The absence of an element can be seen from the deficiency symptoms developed such as limited growth and leaf chlorosis. The symptoms can be visible in the early growth stages and thus can be used to draw conclusions for fertilizer recommendation. The main principle of this technique is the supply of major and trace elements to plant roots in a nutrient solution except for the manipulation of the element that is object of study (Cardoso *et al.,* 2004). The roots of the plants in the soil compartment (upper pot) pass through a mesh that forms the bottom of the pot and grow into the nutrient solution in a container (lower pot) placed under the upper pot. The plant is therefore provided with two sources of plant nutrients (test soil and nutrient solution) for its simultaneous uptake. With this approach, the nutrients other than the nutrient to be tested are available to plants in non-limiting amounts from the nutrient solution in the lower pot. The nutrient that is omitted from the solution can only be taken up by plants from the soil (Janssen, 1990). In this way, the nutrients are supplied to the plants without mixing them with the soil and without disturbing the native conditions of the soil.

This method has been used in a number of tropical countries as it serves as a tool for diagnosis of symptoms observed in the field for specific crops. A few examples of the experiments are shown below;

Table 1: Experiments carried out using double pot technique

Experimental soils	Plant species	Nutrients tested
Cameroon	Millet, rye grass, sorghum	P and K
Colombia	Maize	N, P, S, K, Ca and Mg
Ecuador	Cocoa	N, P, S, K, Ca, Mg, Fe, Mn, Zn, Cu, Mo and B
Netherlands	Rape and sunflower	P
Netherlands	Semi-natural grass	N, P, S, K, Ca and Mg
Suriname	Maize	N, P, K, Ca and Mg

Source: (Janssen, 1990).

2.7 Economic analysis

A new technology can be evaluated in terms of its impact on the productivity, profitability and sustainability of the farming systems (Boughton *et al.,* 1990). The CIMMYT, (1988) approach to the economic analysis of agronomic data utilizes partial budgeting combined with marginal analysis. Partial budgeting analysis is simple but effective technique for assessing the profitability of the new technology with an existing enterprise. It provides a foundation for comparing relative profitability of alternative treatments and testing how robust profits are in the event of changing product or input prices. The most important values in partial budget analysis are gross field benefits (GFBs), total costs that vary (TCV) and net field benefits (NFBs). (Boughton *et al.,* 1990; CIMMYT, 1988). The costs and benefits of a given enterprise are established from the prevailing market prices for the inputs and outputs in a particular area.

CHAPTER THREE

MATERIALS AND METHODS

3.1 Introduction

The study was conducted in two phases. Phase one of the experiment focused on the identification of limiting nutrients in selected soils using the double pot technique (modified nutrient omission technique). This experiment was established in a greenhouse located at the University of Eldoret. Phase two (field experiments) focused on assessment of the response of soybean to the application of "Sympal" (0:23:15 plus 10% CaO, 4% S and 1% MgO) fertilizer blend for legumes, farmyard manure, micronutrients (B and Mo) and establishing the effect of liming on nutrient availability and yield of soybean in Acrisols and Ferralsols of Kakamega and Busia Counties.

3.2 Greenhouse experiment

3.2.1 Set up of the double pot experiment

Assessment of limiting nutrients was carried out in the greenhouse at the University of Eldoret located at 0^{o} 34' N and 35^{o} 18' E. The temperatures in the greenhouse ranged from a minimum of 18.3 to a maximum of 35.8^{0}C. The experiment adopted the so called double pot technique (Janssen, 1974). In this technique, two pots were used whereby the upper pot (pot 1, plate1) which had a gauze fitted at the bottom was filled with 250 g of soil. The lower pot (pot 2, plate 1) was filled with nutrient solution and had a lid to support the upper pot. A space of approximately 1 cm was left between the bottom of the upper pot and the nutrient solution to allow oxygen supply to the plant roots. Three seeds were planted in the soil. In this experiment, the upper pots were provided by parts of a common sewage pipe with 9 cm diameter. A mesh was cut into small pieces and tied to their bottom, to prevent the soil from falling into the solution, but providing passage for the roots. As bottom pots, small plastic pots of 2 litre volume were used.

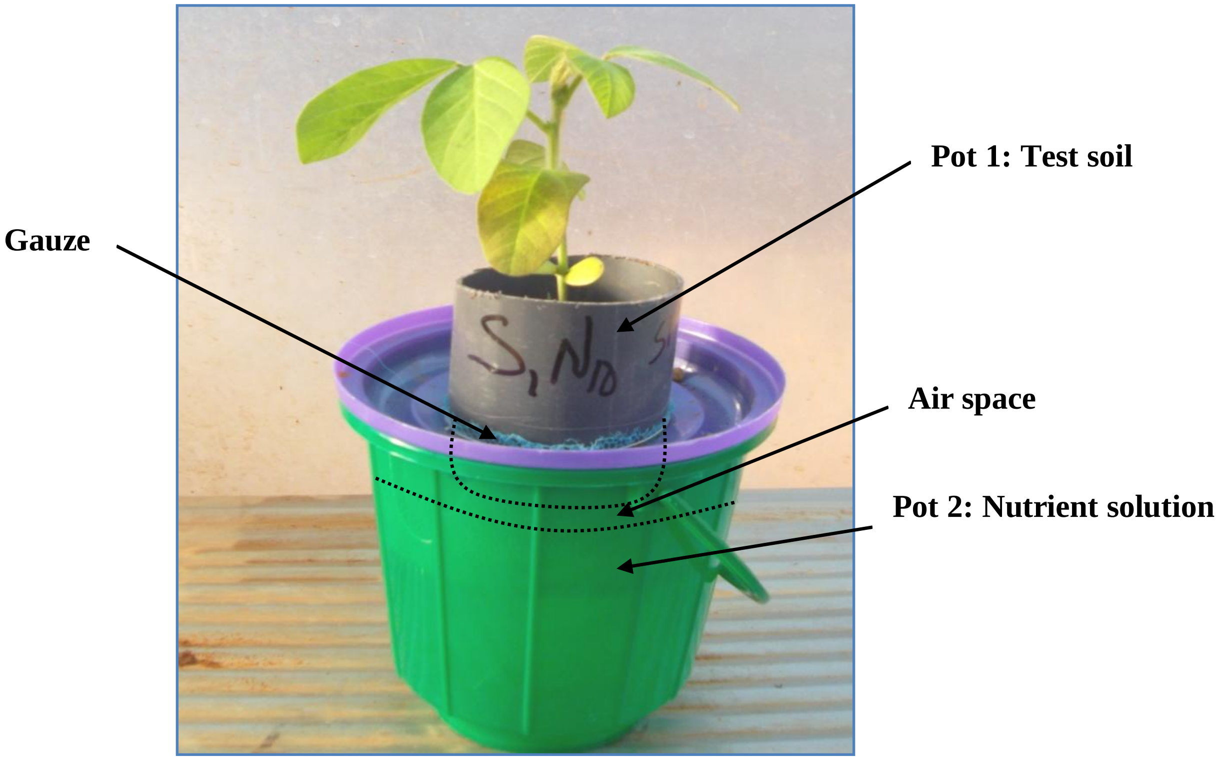

Plate 1: Set up of the double pot experiment

3.2.2 Soils used in the greenhouse experiment

The soils that were used in the greenhouse experiment were collected from five (5) distinct locations representing major soybean growing areas in western Kenya. These areas have been identified as having soils with poor response to P-fertilization and inoculation when growing soybean plants (Table 2). From each site approximately 60 kg of soil were collected by taking the top soil (0-20 cm) using a hand hoe. The zig-zag method was used in the sampling procedure. This was done by dividing the land into two equal portions and collecting samples transversally to ensure that the land was well represented. The soil portions from one site were mixed to come up with a composite representative sample. The soils were then air-dried, sieved to pass through a 5 mm sieve then put on the top pot (Plate 1), each carrying 250 g of soil. The field capacity of each of the soils was established by saturating the soils with water and covering them using perforated polythene papers. They were then weighed and then left for 48 hours in the greenhouse conditions. They were then reweighed and the difference between the weight of the container plus saturated soil and that of the container plus soil after 48 hours represented the field capacity. From the remaining soils of each location, a sub-sample of about 250 g was taken for chemical and physical characterization in the laboratory.

Table 2: Location and characteristics of the sites where experimental soils were sourced

Site	**Sub-location**	**District**	**Latitude**	**Longitude**	**Altitude (masl)**	**Soil type**	**Average Rainfall and temperature (per annum)**
Masaba Central	Masaba	Butere	034^0 27’ 38.2’’E	00^0 11’59.9’’N	1331	Chromic Acrisols	1685 – 1882 mm 13.9 – 30.2^{0}C
Kakamega	Khwisero	Kakamega south	034^0 40’ 20.4’’E	00^0 12’ 26.0’’ N	1488	Ferralo-humic Acrisols	1730 – 1929 mm 14.1 – 27.1^{0}C
Kakamega	Shikhulu	Kakamega South	034^0 40’ 05.4’’ E	00^0 12’ 14.6’’ N	1508	Ferralo-humic Acrisols	1730 – 1929 mm 14.1 – 27.1^{0}C
Butere	Emutsatsa	Butere	034^0 27’ 56.9’’ E	00^0 11’ 51.35’’ N	1344	Chromic Acrisols	1685 – 1882 mm 13.9 – 30.2^{0}C
Butula	Bukhalalire	Butula	034^0 16’ 48.9’’ E	00^0 19’ 11.8’’ N	1219	Rhodic Ferralsols	1790 – 2016 mm 15.8 – 28.6^{0}C

Source: Jaetzold and Schmidt, (2005).

3.2.3 Nutrient treatments

The composition of the nutrient solutions were based on Hoagland half strength solution (Hoagland and Arnon, 1950), according to specific requirements of soybean. The ion concentrations were in milli-moles per litre (mmol/ l) for the macro-nutrients and micro-moles per litre (µmol/l) for the micro-nutrients (Table 4). The treatments included one positive control (complete nutrient solution), negative control (only distilled water), 5 treatments where one element was omitted from the nutrient solution, one treatment where the micro-nutrient mixture was omitted from the nutrient solution, one treatment with an additional nitrogen source in the nutrient solution and one treatment with lime addition to the soils (Table 3). The nutrients tested were P, K, Mg, Ca, S, and micronutrients (B, Mo, Mn, Cu and Zn) (Table 4). The purpose of the complete plus N treatment was to help in assessing whether the poor performances of plants with omitted elements was because of the element in question alone or also due to nitrogen limitation. This is also because of the nitrogen deficiencies observed in earlier double pot experiments with soybean as test crop (Wietske, 2012; Foli, 2012). Lime treatment was also included because all the experimental soils were acidic. Lime and the plus nitrogen treatments were added only to the complete treatment and were compared to the complete treatment during analysis of the results. The lime requirements of the soils were determined based on Rutgers, (2013) with the target pH of 6.2.

Table 3: Treatments used in the experiment

No.	Treatment	Macro nutrients						Micronutrients	Lime
		N	P	K	Mg	Ca	S		
1	Complete	-	+	+	+	+	+	+	-
2	Complete plus lime	-	+	+	+	+	+	+	+
3	Control	-	-	-	-	-	-	-	-
4	P omitted	-	-	+	+	+	+	+	-
5	Complete plus N	+	+	+	+	+	+	+	-
6	K omitted	-	+	-	+	+	+	+	-
7	Mg omitted	-	+	+	-	+	+	+	-
8	Ca omitted	-	+	+	+	-	+	+	-
9	S omitted	-	+	+	+	+	-	+	-
10	Micro-nutrients omitted	-	+	+	+	+	+	-	-

KEY: + (Nutrient included in the nutrient solution)

- (Nutrient omitted from nutrient solution)

3.2.4 Nutrient salts used in the experiment

A variety of salts (Table 4) were used to make nutrient solutions that were used to supply nutrients to soybean plants.

Table 4 : Nutrient salts and rates used to prepare nutrient solutions

Element	Salts	Rate of application
Macro – nutrients		**(milli mols/l)**
N and Ca	$Ca(NO_3)_2.4H_2O/NH_4NO_3/$ $CaCl_2.2H_2O$	N= 7.5 Ca = 2.5
P	H_3PO_4	P = 0.5
Mg, S and K	$MgSO_4/K_2SO_4$	Mg = 1 SO_4^- = 1 K = 3
Micro – nutrients		**(Micromoles/l)**
B	H_3BO_3	7.13
Mo	$Na_2MoO_4\text{-}2H_2O$	0.01
Zn	$ZnSO_4$	0.96
Cu	$CuSO_4$	1.04
Mn	$MnCl_2$	7.4

Source: Paradiso *et al.*, (2012); modified from Hoagland solution (Hoagland and Arnon, 1950).

Sodium hydroxide and hydrochloric acid were used to regulate the pH of the nutrient solutions.

3.2.5 Management of experiment

Soybean variety TGX 1740-2F (SB19) was planted in the experiment. The seeds were inoculated using the commercial Biofix inoculant containing *Bradyrhizobium japonicum* strain USDA 110 which was acquired from MEA fertilizers ltd. Three seeds were sown per pot and were thinned to single uniform plants per pot 7 days after emergence. After thinning, soils in the pots were covered with gravel to reduce evaporation. The pots were watered daily with distilled water to keep the soils at field capacity. The nutrient solution was renewed at 3 weeks after planting by pouring out the old solution and adding a new solution into the container. To buffer the pH of the alkaline nutrient solutions one 1 molar HCl was used while 0.1 molar NaOH was used to buffer the pH of the acidic nutrient solution (-K). The target pH was 6.5.

3.2.6 Experimental design

The greenhouse experiment was laid out in a Completely Randomized Design (CRD) whereby the treatments were replicated four times. To allow for the destructive sampling at three time intervals, extra pots were included. There were 120 pots per soil (10 treatments x 4 replications x 3 extra pots per replication) and therefore there were a total of 600 pots for the five soils. Each pot had 250 g of soil leading to a total of 150 kg for all the soils. The treatments were randomly allocated to the pots (experimental units). The experimental factors were the soils, nutrient treatments and the lime treatment.

3.2.7 Data collection

The data collected from the double pot experiment included; stem heights and shoot dry weights. Initial measurements were taken 14 days after emergence (DAE) and thereafter repeated every 7 days (14, 21 and 28 DAE). The destructive sampling was done at the three time intervals to help in determination of the relative growth rate and nutrient sufficiency quotients. The plants samples were oven dried at 65^0C for 48 hrs to determine the dry weights. After drying, the plant samples at the final harvest were ground. The four samples from each replicate representing one treatment were aggregated. The samples were then sent to Crop Nutrition Laboratory Services Ltd, Nairobi for elemental analysis using Inductively Coupled Plasma (ICP). The analysis carried out established, N, P, K, Ca, Mg, Mn, B, Zn and Cu concentrations in the plant samples. Visual observations for any deficiency symptoms were also carried out daily from the 8th day after emergence to the end of the experimental period.

3.2.8 Determination of relative growth rate and nutrient sufficiency quotients (SQ's)

The relative growth rate based on shoot dry weights was calculated between two time intervals 14 -21 DAE and 21 – 28 DAE. This reflected the net growth of the plants growing on different nutrient solutions. The relative growth rates obtained were then used to calculate the nutrient sufficiency quotients, to compare treatments with specific nutrient omission and the complete treatment. The SQ's were then multiplied by 100 to reflect the percentage growth of a given treatment compared to the complete treatment and thus help show the extent of limitation of a given element in each soil. The formulas used in calculation were;

$Rs = (\ln S2 - \ln S1) / (t2 - t1)$ (Janssen, 1974)

Where; Rs = Relative growth rate, S = shoot dry weights in g and t = time in days

The following formula was used to calculate SQ’s;

SQ_K = (RS)-x/ (RS) C (Seitz, 2013; Foli, 2012)

Where; SQ_x = Sufficiency quotient for x, where x is the nutrient element in question, (RS)-x = Relative growth rate of plants growing in nutrient solutions with x (nutrient element in question) omission and RS(C) = Relative growth rate of plants growing on complete nutrient solution.

3.2.9 Data analysis

Analysis of variance was done to compare the effects of the different treatments on growth and performance of soybean using Genstat 12th edition. The effects of the different treatments were compared by computing the least square means and their standard errors of difference (SED). All the single nutrient treatments were compared to the complete treatments (Janssen, 1974) using the least significant differences to establish their extent of limitation. If a treatment was significantly lower than the complete treatment, then it was considered to be limiting in the soils. Significance of the difference was evaluated at $P \leq 0.05$ significant level. Lime and nitrogen treatments were compared to the corresponding treatment (complete) and not to the other nutrient treatments. Each soil was analyzed separately to identify limiting nutrients specific to that soil.

The model that was used for analysis was;

$Y_{ij} = \mu + T_i + \varepsilon_{ij},$ Where μ= overall mean $Ti_{=}$ treatment effect and ε_{ij} = Overall experimental error

To analyze for the relative growth rate, time was considered as a factor and therefore the following model was used;

$Y_{ij} = \mu + T_i + T_j + C_{ij} + \varepsilon_{ij},$ Where μ= overall mean, $T_{i\,=}$ treatment effect, T_j = time effect, C_{ij} = interaction effect of treatments and time and ε_{ij} = Overall experimental error

3.3 Field experiments

The field experiments were carried out in Butere (Masaba central) and Butula. Site characteristics of the 2 districts are described in (Table 2). The sites were chosen based on the initial soil characterization and also based on the results from the green house experiment. Apart from P, K and Mg, both sites showed micro-nutrient limitations and thus there was need to test their response alongside other nutrients (P, K, Ca, Mg and S).

3.3.1 Description of materials used

3.3.1.1 Sympal

Sympal is a new fertilizer blend for legumes from MEA ltd, Kenya and its composition is; 23% P_2O_5, 15% K_2O, 10% Ca, 4% S and 1% MgO. Being relatively new in the market, there is need to test the efficiency of this fertilizer.

3.3.1.2 Farmyard manure

The farmyard manure used in the experiment was obtained from Baraton University's farm. It had the following characteristics (Table 5).

Table 5: Chemical characteristics of farmyard manure used in the experiment

pH (H_2O)	Mg cmol/kg	Ca cmol/kg	K cmol/kg	P %	S mg/kg	N %
8.74	39.17	41.25	41.15	0.85	235	2.05

3.3.1.3 Boric acid and sodium molybdate 2-hydrate

Boric acid (H_3BO_3) and sodium molybdate 4-hydrate (Na_2MoO_4-$2H_2O$) were used as a source of boron and molybdenum respectively. They were also used to provide the same elements in the double pot experiment.

3.3.1.4 Calcium oxide

Calcium oxide also known as quicklime was used to raise the soil pH of the research sites. CaO has a calcium carbonate equivalence of 150-175%.

3.3.1.5 Biofix

Biofix is a legume inoculant containing *Bradyrhizobium japonicum* strain USDA 110. It is a product of University of Nairobi manufactured by MEA ltd, Nakuru.

3.3.2 Experimental design and treatments

The experiment was laid out in a Randomized Complete Block Design with three replications. The treatments were; **i)** Control**, ii)** Sympal alone, **iii)** Farmyard manure alone**, iv) S**ympal plus farmyard manure**, v)** Sympal plus micro-nutrients**, vii)** Farmyard manure plus micro-nutrients**, viii)** Farmyard manure plus lime**, ix)** Sympal plus lime**, x)** Sympal plus micro-nutrients plus lime. One variety of soybean (SB 19) was planted in plots measuring 4.5 m by 3 m and separated by a path of 0.5 m. Sympal and farmyard manure were applied at a rate of 30 kg P/ha (0.41 kg of Sympal and 5.4 kg of farmyard manure per 3.5 m^2). Lime was applied at the rate of 2 t/ha (2.7 kg per 13.5 m^2). The rates used for B and Mo were 0.44 kg B / ha (0.60 g per 13.5 m^2) and 0.57 kg Mo / ha (0.8 g per 13.5 m^2) respectively as per Cooke (1982). Biofix was applied at the rate of 10 g/ kg of seed. This was done by wetting the seeds with the gum Arabic followed by mixing with the Biofix. The seeds were then planted immediately to avoid drying and death of the bacteria.

3.3.3 Trial establishment and management

Land preparation was done by ploughing to a depth of 15-20 cm using a hand hoe. The experiment was established at the onset of the rainy season (28th March 2013) at a spacing of 45 cm by 5 cm giving a population of 444,444 plants / ha. Sympal was applied by banding at the time of planting i.e. 2-5 cm from the planting rows to avoid direct contact of seed with fertilizer. Farmyard manure and lime were also applied just before planting by broadcasting followed by incorporation into the soil by light tillage using a hand hoe. The micro-nutrients were applied as foliar sprays in split application at 4 weeks and 6 weeks after emergence to avoid leaf damage.

All the soybean seeds used in the experiment were inoculated with Biofix inoculant before planting. The experimental plots were kept weed free by weeding twice using a hand hoe. Earthing up was done for the plots with lime to raise the soil and thus prevent spillage of lime and other treatments to adjacent plots.

3.3.4 Data collection

Before the onset of the experiment, information about the sites was collected and this included the global positioning system (GPS) readings (Table 2). At 50% podding, the above ground biomass from an area 0.5 m^2 in each plot was dug out of the soils with nodules and roots intact and separated into above (stems) and below ground (roots) parts. The above ground fresh biomass was weighed in the field and samples taken to the laboratory for oven drying followed by determination of the dry weights. The roots were washed, nodules detached, dried and dry weight determined. All nodules were sliced before drying to determine whether they were actively fixing nitrogen. Active nodules are pink, red or brown, while non-active nodules are white or light green (Vanlauwe *et al.*, 2003). Harvesting was carried out in the net plot of 5 m^2. The measurements carried out during harvesting included; Height of ten randomly selected plants, number of pods per plant, total stover + haulm yield, and grain yield.

3.3.5 Data analysis

Analysis of variance was also done to compare the effects of the treatments on soybean performance and on soil pH, available P and total N using GENSTAT 12th edition. The effects of the different treatments were also compared by computing their least square means and their standard errors of the difference (SED). Separate analysis was done per site. The model that was followed for data analysis was;

$Y_{ij} = \mu + T_i + R_j + \varepsilon_{ij}$; Where μ = overall mean T_i = treatment effect, R_j = replication/block effect and ε_{ij} = Overall experimental error

3.4 Laboratory soil analysis

Initial soil analysis was done for; pH, total nitrogen, total carbon, available phosphorus, exchangeable cations and particle size. Final soils analysis was done for pH for the green house experiment and soil pH, total soil N and available soil P for the field experiment.

3.4.1 Soil particle analysis

This was carried out by the hydrometer method to estimate the percentage of sand, silt and clay contents of the soils which is often reported as percentage by weight of oven-dry and organic matter-free soil. The analyses are usually performed on air-dry soil. Based on the proportions of different particle sizes, a soil textural category is assigned to the sample. The first stage in particle size analysis was the dispersion of the soil into the individual particles. These are the sand (2.00 – 0.05 mm), silt (0.05 – 0.002 mm) and clay (<.002 mm) fractions. Individual soil particles are often bound into aggregates hence the requirement for dispersion. The hydrometer method of silt and clay measurement relies in the effects of particle size on the differential settling velocities within a water column.

3.4.2 Soil pH

Measurements of pH are expressed as the inverse log of the hydrogen ion concentration. Soil pH was measured on 2.5:1 soil to water suspension on a glass electrode.

3.4.3 Extractable phosphorus

This was carried out using the Olsen, (1954) method. The principle behind this method is that the soil is extracted with 0.5 M solution of sodium bicarbonate. In calcareous, alkaline or neutral soils containing calcium phosphate, this extractant decreases the concentration of calcium in solution by precipitating Ca as $CaCO_3$. The result is an increase of the P concentration in the solution. In acid soils containing Al and Fe phosphate, the P concentration in the solution increases as the pH rises. Precipitation reactions in acid and calcareous soils are reduced to a minimum because the concentrations of Al, Ca and Fe remain at a low level in this extractant. Basically, the concentration of phosphates present is related to the blue complex produced by the reduction, in the presence of ascorbic acid, of the phosphomolybdate (molybdophosphoric acid) formed when acidic ammonium molybdate reacts with phosphate (Murphy and Riley, 1962), and is measured calorimetrically at 880 nm wavelength on a spectrophotometer.

3.4.4 Total nitrogen

The content of total nitrogen was measured in a digest obtained by treating soil and plant sample with hydrogen peroxide+sulpuric acid selenium and salicylic acid. The principle takes into account the possible loss of nitrates by coupling them with salicylic acid in an acid media to form 3-nitrosalicylic and or 4-nitrosalicylic. The compounds were reduced to their corresponding amino acid forms by the soil organic matter. The Analysis of total nutrients required complete oxidation of organic matter. The hydrogen peroxide oxidised the organic matter while the selenium compound acted as a catalyst for the process and the H_2SO_4 completed the digestion at elevated temperatures.

3.4.5 Exchangeable cations

These were determined using the Mehlich three stock solution methods. It involved the extraction of the cations from the soils using Mehlich 3 extracting solution (0.2 N CH_3COOH + 0.25 N NH_4NO_3 + 0.015 N NH_4F + 0.013 N HNO_3 + 0.001 M EDTA).The final filtrate was used to establish the concentrations of the cations. The amounts of exchangeable K, Ca and Mg were determined using Inductively Coupled Plasma (ICP) (Mehlich, 1984).

3.4.6 Organic carbon

Organic carbon was determined by the sulphuric acid and aqueous potassium dichromate ($K_2Cr_2O_7$) mixture. After complete oxidation from the heat of solution and external heating, the unused or residual $K_2Cr_2O_7$ (in oxidation) was titrated against ferrous ammonium sulphate. The used $K_2Cr_2O_7$, the difference between added and residual $K_2Cr_2O_7$, gave a measure of organic C content of the soil. The chemical reaction in the method is;

$$2Cr_2O_7^{2-} + 3C + 16\ H^{+} \rightarrow 4Cr^{3+}\ 3CO_2 + 8\ H_2O$$

3.5. Economic analysis

To determine the economically viable treatments, partial budgeting was carried out to compare the costs and the benefits of the treatments. Gross field benefits (GFBs) were calculated based on the adjusted yield (10% downwards the attained yield). Research has found out that farmers using the same technology would obtain yields which are 10% lower than those found by researchers (Kisinyo, 2011). The prices of output were based on 2013 market price of the soybean using the following formula;

GFBs = adjusted yield * Kshs. 55 (market price per kg)

The prevailing rates paid to the labourers at each site were used to calculate the labour costs that vary. The input and transport costs were also established based on the prevailing market prices and transport costs in each site. The total costs that vary (TCV) were then calculated follows;

TCV = labour costs + input costs + transport costs

The accruing net benefits (GFB – TCV) and costs that vary were than compared across the treatments in dominance analysis. This was based on the criterion that any treatments that had net benefit equal or lower than that of another treatment with lower costs that vary is dominated (D) and thus not considered for investment by the farmer. Marginal analysis was done on the un-dominated treatments. Marginal rate of return (MRR) was calculated as follows;

MRR = (marginal cost/ marginal net benefits) * 100.

A minimum rate of return of 50% was considered worthy for adoption by the farmers CIMMYT, (1988),

CHAPTER FOUR

RESULTS

4.1 Soil analysis

4.1.1 Initial soil characterization

The results in Table 6 indicate that the soils from all the sites were strongly acidic (4.50 to 5.00) except for the soils from Kakamega (Khwisero sub-location) which were moderately acidic (5.08). Masaba Central had the lowest pH value (4.65) followed by the soils from Butere. Soils from Butula and Kakamega south (Shikhulu sub-location) had the same pH value of 4.99. Percentage nitrogen levels were moderate for all the soils (0.12 to 0.25). The levels of extractable P were low in the soils from all the sites (< 10 mg/kg). Exchangeable potassium was available in low levels in the soils from all the sites (< 0.2 cmol/kg). The amount of exchangeable calcium was low (< 2 cmol/kg) in Masaba and Butere (1.10 and 1.03 cmol/kg respectively), while the levels were moderate (2 to 10 cmol/kg) in both sites from Kakamega and in Butula. Magnesium levels were moderate (1.70 cmol/kg) in the soils from Kakamega (Khwisero sub-location), and low (< 1 cmol/kg) in all the other sites.

The measured organic carbon contents were moderate (1.5 to 3%) in the soils from Kakamega and in Butula, while those from Masaba central and Butere had low (0.5 to 1.5%) levels of percentage carbon. All the soils fell in the sandy clay loam textural class except for those soils from Butere which were in the sandy loam class.

Table 6: **Physical and chemical properties of the soils used in the experiment.**

Parameters	**Masaba**	**Kakamega South (Khwisero)**	**Kakamega South (Shikhulu)**	**Butere**	**Butula**
pH (water)	4.65	5.08	4.99	4.91	4.99
Total N (%)	0.13	0.24	0.20	0.13	0.15
Available P (mg/kg)	4.43	7.27	4.58	2.11	6.18
Organic C (%)	1.50	3.13	2.59	1.25	1.58
C:N ratio	11.53	13.04	12.95	9.62	10.53
K (cmol/kg)	0.16	0.18	0.17	0.10	0.16
Ca (cmol/kg)	1.10	4.34	3.22	1.03	2.49
Mg (cmol/kg)	0.51	1.70	0.93	0.39	0.98
TEXTURE					
Sand (%)	54	48	52	60	58
Clay (%)	30	32	32	18	24
Silt (%)	16	20	16	22	14
Textural class	Sandy clay loam	Sandy clay loam	Sandy clay loam	Sandy loam	Sandy clay loam

4.2 Identification of the nutrients (macro and micro) that are limiting soybean production in Acrisols and Ferralsols of western Kenya

4.2.1 Visual observations

Daily visual observations were carried out from eight days after emergence. This was to help note any deficiency symptoms that were conspicuous. The common deficiency observed was that of magnesium where there was interveinal leaf yellowing especially in treatments of plus nitrogen and a few of magnesium omitted treatments across the soils (Plate 2A). Potassium deficiencies were also noted mainly on older leaves of potassium omitted treatments and also on several other treatments. These were characterized by leaf yellowing with tissue necrosis along the leaf margins (Plate 2B). There was an early leaf drop in the plants growing in the treatments with both magnesium and potassium deficient treatments, and this led to low biomass being recorded by the same treatments especially at the final harvest. Plants growing in the control (only distilled water) and in the treatments where phosphorus and calcium were omitted maintained green leaf colour to the end of the experimental period. Other treatments (complete, minus sulphur, minus micro-nutrients and complete plus lime) had pale yellow leaves at the time of harvesting (Plate 3G) indicating possible deficiencies of nitrogen.

Addition of nitrogen to the nutrient solution had a variable response across the soils. Soils from Kakamega and Butula produced very leafy green plants while in the other soils, the foliage was less and there was a burning characteristic at the edge of the leaves. There was a well developed rooting system in all the treatments (3E) except in calcium omitted treatments and also in some of the plus nitrogen treatments (3F). Before the plumule emerged from the soil, the radical had already penetrated through the wire gauze into the nutrient solution in all the treatments across all the soils. The roots of minus calcium treatment darkened with the progress of the experiment.

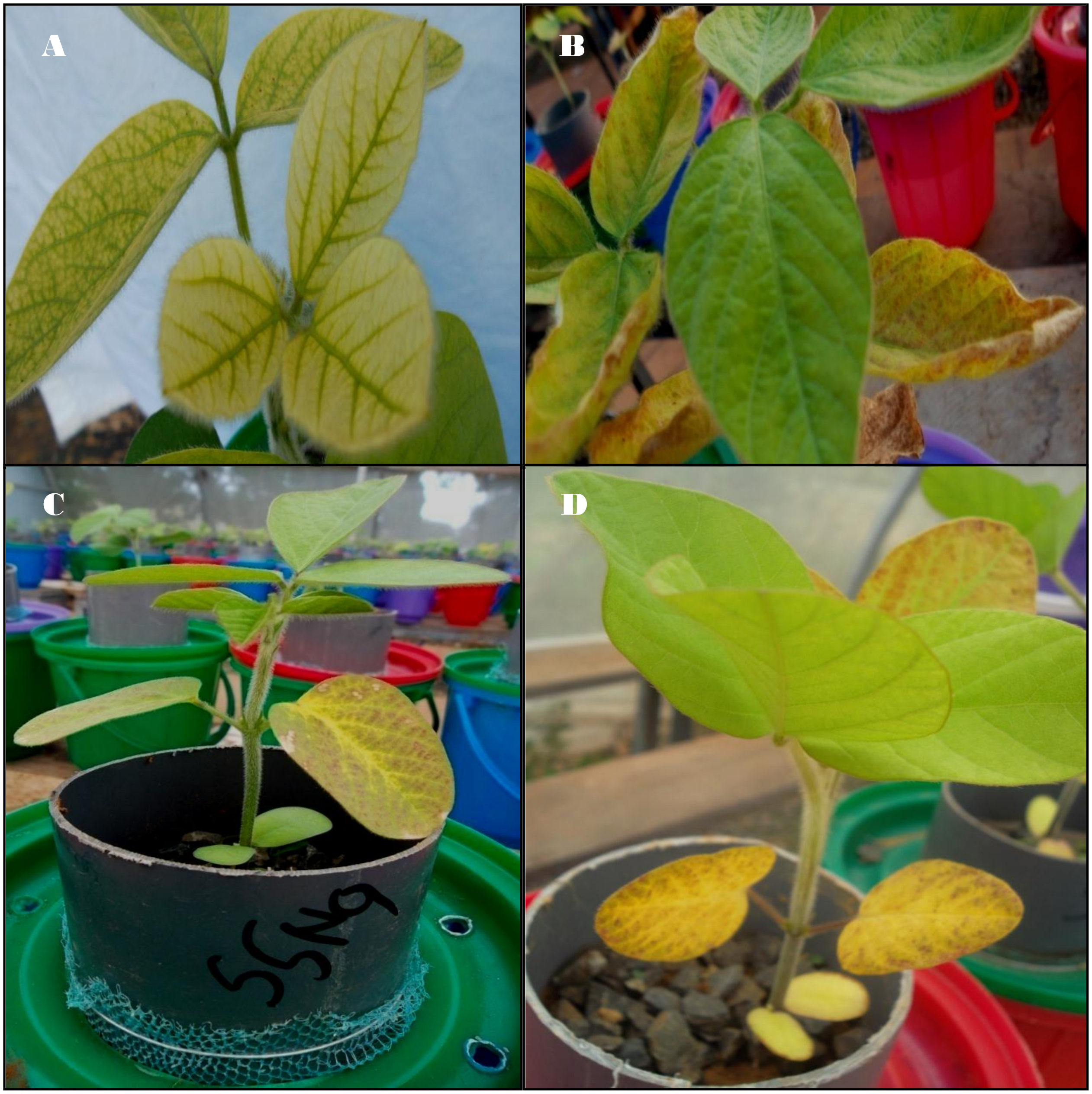

Plate 2: Nutrient deficiency symptoms noted during plant growth. A – Mg deficiency in plus nitrogen treatment in Kakamega (Shikhulu Sub-location), B – K deficiency in minus K treatments in Soils from Kakamega (Shikhulu sub-location), C – P deficiency in minus magnesium treatments in soils from Butula, D – Micro-nutrient deficiencies in minus micro-nutrient treatments in soils from Masaba Central.

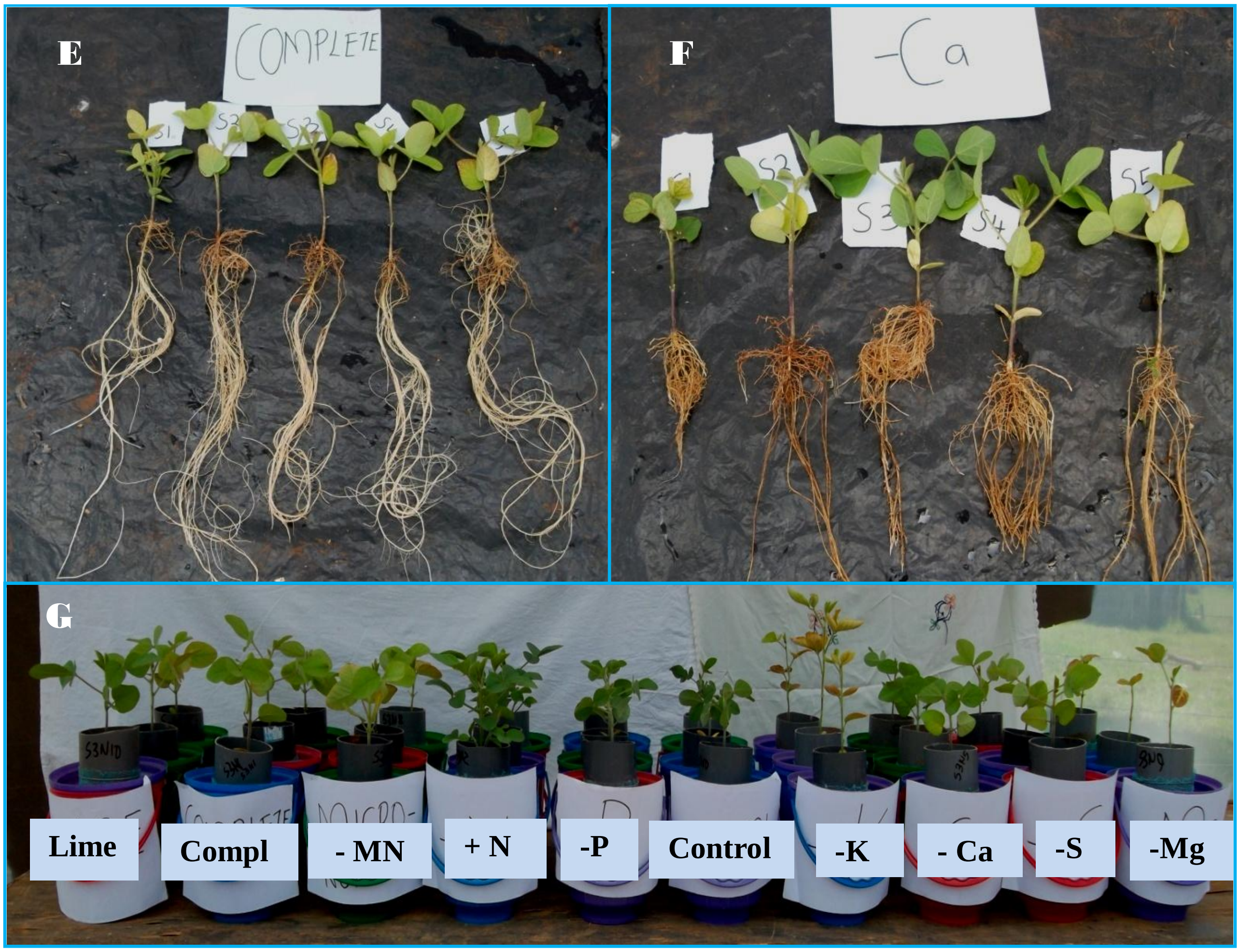

Plate 3: **Soybean plants at the time of harvest. E – Plants growing on complete nutrient solution on different soils, F – Plants growing on minus calcium nutrient solution on different soils, G – Plants growing on different nutrient treatments on soils from Kakamega (Shikhulu sub-location); Compl – complete treatment, MN – micro-nutrients.**

4.2.2 Trends of shoot nutrient concentration as influenced by different treatments in different soils.

Tables 7 show the shoot nutrient concentration in different soils. The nutrient concentration of the plant tissues analyzed at the final harvest was compared between the complete treatment, the control and the treatment with specific nutrient omission. Nitrogen added treatments accumulated high amounts of N in their plant tissues compared to the complete and control treatments in all the soils except those from Masaba central. The control treatment accumulated high amounts of N than the complete treatment in soils from Masaba central, Butere and Butula. Nutrient omission from the nutrient solutions led to their respective low accumulation in the plant tissues compared to the complete treatment in soils from all the sites. Omission Ca and Mg from the nutrient solution led to their low concentration than the control treatment in the plant tissues in all the soils. Phosphorus omission resulted in its low concentration in their plant tissues than the control treatment in soils from Kakamega (Khwisero and Shikhulu sub-locations) and Butere (Table 7). Liming improved nitrogen concentration than the complete treatment in soils from Masaba central, Kakamega (Shikhulu sub-location), Butere and Butula. It improved P concentration in plant tissues compared to the complete treatment only in soils from Kakamega (Shikhulu sub-location). Concentration of Mg in plant tissues was improved by liming compared to the complete treatment in all the soils.

Generally, elimination of Ca from the nutrient solution led to low accumulation of several elements e.g. P, N, Ca and all micro-nutrients in the shoots across all the soils. Omission of K and Mg from the nutrient solution led to increased concentration of the other elements in all the soils.

Table 7: Nutrient concentration in plant tissues in soils from different sites.

		%					ppm			
Sites	**Treatments**	**N**	**P**	**K**	**Mg**	**Ca**	**Mn**	**B**	**Zn**	**Cu**
Masaba central	Control	3.51	0.42	1.69	0.29	0.87	706.00	28.70	55.30	5.16
	Treatment	2.10	0.82	0.97	0.14	0.29	513.00	25.50	22.70	3.87
	Complete	2.25	0.95	2.92	0.41	1.51	679.00	41.00	56.30	6.79
	Plus lime	2.31	0.70	2.23	0.32	1.61	183.00	36.70	25.20	4.00
Kakamega (Khwisero Sub-location)	Control	1.88	0.08	0.62	0.44	0.76	259.00	31.20	48.10	3.08
	Treatment	5.72	0.07	0.78	0.37	0.47	230.00	28.20	36.60	3.08
	Complete	2.57	0.82	2.21	0.47	1.63	232.00	37.30	51.40	3.48
	Plus lime	1.52	0.75	2.17	0.40	1.42	140.00	37.20	43.30	4.57
Kakamega (Shikhulu Sub-location)	Control	1.88	0.07	0.61	0.36	0.72	218.00	24.70	52.40	2.68
	Treatment	4.04	0.06	1.24	0.26	0.52	170.00	31.20	45.30	2.37
	Complete	1.94	0.60	2.21	0.36	1.34	189.00	32.50	48.20	2.62
	Plus lime	1.99	0.81	2.62	0.42	1.76	153.00	43.30	52.50	3.77
Butere	Control	2.57	0.12	0.64	0.32	0.59	758.00	20.50	55.40	3.41
	Treatment	4.30	0.07	1.14	0.19	0.26	819.00	29.50	24.70	4.00
	Complete	1.83	0.78	2.60	0.38	1.38	647.00	31.20	36.30	5.58
	Plus lime	2.10	0.64	2.02	0.31	1.52	192.00	34.90	27.30	2.29
Butula	Control	2.73	0.06	0.58	0.27	0.48	198.00	19.40	35.80	2.25
	Treatment	4.14	0.06	1.78	0.25	0.41	314.00	29.80	34.10	2.50
	Complete	2.15	0.82	2.34	0.37	1.38	190.00	32.60	42.10	3.16
	Plus lime	3.20	0.78	2.30	0.33	1.58	113.00	34.50	30.20	3.27

Complete (all macro (except N) and all micro-nutrients), Control (only distilled water), Treatment (specific nutrient omission from the nutrient solution, for plus N treatment, it shows N addition to the nutrient solution) and Plus lime (treatment with lime addition to the soils).

4.2.3 Plant height

The treatments differed significantly ($P \leq 0.05$) in the soils from Masaba central, Kakamega (Khwisero and Shikhulu sub-locations) and ($P \leq 0.001$) in soils from Butere and Butula in terms of plant height in the final harvest. Omission of Ca, Mg and K from the nutrient solutions in the soils from Masaba central led to significantly lower plant heights compared to the complete treatment (Table 8). In the soils from Kakamega (Khwisero), omission of Mg and P from the nutrient solution led to significantly lower plant heights than the complete treatment while in soils from Shikhulu Sub-location, P omission led to significantly lower plant heights compared to the complete treatment. Omission of K from the nutrient solution in the soils from Butere led to significantly ($P \leq 0.05$) lower plant heights than the complete treatment. In the soils from Butula, omission of Ca, K, Mg, P and S from the nutrient solutions led to significantly lower plant heights compared to the complete treatment (Table 8).

Table 8: Plant heights (cm) at final harvest (28 DAE) for the different treatments in the soils from the different sites.

		Kakamega			
	Masaba	**Khwisero**	**Shikhulu**	**Butere**	**Butula**
Complete	18.15^{c}	19.10^{c}	18.48^{c}	16.05^{cd}	19.33^{e}
Control	12.57^{a}	13.90^{a}	13.33^{a}	11.68^{a}	13.60^{a}
Minus Ca	15.00^{ab}	18.10^{bc}	16.82^{bc}	17.43^{d}	17.70^{d}
Minus K	14.43^{ab}	18.55^{bc}	17.77^{bc}	12.85^{ab}	14.57^{ab}
Minus Mg	14.60^{ab}	15.93^{ab}	15.72^{abc}	14.38^{bc}	16.05^{bc}
Minus MN	16.43^{bc}	20.30^{c}	17.32^{bc}	17.95^{d}	17.77^{de}
Minus P	17.15^{bc}	16.05^{ab}	15.25^{ab}	14.30^{bc}	15.38^{b}
Minus S	15.82^{bc}	19.35^{c}	18.23^{bc}	17.52^{d}	17.18^{cd}
F. Probability	0.019	0.001	0.028	<.001	<.001
LSD	2.929	2.758	3.035	2.214	1.617
% C.V.	12.9	10.7	12.5	9.9	6.7

Statistical analysis and treatment comparisons done per soil (along the column). Means followed by the different letters are significantly different (LSD, $P \leq 0.05$) MN – micronutrients.

4.2.4 Effects of treatments on shoot dry weights at final harvest.

The treatments differed significantly ($P \leq 0.001$) from each other in all the soils except in soils from Kakamega (Khwisero sub-location) which differed significantly at ($P \leq 0.05$) from each other with respect to shoot dry weights. The control treatments had significantly lower shoot dry weights compared to the complete treatment in all the soils (Table 9). In soils from Masaba central, omission of K, Mg, P, Ca and micro-nutrients resulted in significantly lower shoot dry weights compared to the complete nutrient treatment. Omission of S from the nutrient solution resulted in non-significantly ($P \leq 0.05$) different shoot dry weight to that of the complete nutrient treatment. When Mg and P were excluded from the nutrient solution in soils from Kakamega (Khwisero sub-location), there was significantly lower shoot dry weights compared to that of the complete treatment. There was no significant difference in shoot dry weights between the complete treatment and when K, Ca, S and micro-nutrients were omitted from the nutrient solutions with Ca, S and micro-nutrients exclusion resulting in higher dry weights than the complete treatment.

In soils from Kakamega (Shikhulu sub-location), omission of K, P and Mg resulted in significantly lower shoot dry weights compared to the complete treatment (Table 9). There were no significant differences when Ca, S and micro-nutrients were omitted from the nutrient solution from the complete treatment. In Butere, significantly lower yields were obtained when K and Mg were omitted from the nutrient solution. Omission of Ca and micro-nutrients resulted in significantly higher shoot dry weights than the complete treatment. Omission of S and P nutrients resulted in shoot dry weights that were not significantly ($P \leq 0.05$) different from the complete treatment (Table 9). When K, P, Mg and micro-nutrients were omitted from the nutrient solution in soils from Butula, there were significantly lower shoot dry weights compared to the complete treatment. Omission of Ca and S resulted in shoot dry weights that were not significantly different from that of the complete treatment with calcium omission having higher shoot dry weights (Table 9).

Omission of K resulted in lower shoot dry weights than the control (distilled water only) treatment in soils from Masaba central, Kakamega (Shikhulu sub-location), Butere and Butula. However, omission of K and the control (distilled water only) treatment were not significantly different from each other. Omission of Mg resulted in lower shoot dry weights than the control treatment (distilled water only) in soils from Kakamega (Shikhulu and Khwisero sub-locations) and in Butula (Table 9).

Table 9: Biomass (g/plant) for the different treatments in the soils from different sites.

		Kakamega			
	Masaba	**Khwisero**	**Shikhulu**	**Butere**	**Butula**
Complete	0.52^{d}	0.57^{bcd}	0.59^{c}	0.37^{cd}	0.57^{de}
Control	0.27^{b}	0.38^{a}	0.41^{b}	0.22^{ab}	0.38^{bc}
Minus Ca	0.37^{bc}	0.68^{cd}	0.57^{c}	0.56^{e}	0.64^{e}
Minus K	0.15^{a}	0.49^{abc}	0.28^{a}	0.15^{a}	0.20^{a}
Minus Mg	0.29^{b}	0.33^{a}	0.29^{a}	0.25^{ab}	0.29^{ab}
Minus MN	0.41^{c}	0.70^{d}	0.48^{bc}	0.49^{e}	0.43^{c}
Minus P	0.41^{c}	0.40^{ab}	0.45^{b}	0.28^{bc}	0.38^{bc}
Minus S	0.43^{cd}	0.62^{cd}	0.48^{bc}	0.48^{de}	0.48^{cd}
F. probability	<.001	0.002	<.001	<.001	<.001
LSD	0.1091	0.1882	0.1214	0.1167	0.1332
% C.V.	20.8	24.8	18.8	22.9	21.7

Statistical analysis and treatment comparisons done per soil (along the column). Means followed by the different letters are significantly different (LSD, $P \leq 0.05$) MN – micronutrients.

4.2.5 Effect of liming and nitrogen application on shoot dry weights in different sites.

There was significantly (P ≤ 0.05) high shoot dry weights in treatments with lime addition in the soils from Masaba central and Butere. Liming did not significantly influence the shoot dry weight accumulation in Kakamega (Khwisero and Shikhulu sub-location) and in Butula. The lime added treatments had higher biomass as compared to the complete treatment in Kakamega (Shikhulu Sub-location) and Butula (Figure 1). Addition of nitrogen to the nutrient solution increased shoot dry weights significantly in soils from Kakamega (Khwisero and Shikhulu sub-locations) and in Butula. In Masaba central, the complete treatment had significantly higher shoot dry weights compared to the plus nitrogen treatment, while in Butere, the two treatments had the same shoot dry weights (Figure 2).

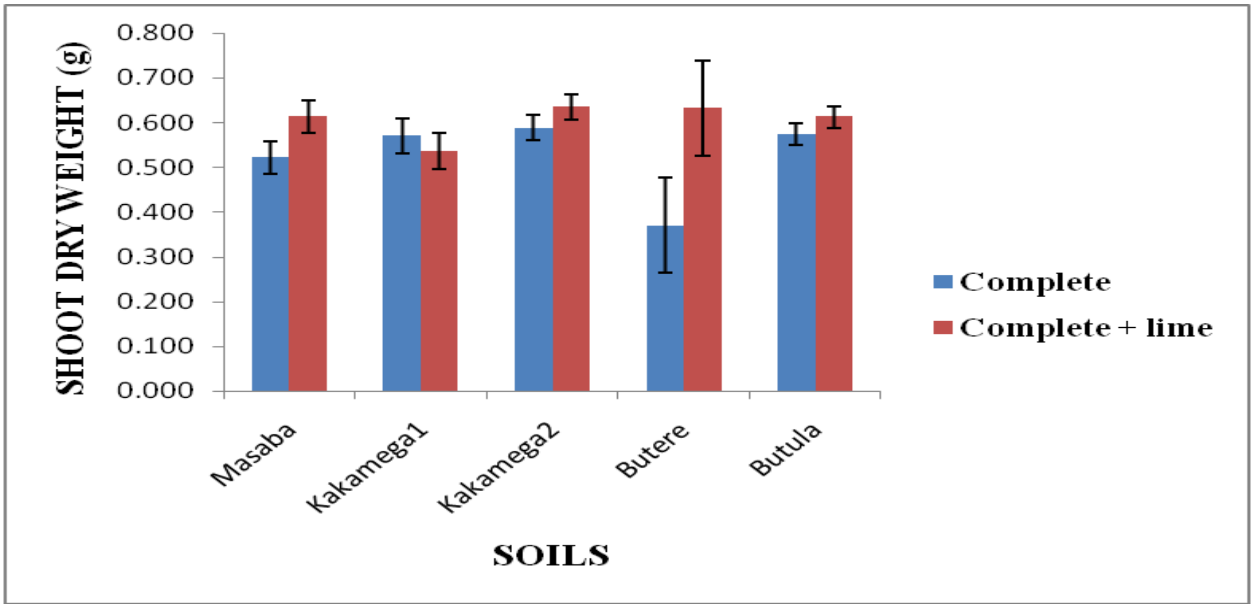

Figure 1: Effects of liming on soybean shoot dry weights in soils from the different sites. Kakamega1 (Khwisero sub-location), Kakamega2 (Shikhulu sub-location), Error bars represent LSD.

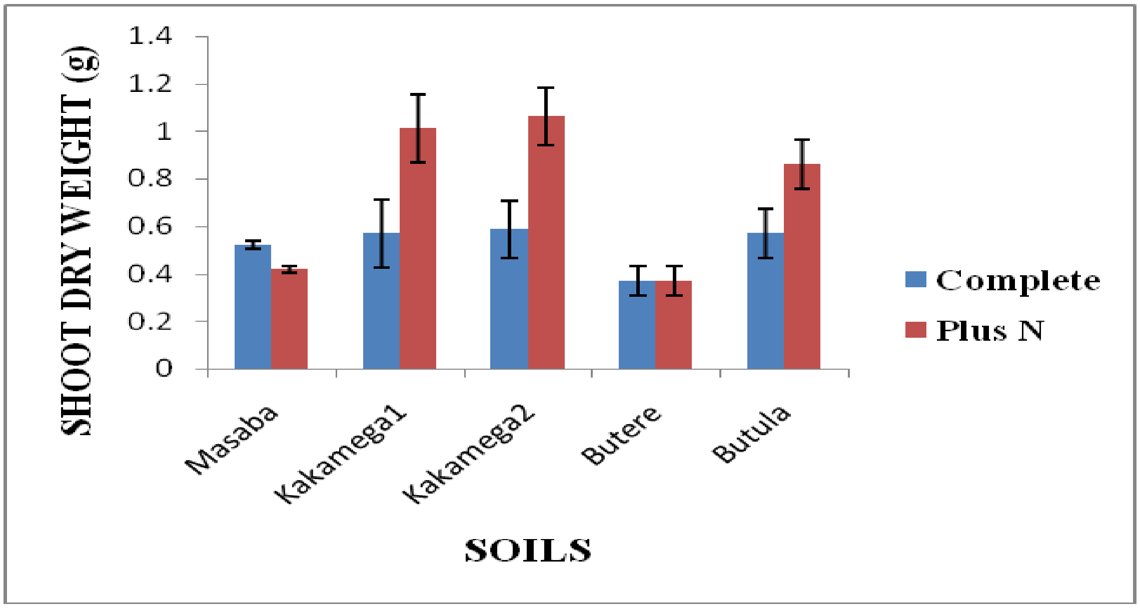

Figure 2: **Effects of nitrogen application to the nutrient solution on soybean shoot dry weights in soils from the different sites. Kakamega1 (Khwisero sub-location), Kakamega2 (Shikhulu sub-location), the error bars stand for LSD.**

4.2.6 Nutrient sufficiency quotients.

The sufficiency quotients were multiplied by 100 to show the percentage growth of a nutrient treatment compared to the complete treatment. From this experiment, omission of Ca from the nutrient solution in soils from Kakamega (Shikhulu sub-location), micro-nutrients in the soils from Kakamega (Khwisero sub-location) and Butere and S omission from the nutrient solution in the soils from Kakamega (Khwisero sub-location) and Butula had sufficiency quotients greater than 100% (Table 10). Omission of Mg and K from the nutrient solution led to lowest sufficiency quotients in soils from all the sites. The sufficiency quotients of minus K were negative in Butula while those of minus Mg were negative in Kakamega (Shikhulu sub-location) (Table 10). The control treatment had low sufficiency quotients in Masaba central and Butere but these values were higher than those of minus Mg and minus K. In soils from the other sites, they had higher sufficiency quotients, even greater than P omitted treatments in Kakamega (Khwisero and Shikhulu sub-locations) and in Butula. Also greater than minus Ca and minus micro-nutrients in Butula (Table 10).

Lime addition to the soils led to higher nutrient sufficiency quotients than the complete treatment in soils from Butere and Butula. In soils from Masaba central and Kakamega (Khwisero and Shikhulu Sub-locations) the sufficiency quotients of the limed treatments were lower than those of the complete treatments. Nitrogen application to the nutrient solution increased the nutrient sufficiency quotients than the complete treatment in all the soils except those from Masaba central (Table 10).

Table 10: Percent nutrient sufficiency quotients for different treatments in soils from the different sites.

		Kakamega			
SITE/TREATMENT	**Masaba**	**Khwisero**	**Shikhulu**	**Butere**	**Butula**
Complete	100	100	100	100	100
Control	42	85	74	42	96
Minus Ca	85	97	113	79	94
Minus K	18	64	48	1	-4
Minus Mg	35	28	-1	50	30
Minus micro-nutrients	69	126	77	124	92
Minus P	68	68	71	49	82
Minus S	78	122	91	86	106
Plus lime	83	82	84	129	112
Plus N	61	178	145	115	110

4.2.7 Soil differences in terms of shoot dry weights (g) and plant heights (cm) at the time of harvest.

Soils from Kakamega (Khwisero sub-location) had significantly ($P \leq 0.05$) higher shoot dry weights than soils from all other sites. It also had higher heights compared to the other soils and was not significantly different from the soils of Kakamega (Shikhulu sub-location). Butere soils had lowest plant heights and shoot dry weights as compared to the other soils but was not significantly different from the soils of Masaba central in plant height, and from Masaba central and Butula in shoot dry weights (Figures 3a and 3b).

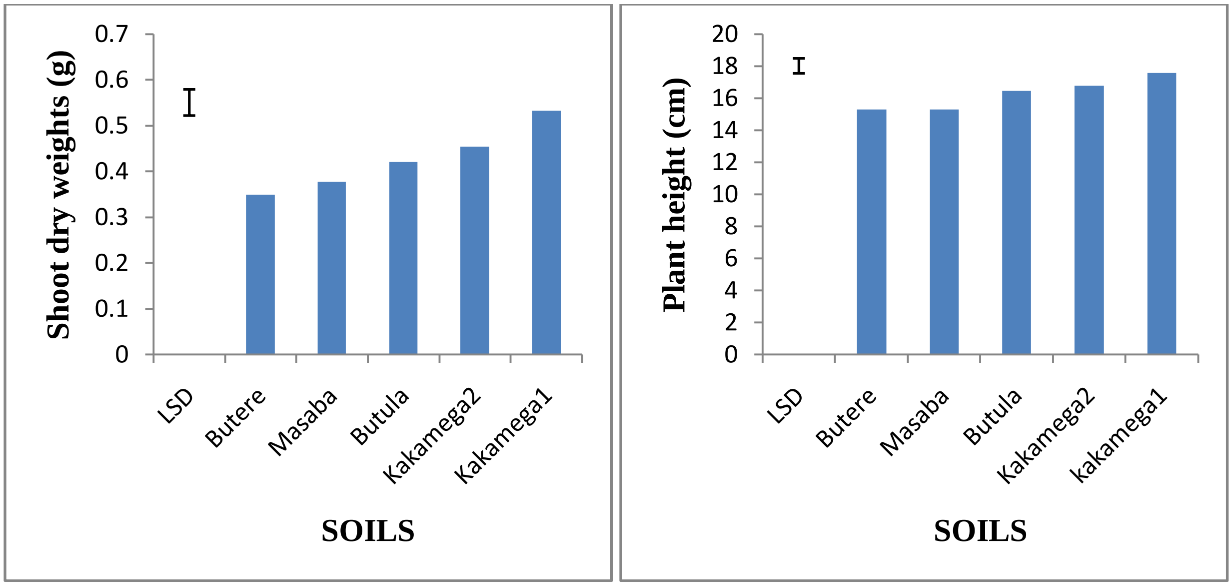

Figure 3a **Figure 3b**

Figure 3: **Mean shoot dry weights (g) and plant heights (cm) across different sites. Kakamega1 (Khwisero Sub-location), Kakamega2 (Shikhulu Sub-location). The bars in the graphs represent LSD.**

4.2.8 Effects of the treatments on soil pH.

The treatments differed significantly ($P \leq 0.05$) in terms of soil pH in all the sites except in Kakamega (Khwisero and Shikhulu sub-locations). Application of lime to the soils from Masaba central, Butere and Butula raised the soil pH significantly ($P \leq 0.05$) compared to the other treatments. The soil pH of Mg omitted treatment in soils from Butula did not differ significantly from the lime treatment. In soils from Kakamega (Khwisero and Shikhulu sub-locations), addition of lime to the soils led to higher soil pH values but this was not significantly different from the other treatments (Table 11).

Table 11: Effects of the different treatments on soil pH of the soils from different sites.

		Kakamega			
	Masaba	**Khwisero**	**Shikhulu**	**Butere**	**Butula**
Complete	4.64^{a}	5.14^{a}	5.18^{a}	4.89^{a}	4.91^{ab}
Control	4.55^{a}	5.12^{a}	5.16^{a}	4.81^{a}	5.13^{b}
Minus Ca	4.56^{a}	5.25^{a}	5.14^{a}	5.12^{b}	5.03^{b}
Minus K	4.60^{a}	5.12^{a}	5.09^{a}	4.80^{a}	5.05^{b}
Minus Mg	4.67^{a}	5.19^{a}	5.06^{a}	4.85^{a}	5.20^{bc}
Minus micro-nutrients	4.68^{a}	5.05^{a}	4.75^{a}	4.91^{ab}	4.98^{b}
Minus P	4.70^{a}	5.13^{a}	4.91^{a}	4.97^{ab}	4.91^{ab}
Minus S	4.66^{a}	5.06^{a}	5.13^{a}	4.84^{a}	4.96^{ab}
Plus lime	5.07^{b}	5.30^{a}	5.24^{a}	5.41^{c}	5.53^{c}
Plus N	4.54^{a}	5.02^{a}	4.81^{a}	4.78^{a}	4.63^{a}
LSD	0.1965			0.2170	0.3457
F. Probability	0.005	NS	NS	0.002	0.013
% C.V.	1.9	2.1	3.4	2	3.1

Statistical analysis and treatment comparisons done per soil (along the column). Means followed by the different letters are significantly different (LSD, $P \leq 0.05$) MN – micronutrients.

4.3 Effects of sympal and farmyard manure as sources of macro-nutrients (P, K, Mg, Ca and S) and micro-nutrients (B and Mo) application on selected soil chemical properties and soybean performance.

4.3.1 Effects of the treatments on selected soil chemical properties (pH, available P and total soil N).

The treatments differed significantly ($P \leq 0.05$) in terms of soil pH and total N and ($P \leq 0.001$) in terms of available P in Masaba central. In Butula, the treatments were not significantly different in terms of soil pH and total soil N. They were significantly different ($P \leq 0.001$) in terms of available P. In Masaba central, manure application led to high soil pH and was not significantly different from application of manure plus micro-nutrients and sympal plus micro-nutrients. Sympal application led to higher available soil P but was not significantly different from sympal plus manure, manure plus micro-nutrients and manure alone. The highest total soil N was obtained after manure application but was not significantly different from manure plus micro-nutrients application (Table 12).

In Butula, the highest soil pH was obtained after sympal application but was not significantly different from all the other treatments. Combination of sympal and manure led to high available soil P but was not significantly different from the application of sympal and sympal plus micro-nutrients. Sympal application led to high total N but was not significantly different from all the other treatments (Table 12).

Table 12: Effects of the treatments on soil pH, available P and total soil N in Masaba central and Butula.

	Masaba Central			**Butula**		
TREATMENTS	**pH (H2O)**	**P (mg/kg)**	**%N**	**pH (H2O)**	**P (mg/kg)**	**%N**
Control	4.70^{a}	9.31^{a}	0.16^{ab}	4.95^{a}	7.45^{a}	0.17^{a}
Sympal+manure	4.78^{ab}	13.71^{bc}	0.16^{ab}	5.22^{a}	13.04^{c}	0.18^{a}
Sympal	4.84^{ab}	14.06^{c}	0.15^{a}	5.35^{a}	11.79^{c}	0.19^{a}
Sympal+MN	4.88^{abc}	9.31^{a}	0.15^{a}	5.07^{a}	12.92^{c}	0.18^{a}
Manure+MN	4.96^{bc}	12.37^{bc}	0.17^{bc}	5.04^{a}	9.99^{b}	0.18^{a}
Manure	5.08^{c}	11.92^{bc}	0.18^{c}	5.11^{a}	9.79^{b}	0.18^{a}
F.probability	0.049	<.001	0.022	0.058	<.001	0.867
LSD	0.234	1.840	0.014	0.254	1.445	0.024
% CV	2.7	8.6	4.8	2.8	7.3	7.2

Different letters within each column shows significant difference to protected LSD ($P \leq 0.05$). MN – micro-nutrients.

4.3.1 Effects of the treatments on soybean nodulation.

The treatments differed significantly ($P \leq 0.05$) in terms of nodule number, nodule dry weight and number of active nodules in Masaba central. In Butula, the treatments differed significantly ($P \leq 0.05$) in terms of nodule number and number of active nodules and ($P \leq 0.001$) in terms of nodule dry weight (Table 13). Application of sympal plus micro-nutrients in Masaba central led to a high nodule number (30) and was not significantly different from application of sympal plus manure (28 nodules), manure plus micro-nutrients (21 nodules) and manure alone (19 nodules). Application of sympal plus manure led to highest nodule dry weight (2.47 g) and was not significantly different from application of manure alone. Application of sympal plus manure significantly increased the number of active nodules and was not significantly different from application of micro-nutrients in combination with sympal and manure (Table 13).

In Butula, application of sympal led to higher nodule numbers (64) and was not significantly different from the application of manure alone and sympal plus manure. Manure alone led to higher nodule dry weights (4.03 g) and was not significantly different from the application of sympal alone and sympal plus manure. Application of manure plus micro-nutrients and sympal plus micro-nutrients did not differ significantly from each other. Application of sympal alone led to significantly higher number of active nodules and was not significantly different from application of sympal plus manure and manure alone (Table 13).

Table 13: Effects of the treatments on nodule number, nodule dry weight and number of active nodules in Masaba central and Butula.

	Number of nodules		**Nodule dry weight**		**Number of active nodules**	
	Masaba	**Butula**	**Masaba**	**Butula**	**Masaba**	**Butula**
Control (inoculated)	12^{a}	37^{a}	1.04^{a}	1.93^{a}	12^{a}	24^{a}
Sympal alone	14^{a}	64^{d}	1.51^{ab}	3.97^{c}	13^{a}	49^{c}
Manure alone	19^{ab}	56^{cd}	2.00^{bc}	4.03^{c}	16^{a}	43^{bc}
Manure+MN	21^{ab}	43^{ab}	1.62^{ab}	3.09^{b}	20^{bc}	33^{ab}
Sympal + manure	28^{b}	55^{cd}	2.47^{c}	3.96^{c}	24^{c}	46^{c}
Sympal+MN	30^{b}	50^{bc}	1.39^{ab}	2.74^{b}	22^{c}	36^{b}
F. Probability	0.030	0.005	0.008	<.001	0.002	0.003
LSD	10.1	11.28	0.6031	0.770	4.763	9.92
% C.V.	26.3	12.2	19.2	12.7	14.3	14.0

Different letters within each column shows significant difference to LSD ($P \leq 0.05$).

MN – micro-nutrients.

4.3.2 Effects of the treatments on above ground biomass yield accumulation.

All the treatments had higher above ground biomass compared to the control treatment in both sites. The treatments differed significantly from each other ($P \leq 0.05$) in Masaba central, while there were no significant differences between the treatments in Butula. In Masaba central, application of manure alone had higher above ground biomass and was not significantly different from the application of sympal plus manure, manure plus micro-nutrients and sympal plus micro-nutrients. Application of sympal alone did not differ significantly from the control treatment (Figure 4). In Butula, application of manure alone had the highest above ground biomass followed by the application of manure plus micro-nutrients and was not significantly different from the other treatments (Figure 4).

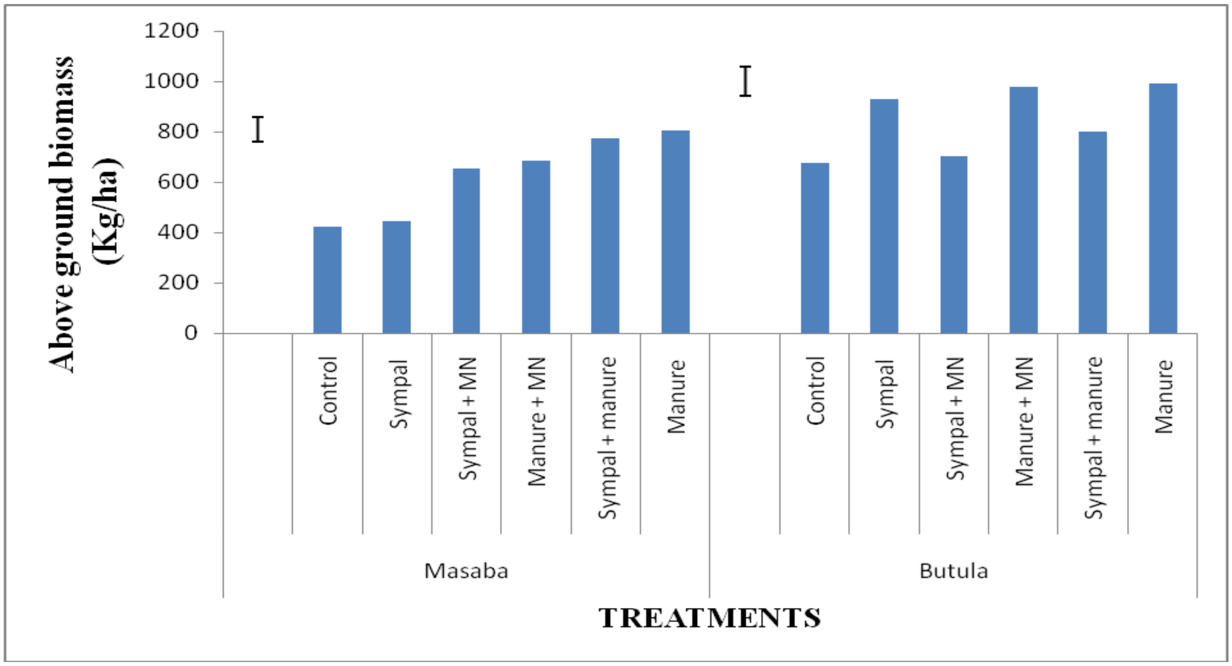

Figure 4: Above ground biomass yields of soybean as affected by the different treatment in Masaba central and Butula; where MN = micro-nutrients. Error bars stand for LSD.

4.3.3 Effects of the treatments on soybean grain yields.

All the treatments increased the grain yields compared to the control treatment in both sites except for the application of sympal alone in Butula. The treatments differed significantly from each other ($P \leq 0.05$) in both sites. Application of micro-nutrients increased grain yields in both sites. In Masaba central, application of sympal plus micro-nutrients had the highest grain yields (0.98 t/ha) followed by manure plus micro-nutrients (0.95 t/ha). They were not significantly different from the application of manure alone and sympal plus manure (Figure 5). In Butula, application of manure plus micro-nutrients had higher grain yields (1.4 t/ha) and was followed by sympal plus micro-nutrients (1.2 t/ha). These treatments were not significantly different from sympal plus manure. Application of manure alone and sympal alone did not differ significantly from the control treatment in both sites (Figure 5).

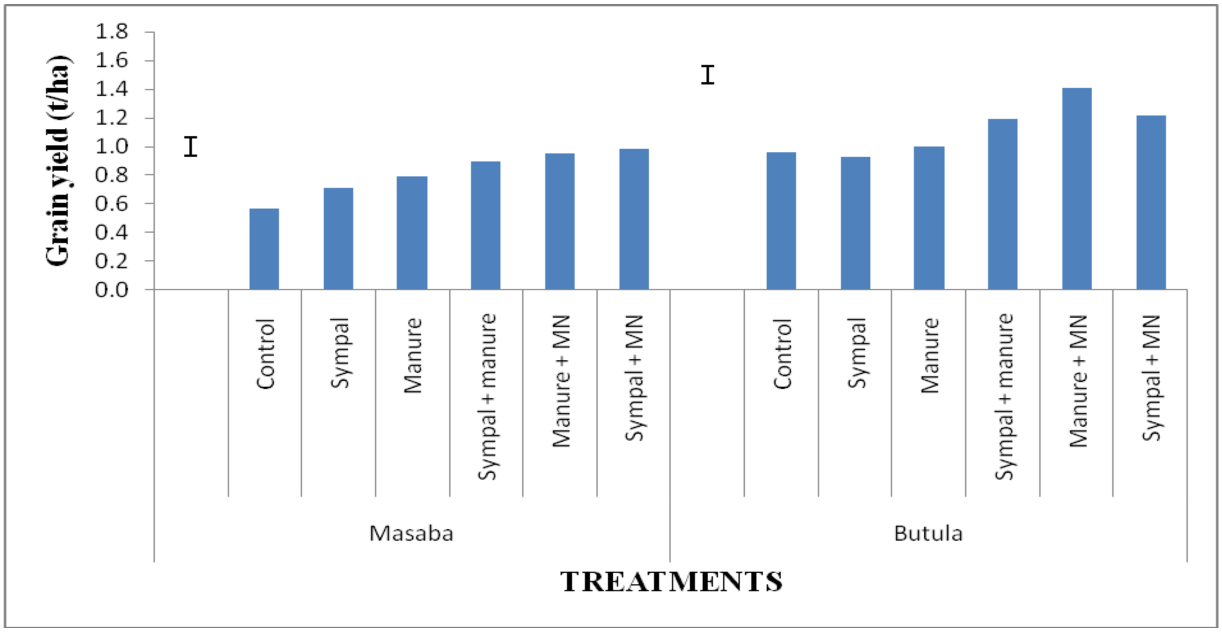

Figure 5: Grain yields of soybean as affected by the different treatments in Masaba central and Butula; where MN = micro-nutrients. Error bars stand for LSD.

4.4 Effects of liming on selected soil chemical properties (pH, available P, and total N).

The treatments differed significantly ($P \leq 0.05$) in their effect on soil pH in both sites. Lime applied treatments differed significantly from the un-limed treatments in both sites. Manure application alone did not differ significantly from the limed treatments in Masaba central. The pH of the soils was raised by 0.60 and 0.85 units in Masaba central and Butula respectively by lime application (Table 14). The treatments also differed significantly ($P \leq 0.001$) in both sites in their effect on soil available P. Lime application significantly increased the soil available P over the control treatments in both sites. It had significantly lower available P than sympal in Masaba central. The effect of the treatments on soil available N was significantly ($P \leq 0.05$) different in Masaba central while there was no significant differences in Butula. Sympal plus lime had the highest level of soil total N (0.15% N) in Masaba central and was not significantly different from application of manure. In Butula, lime application increased soils total N nitrogen over the control treatment and were not significantly different from each other and also from the other treatments (Table 14). Some of the treatments had the same total N values as the initial soil before planting (Tables 6 and 14).

Table 14: Effects of lime on soil pH, available P and total soil N in Masaba central and Butula.

	Masaba Central			Butula		
TREATMENTS	**pH (H_2O)**	**P (mg/kg)**	**%N**	**pH (H_2O)**	**P (mg/kg)**	**%N**
Control	4.70^{a}	9.31^{a}	0.16^{a}	4.95^{a}	7.45^{a}	0.17^{a}
Sympal	4.84^{ab}	14.06^{d}	0.15^{a}	5.35^{b}	11.79^{cd}	0.19^{a}
Sympal+MN	4.88^{ab}	9.31^{a}	0.16^{ab}	5.07^{a}	12.92^{d}	0.18^{a}
Manure	5.08b^{c}	11.92^{bc}	0.18^{cd}	5.11^{a}	9.79^{b}	0.18^{a}
Sympal+MN+lime	5.11b^{c}	11.03^{b}	0.16^{ab}	5.79^{c}	10.75^{bc}	0.19^{a}
Manure+lime	5.29^{c}	13.07^{cd}	0.17^{bc}	5.78^{c}	10.63^{bc}	0.18^{a}
Sympal+lime	5.36^{c}	11.25^{b}	0.18^{d}	5.72^{c}	10.72^{bc}	0.19^{a}
F. probability	0.003	<.001	0.002	<.001	<.001	NS
LSD	0.295	1.387	0.012	0.212	1.335	0.029
% CV	3.3	6.8	4.1	2.2	7.1	8.6

Different letters within each column shows significant difference to LSD ($P \leq 0.05$). MN – micro-nutrients.

4.5 Effects of liming on soybean grain yields.

Application of lime generally increased grain yields compared to un-limed treatments in both sites. In Masaba central, application of sympal plus micro-nutrients plus lime led to higher grain yields (1.27 t/ha). This was not significantly different from the application of sympal plus lime, manure plus lime and sympal plus micro-nutrients (Figure 6). In Butula, application of sympal plus micro-nutrients plus lime also had higher grain yields (1.85 t/ha) and was not significantly different from the application of manure plus lime (Figure 7).

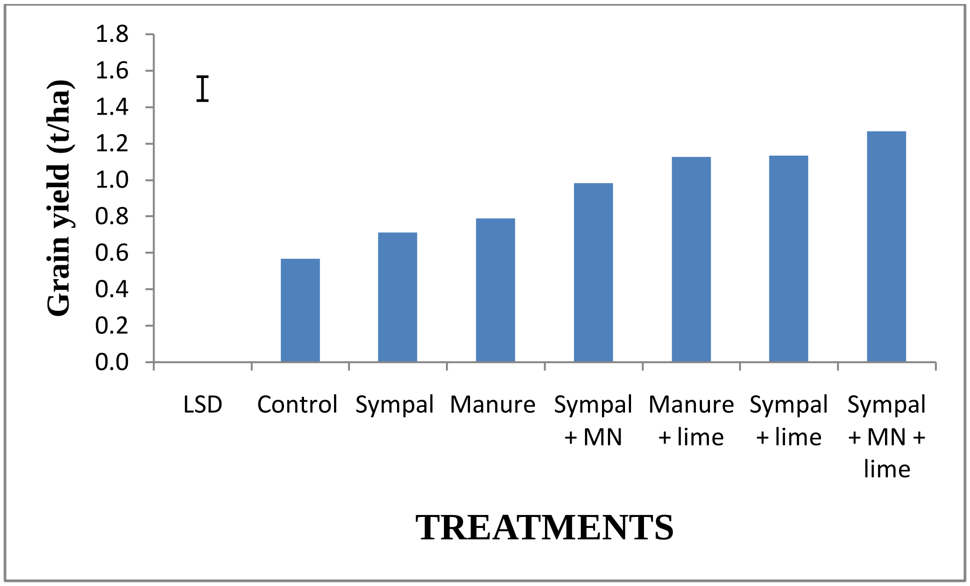

Figure 6: Effect of lime on soybean grain yields in Butere (Masaba central); where MN = micro-nutrients. Error bar stands for LSD.

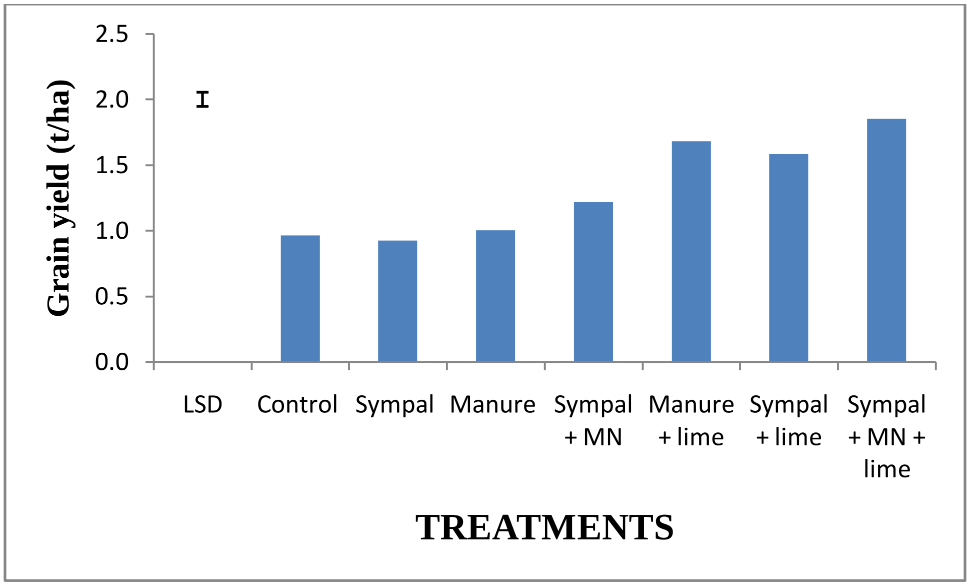

Figure 7: Effect of lime on soybean grain yields in Butula; where MN = micro-nutrients. Error bar stands for LSD.

4.6 Economic analysis.

The treatments with marginal rate of return greater than 50% were considered worth for adoption. At Masaba central, application of manure alone and manure plus micro-nutrients attained economically viable returns while in Butula, application of manure plus micro-nutrients and manure plus lime attained viable returns (Table 15).

Table 15: Gross field benefits, total costs that vary, net field benefits and marginal rate of returns of the treatments used in Masaba central and Butula.

	Treatments	Gross benefits (KES)	TCV (KES)	Net Benefits (KES)	MRR (%)
MASABA	Control	19577	12120	7457	
	Manure alone	39006	18800	20206	190.9
	Manure plus micro-nutrients	48594	20352	28242	517.8
	Sympal	35101	32880	2221	D
	Sympal plus micro-nutrients	47030	34432	12598	D
	Manure plus lime	56024	37200	18824	D
	Sympal plus manure	44298	39560	4738	D
	Sympal plus lime	55707	51280	4427	D
	Sympal + micro-nutrients + lime	62642	52832	9810	D
BUTULA	Control	44402	12160	32242	
	Manure alone	49500	18840	30660	D
	Manure plus micro-nutrients	69795	20392	49403	208.5
	Sympal	45788	33160	12628	D
	Sympal plus micro-nutrients	60143	34712	25431	D
	Manure plus lime	83111	37240	45871	808.54
	Sympal plus manure	59153	39840	19312	D
	Sympal plus lime	78210	51560	26650	D
	Sympal + micro-nutrients + lime	91575	53112	38463	D

D – Dominated treatments

CHAPTER FIVE

DISCUSSION

5.1 Initial soil characterization

Soybean plant grows well at pH levels between 6.0 and 7.0, with the optimum value of 6.3 to 6.5 (Staton, 2012). All the soils used in the experiment were acidic (Kanyanjua *et al.,* 2002) and thus were below the optimal range required for soybean growth and development. The nutrient levels in all the soils ranged from low to moderate levels and thus shows the ability of the soils to respond to inoculation and P fertilizer application in all the sites due to low soil fertility, especially the low nitrogen and phosphorus levels (Mungai and Karibiu, 2009). Sanginga, (2003) reported that there is a clear response to inoculation, P and K fertilizer application in low fertile soils but other nutrient deficiencies appear to be the major constraint.

Based on the low to moderate levels of carbon in the different sites, the soils can be said to be of low to moderate fertility based on carbon and nitrogen levels (Okalebo *et al*., 2002). The low levels of carbon also indicates that there is low amounts of organic matter in the soils, thus application of organic matter will help in improving the soil fertility status of these soils (Otieno *et al.,* 2009). The low levels of the cations in the soils may be attributed to soil acidity and thus may have been replaced by H^+ (Kisinyo *et al*., 2013; Kiplagat *et al*., 2010).

5.2 Identification of the nutrients (macro and micro) that are limiting soybean production in Acrisols and Ferralsols of western Kenya

5.2.1 Visual observations and plant tissue nutrient concentrations

Hydroponic system has been frequently used to study the effects of mineral nutrient deficiencies on plant growth and physiology. This is because it is important in identification of the visual symptoms for diagnostic purposes (Shavrukov *et al.*, 2012). Several deficiency symptoms were noted in the double pot experiment. Interveinal yellowing of the leaves can be attributed to magnesium deficiency (Stevens *et al.*, 2002). This can be related to the low Mg concentration in plant tissues growing in magnesium omitted treatments. These values were below the sufficiency ranges (0.3 to 0.6%) for soybean plant at early growth stages (Sabbe *et al.*, 2011). This indicates that the magnesium levels in the soils were low (as shown by the initial soil analysis). Since the element was not provided by the nutrient solution, there was insufficient amount for plant uptake from the soils. This treatment also accumulated high amounts of other nutrients such as, P, N, K, Ca, B and Zn. The low accumulation of Mg in plant tissues growing in Mg omitted treatments and high accumulation of other elements may suggest that the poor performance of the plants may be due to Mg deficiency. The occurrence of Mg deficiency symptoms especially in plants growing in plus N treatments can be associated with NH_4^+ and Mg^{2+} competition for the ion sites in the root surfaces (Marschner, 1995). This can be seen by the low concentrations of Mg in the plant tissues after the control and minus Mg treatments in soils from Kakamega (Khwisero sub-locations), Butere and Butula.

The plants grown on potassium omitted nutrient solution had older leaves turning yellow with tissue necrosis along the leaf margins which could be attributed to K deficiency (Ruiz Diaz *et al.*, 2011). The concentration of K in the shoot tissues of the plants growing in K omitted treatments were lower in all soils compared to the other treatments. These concentrations were below the sufficiency ranges (1.7 to 2.5%) for soybean plant tissues at early growth stages except in soils from Butula whose concentration was 1.78% (Sabbe *et al.,* 2011). The same plants also had high concentration of other elements such as P, Mg, Ca, Cu, Mn and Zn. This shows that K may be limiting in the soils because of high concentration of other nutrients and thus growth is limited by low K. These findings are in agreement with those of Wietske (2012). The presence of other elements in high concentrations in K omitted treatments can be attributed to K competitive ability especially against Ca and Mg (Moore *et al.,* 2014). This competition occurs because there are a limited number of ion carrier sites on the root plasma membrane and thus the ions of the same ion strength can out compete each other for the sites (Marschner, 2011).

Apart from the dark green leaves and stunted plants grown in minus P treatments, some plants exhibited interveinal reddening which is associated with P deficiency in soybean plants at some instances (Slaton *et al.,* 2011). This can be attributed to lower P concentrations in plant tissues growing in P omitted treatments in all the soils. These concentrations were below the sufficiency ranges (0.3 to 0.6%) required for soybean at early growth stages (Sabbe *et al*., 2011). The levels were above the ranges in Masaba central (0.82%). From the initial soil analysis, soils from all the sites had low levels of soil P and thus there were very minimal amounts available for plant uptake in P omitted treatments. The poor root development in Ca omitted treatments can be associated with roles of calcium in root growth. Calcium is involved in cell growth, both at the plant terminal and the root tips. Absence of Ca leads to browning and dying of the root tips and thus leading to poorly developed root systems. Although all the growing tips are sensitive to Ca deficiency, those of the roots are affected more severely (Roy *et al.*, 2006). The low concentration of other elements such as; P, Mg, K, B, Cu, Mn and Zn in Ca omitted treatments can also be attributed to poor root development and thus influencing the nutrient uptake.

5.2.2 Effect of the nutrient treatments on soybean growth

All the treatments were compared against the complete treatment to determine their extent of limitation; this is because theoretically the complete treatment has the best performance because of the optimal conditions for growth. If the treatment was significantly lower than the complete treatment, it meant that the element was limiting (Janssen, 1974). The poor performance of plants growing in minus K treatments indicated that K is limiting in these soils. It has been reported that relatively large amounts of K are required by high yielding soybean varieties. This is because K is associated with the improvement of nitrogenase activity and thus enhancing BNF and finally the grain yields (Roy *et al.*, 2006; Uchida, 2000). Decrease in yield of soybean and corn were found in absence of K application and thus suggesting the need to apply the fertilizer according to the requirements of the plants (Myint *et al.*, 2009). The poor performance of plants growing in magnesium omitted treatments may be attributed to the important roles played by magnesium in the plants. Magnesium is an important component of chlorophyll which helps in capturing energy from the sun for growth and development; Mg also plays an important role in activation of a number of enzymes important in protein synthesis and P reactions (Olugundudu and Adenkule, 2013). Kanyanjua *et al.*, (2002) reported that Mg deficiencies are widely reported and has resulted in ailments such as grass tetany in ruminant animals feeding on grasses with Mg deficiency. Most field Mg deficiencies are induced by competing cations such as, K^+, NH_4^+, Ca^{2+} and Mn^{2+} (Marschner, 1995). Sanginga, (2003) also reported the occurrences of Mg deficiency symptoms in the field trials. These were more pronounced in cases where K fertilizers were used when there was little Mg available in the soil.

The poor performance of plants growing in P omitted treatments in terms of shoot dry weights in all the soils except in soils from Butere and in terms of plant height in soils from Kakamega (Khwisero and Shikhulu sub-locations) and in Butula, indicates that P is limiting in these soils. This finding agrees with many findings that P is one of the most limiting elements affecting soybean production in soils of western Kenya and this can be attributed to the widespread occurrence of soils with high P fixation capacity (Vanlauwe *et al.,* 2003). Despite the poor root development in Ca omitted treatments as explained earlier, the plants growing in this treatment were not significantly different from the complete treatment in terms of shoot dry weight in all the sites except in Masaba central and plant height in all the sites except in Masaba central and Butula. This can be attributed to the soil Ca status (moderate) especially for the soils from Kakamega (Khwisero and Shikhulu sub-locations) and Butula and also to the insignificant difference between the soil pH of minus calcium and complete treatments at the end of the experimental period.

Omission of S from the nutrient solution was not significantly different from the complete treatment in terms of plant height and shoot dry weights. S deficiency in plants is mainly indicated by leaf chlorosis just like in nitrogen deficient plants. Due to its importance in synthesis of amino acids, S availability is mainly assessed by the analysis of the grains to establish their contents (Marschner, 1995). The poor performance in plants growing in minus micro-nutrients treatments in soils from Masaba central and Butula indicates that the micro-nutrients may be among some of the plant nutrients limiting soybean production. Micro-nutrients are very important in soybean nutrition. For instance, Jabbar and Saud (2012) reported that maximum production in leguminous plants can be obtained through effective nodulation and molybdenum application and this is well expressed in terms of yield and nitrogen concentration in the plant tissues. This can be seen in the low accumulation of nitrogen in minus micro-nutrients treatments.

5.2.3 Effect of lime and nitrogen application on soybean performance and soil pH

The positive influence of lime application on shoot dry weights especially in Masaba central and Butula can be attributed to its ability to raise the soil pH by replacing the H^+ on the cation exchange complex with Ca^{2+}. The H^+ then combines with the hydroxyl ions (OH^-) to form water. Calcium ions in the liming material also replace the aluminium and manganese ions from the exchange sites and thus increasing the cation saturation in the soil solution (Kiplagat, 2013). This is evidenced by the rise in soil pH with lime application to the soils (Table 6 – initial soil analysis and Table 11 – final soil analysis).This might have contributed to the better performance of plants growing on lime added treatments.

Lime addition to the soils however, did not significantly influence nutrient uptake of plant nutrients compared to other treatments as expected. This may be due to the good supply of the nutrients from the nutrient solution in the lower pot that was not influenced by lime application. Manganese concentrations in lime added treatments were lower than all the treatments. Lime reduces manganese concentration through mass action when applied to the soils by raising the soil pH. Andric *et al*., (2012) found out that liming reduced Mn concentrations in the soil, but the concentration in the leaves were sufficient but decreased with subsequent seasons under liming.

Addition of nitrogen to the nutrient solution led to variable responses of soybean in the different soils. The high foliage production in plants growing in nitrogen added treatments in soils from Kakamega (Khwisero and Shikulu sub-locations) and in Butula can be attributed to slightly higher nitrogen levels than those of Butere and Masaba central, though all of them were under moderate levels (0.12 to 0.25). These can also be attributed to the moderate carbon levels in the soils from Butula and Kakamega. It has been found out that nitrogen supply to plants increases leaf area and canopies. In dicots, impact of nitrogen supply in hydroponic systems on leaf growth is due to increased cell growth as observed by other earlier studies (Olugundudu and Adekunle, 2013).

5.2.4 Nutrient Sufficiency quotients (SQ)

The nutrient SQ is an index that can be used to assess the nutrient availability. It tells the ability of a soil to supply plant nutrients and thus can be used for fertilizer recommendations (Janssen, 1990; Foli. 2012). SQs help in indicating those nutrients which are present in insufficient amounts. All the treatments apart from the complete treatment should therefore have a sufficiency quotient of below 100%. This is because the complete treatment is provided with all the nutrients (Janssen, 1974). From the study, those nutrients with SQs near 100 are less limiting, those of the elements more than 100 are not limiting while those which are less and farthest from 100% are more limiting (Foli 2012; Seitz 2013). In soils from Masaba central, Kakamega (Shikhulu Sub-location), Butere and Butula, K and Mg omitted treatments had lower SQ’s and thus limiting in the soils. In soils from Kakamega (Khwisero sub-location), Mg alone had low SQs and thus limiting in the soils while P had low SQs in soils from Butere and thus also limiting. The high SQ's of the nitrogen added treatments can be attributed to the high shoot dry weights obtained due to increased foliage.

5.2.5 Soil differences in terms of Shoot dry weights (g) and plant heights (cm)

Performance of plants in terms of shoot dry weights and plant heights per soil can be directly related to the initial soil status. Soils from Kakamega (Khwisero Sub-location) performed better compared to the other soils (Figure 3) and had high concentration of the different elements compared to the other sites from the initial soil analysis (Table 6). Soils from Butere on the other hand had lower shoot dry weights and plant heights compared to the other sites and had lower concentration of the elements from the initial soil analysis compared to the other soils. The percentage clay content in the soils from Butere was low compared to the other soils and thus fell in the textural class sandy loam. The low nutrient accumulation in this soil can therefore be attributed to low clay content and thus their low capacity to store nutrients. This is because clay has a large surface area of negative charges that attract cations and hold them in the exchange sites thus improving the nutrient retention of a given soil (Brady and Weill, 2004). The good performance of the plants growing in soils from Kakamega can be attributed to the carbon contents in the soil, high for Khwisero and moderate for Shikhulu. This shows that there is an adequate amount of organic matter in the soils. Otieno *et al.,* (2007), found out that manure application to the soil increased nodulation and grain yields of common beans, lima bean, green gram and lablab, thus showing the importance of organic matter for legume production.

5.3 Effects of sympal and farmyard manure as sources of macro-nutrients (P, K, Mg, Ca and S) and micro-nutrients (B and Mo) application on selected soil chemical properties and soybean performance

5.3.1 Effects of the treatments on selected soil chemical properties (pH, available P and total N)

The significantly higher soil pH obtained in Masaba central after manure application can be due to the ability of organic manure to raise the soil pH in acid soils. This is because manure is a source of cations which replaces H^+ in the soil surface (Suryantini, 2014). The high levels of the soil available P after sympal application in both sites can be attributed apart from its P content (23% P_2O_5), to its calcium content (10% CaO). This helped in raising soil pH to moderate levels (Table 12) which may have enhanced the reduction of P sorption. Manure application did not differ significantly from its combination with sympal in influencing soil available P in both sites. This is because organic matter has been found to increase available P content of the soils due to its ability to form complexes with cations such as Al^{3+} and Cu^{2+} that form insoluble compounds with P (Karimi *et al*., 2012). The slight changes in soil available N despite inoculation and fertilizer application and also the lower values than that of the control treatment in Masaba central can be attributed to plant uptake upon its fixation. The high soil N obtained after sympal application in Masaba central can be attributed to the increased P levels upon its application. This may have enhanced N fixation due to its important role of energy transfer. Manure on the other hand had significantly high N content in Masaba central and this could be due to its ability to improve the soil conditions for microbial activity and also to its nitrogen content that is released to the soil upon mineralization (Verde *et al.*, 2013). This can also be due to the improved soil pH and the soil P raised above the critical value upon its application.

5.3.2 Effects of the treatments on soybean nodulation

The increase in nodule number, nodule dry weights and number of active nodules after the application of sympal, manure and their combination in both sites over the control treatment may be due to their ability to supply plant nutrients; P, K, Ca, Mg and S. This accounts for the improvement in nodulation since soybean plant requires an adequate supply of major elements for effective nodulation (Musandu and Ogendo, 2001). The significant influence of micro-nutrients (B and Mo) on nodule number and number of active nodules in Masaba central may be due to their importance in soybean nutrition. Mo is an essential component of nitrogenase enzyme where it is involved in the reduction of atmospheric N_2 to NH_3. Research has shown the ability of B and Mo to improve nodule number because both elements have a stimulatory effect on nodule development and in nitrogen fixation (Bellaoui *et al.*, 2014; Nadia and Kandil, 2013). It is suggested that there is a possible synergistic effect between the two nutrients. This is because their application improved chlorophyll concentration of poinsettias compared to when applied singly (Arreola *et al.*, 2008). Sympal and its combination with manure significantly improved nodule number and number of active nodules in Butula. This can be attributed to the significant increase in available P upon the application of these treatments. P acts as a source of energy when Adenosine Triphosphate is converted to Adenosine Diphosphate during nitrogen fixation.

There was a significant increase in nodule dry weights upon manure application over the control treatment with 92.3 and 108.8% in Masaba central and Butula respectively compared to sympal, 44.2 and 105.7% in Masaba central and Butula respectively. This can be attributed to the nitrogen content in manure which may have improved early nodule growth and activity

(Suryantini, 2014). Further improvement of nodulation when manure was combined with sympal may be due to sympal's ability to improve the P contents of the soil.

5.3.2 Effects of the treatments on soybean biomass accumulation and grain yields

Above ground biomass accumulation and soybean grain yields were also improved over the control treatment upon the application of sympal, manure and their combination in both sites. Application of manure improved the shoot biomass over the control treatment; 89.8 and 46.8% in Masaba central and Butula respectively over sympal application 4.4 and 37.6% in Masaba central and Butula respectively. This can be attributed to its nitrogen content and its effect in raising soil N which is important in vegetative growth in a plant. These results are in agreement with those of Ganeshamurthy and Reddy, (2000).

Increased soybean grain yields in both sites after combination of sympal and manure than when applied singly can be attributed to increased phosphorus availability. Phosphorus helps in seed development. Further increase in yields upon the application of micro-nutrients in combination with manure and sympal may be due to their importance in plant growth. Boron helps in pollen tube development, enhances pollen viability and improves flowering and seed set (Ati and Ali, 2011). Foliar application of boron and molybdenum has a marked influence on yield attributing characters such as no. of pods per plant and 100 seed weight (Konthoujam *et al.*, 2012; Nadia and Kandil, 2013; Saker *et al.*, 2002).

5.4 Effects of liming on selected soil chemical properties (pH, available P, total N) and soybean grain yields

The significant increase in soil pH in both sites upon liming can be attributed to the ability of Ca^{2+} ions contained in it to react with H^+, to form weak acids such as water (Kisinyo *et al.*, 2013) thus raising soil pH. The significant increase in soil available P upon liming over the control treatments in both sites can be due to the release of P from the sorption sites by lime reactions as evidenced by the significant increase in soil pH (Anerator and Akinrinde, 2006; Omenyo, 2013). Lime improvement of soil N content over the control treatment especially in Masaba central can be due to its improvement of soil pH which provides favourable environment for microbial activity thus improving N fixation (Suryantini, 2014).

Lime application generally improved soybean grain yields compared to the un-limed treatments in both sites. This may be due to its effect of improving soil pH and available phosphorus in both sites. This was also found out by Bekere, (2013). It has been found out that lime together with *bradyrhizobium* inoculation and also with organic and inorganic fertilizers in soybean significantly improves seed yield (Anerator and Akinirde, 2006).

5.5 Economic analysis

Sympal, lime and their combination did not achieve the economically viable returns in both sites. This can be attributed to the high costs of these materials. Okalebo *et al.,* (2010), attributed the food insecurity encountered in western Kenya to high fertilizer (especially inorganic fertilizers) prices and the poor economies existing in the region. This therefore limits the rate of adoption of the new technologies aimed at replenishing soil fertility. The little practice of liming soils in Sub-

Saharan Africa and especially in western Kenya can also be attributed to the hauling costs of the liming materials (Okalebo *et al.,* 2009). Manure and its combination with micro-nutrients obtained viable economic returns in both sites while the combination of manure and lime had viable economic returns in Butula. This may be due to the low costs of manure compared to inorganic fertilizers. Micro-nutrients on the other hand are required in small quantities and therefore the cost incurred to acquire them is minimal.

5.6 General discussion

Diagnosis for nutrient limitations in different soils revealed that apart from N and P which are known to be widely limiting in these soils, other nutrients such as Mg, K and micro-nutrients are also limiting. Phosphorus, potassium, magnesium and micro-nutrients were found out to be limiting in soils from Masaba central and Butula. Application of farm yard manure, sympal and their combination which are sources of these nutrients improved the soybean grain yields in the fields where these soils were obtained. Further yield increases were obtained with micro-nutrients (B and Mo) application. Liming also improved soybean grain yields by raising the soil pH to favourable ranges. From these results therefore, it is necessary to improve the soil environment and management practices to improve biological nitrogen fixation by enhancing the potential of the legume rhizobium symbiosis. This will result in final yield increases.

CHAPTER SIX

CONCLUSIONS AND RECOMMENDATIONS

6.1 Conclusions

1. The most limiting nutrients in the soils studied for soybean production are K, Mg and P. K is limiting in all the soils except in Kakamega (Khwisero Sub-location), Mg and P are limiting in all the soils. Micro-nutrients are limiting in soils from Masaba Central and Butula while Ca is a limitation in soils from Masaba central

2. Application of sympal, manure and their combination improved soil pH and available P in Masaba central and Butula, and total soil N in Butula site. This led to increased nodule number and dry weight, number of active nodules, shoot biomass and soybean grain yields over the control treatment in both sites. Micro-nutrient (B and Mo) application in combination with sympal and manure further increased soybean grain yields.

3. Soils from all the sites responded well to liming by raising the soil pH, available P and total soil N. It increased soybean grain yields over the no lime application treatments in both sites (Butula and Masaba central).

4. Application of manure alone had economically viable returns in Masaba central while application of manure plus micro-nutrients had economically viable returns in Masaba central and Butula. Manure plus lime led to economically viable returns in Butula.

6.2 Recommendations

1. To increase soybean production in Acrisols and Ferralsols of Western Kenya, fertilizer formulations containing Mg, K, P and Ca should be used. To further increase soybean yields, micro-nutrients (B and Mo) and lime should be used in combination with the fertilizers. This is as a result of their ability to improve soil pH, total N and available P.
2. Farmyard manure may be used to improve soybean production in Acrisols and Ferralsols of western Kenya because it is economically viable to the farmers.

6.3 Way forward

- There is need for more field trials in the different soil types in western Kenya to assess the effects of macro and micro-nutrients application on soil chemical properties and soybean yields.

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APPENDICES

1. GREEN HOUSE EXPERIMENT

Appendix 1: Field capacity determination for the different soils

Soil	Container plus dry soil – g	Container plus wet soil (After 48 hours) – g	Difference in weight (Field capacity) - mls	Amount added daily (mls)
Masaba	331	413	82	25
Kakamega1	333	433	100	30
Kakamega2	331	429	98	29.4
Butere	331	408	77	23.1
Butula	323	409	86	26

Kakamega1 – Khwisero sub-location, Kakamega2 – Shikhulu sub-location

Appendix 2: Determination of lime requirements for the different soils based on Rutgers, 2013.

Soil	Amount of lime (t/ha)	Amount of lime (g/pot)
Masaba	4500	0.5
Kakamega1	2790	0.31
Kakamega2	2790	0.31
Butere	3375	0.37
Butula	2790	0.31

Kakamega1 – Khwisero sub-location, Kakamega2 – Shikhulu sub-location

Appendix 3: Effects of different treatments on shoot nutrient concentration in soils from the different sites

	%P	**%N**	**%Mg**	**%K**	**%Ca**	**B (ppm)**	**Cu (ppm)**	**Mn(ppm**	**Zn (ppm)**
Control	0.15a	2.51ab	0.34b	0.83a	0.68b	24.90a	3.32ab	428a	49.40cd
Minus P	0.21a	2.78ab	0.36b	2.01b	0.93c	32.46bcd	3.14ab	476a	43.58abc
Minus Ca	0.30ab	2.30ab	0.38b	1.99b	0.39a	26.52a	1.88a	410a	31.56a
Plus nitrogen	0.52bc	4.06c	0.35b	2.60cd	1.04c	30.54abc	5.25cd	436a	40.62abc
Minus S	0.66cd	1.98a	0.38b	2.26bc	1.40d	33.94bcd	3.44abc	430a	34.98ab
Plus lime	0.74cd	2.22ab	0.36b	2.27bc	1.58de	37.32de	3.58abc	156a	35.70ab
Complete	0.79d	2.15ab	0.40b	2.46bc	1.45d	34.92cd	4.33bcd	387a	46.86bc
Minus micro-nutrients	0.83d	2.04ab	0.39b	2.49bcd	1.40d	28.84ab	3.16ab	409a	32.68a
Minus Mg	1.40e	2.95b	0.24a	2.99d	1.56de	50.30f	4.95bcd	459a	59.84de
Minus K	1.42e	2.96b	0.59c	1.18a	1.68e	42.56e	5.52d	545a	64.5e
Grand mean	0.703	2.6	0.3772	2.107	1.211	34.23	3.86	414	44
S.E.D	0.1293	0.468	0.0378	0.252	0.0934	2.836	0.951	172.9	6.24
F.Probability	<.001	0.002	<.001	<.001	<.001	<.001	0.011	NS	<.001
C.V.	29.1	28.5	15.8	18.9	12.2	13.1	39	66.1	22.5

Appendix 4: ANOVA for plant height for the different treatments in different soils

Soils	**Source of variation**	**d.f.**	**s.s.**	**m.s.**	**v.r.**	**F pr.**
Masaba central	Treatments	7	85.90	12.27	3.05	0.019
Kakamega 1	Treatments	7	127.50	18.21	3.92	0.006
Kakamega 2	Treatments	7	130.50	18.64	5.22	0.001
Butere	Treatments	7	152.17	21.74	9.44	<.001
Butula	Treatments	7	100.25	14.32	11.66	<.001

Kakameg1 – Khwisero Sub-location, Kakamega2 – Shikhulu Sub-location

Appendix 5: ANOVA for shoot dry weights for the different treatments in different soils

Soils	Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Masaba central	Treatments	7	0.3568	0.050971	9.12	<.001
Kakamega 1	Treatments	7	0.55897	0.07985	4.8	0.002
Kakamega 2	Treatments	7	0.377738	0.053963	7.8	<.001
Butere	Treatments	7	0.612388	0.087484	13.69	<.001
Butula	Treatments	7	0.591397	0.084485	10.14	<.001

Kakamega1 – Khwisero Sub-location, Kakamega2 – Shikhulu Sub-location

Appendix 6: ANOVA for the effects of lime application on shoot dry weights in the different soils

Soils	Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Masaba central	Treatments	1	0.016501	0.016501	6.13	0.048
Kakamega 1	Treatments	1	0.002113	0.002113	0.66	0.446
Kakamega 2	Treatments	1	0.004513	0.004513	2.77	0.147
Butere	Treatments	1	0.040139	0.040139	14.71	0.009
Butula	Treatments	1	0.0032	0.0032	2.76	0.148

Kakamega1 – Khwisero Sub-location, Kakamega2 – Shikhulu Sub-location

Appendix 7: ANOVA for the effects of nitrogen application to the nutrient solution on shoot dry weights in the different soils

Soils	Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Masaba central	Treatments	1	0.02205	0.02205	67.85	<.001
Kakamega 1	Treatments	1	0.39309	0.39309	14.64	0.009
Kakamega 2	Treatments	1	0.45125	0.45125	22.95	0.003
Butere	Treatments	1	0	0	0	1
Butula	Treatments	1	0.16531	0.16531	11.31	0.015

Kakamega1 – Khwisero Sub-location, Kakamega2 – Shikhulu Sub-location

Appendix 8: ANOVA for relative growth rate based on shoot dry weights for the different treatments and soil combination

Soils	Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Masaba central	Treatments	7	0.022604	0.003229	1.21	0.321
	Time	1	0.023716	0.023716	8.88	0.005
	Treatments*Time	7	0.031206	0.004458	1.67	0.145
Kakamega 1	Treatments	7	0.0236806	0.0033829	3.46	0.006
	Time	1	0.0263378	0.0263378	26.91	<.001
	Treatments*Time	7	0.0088639	0.0012663	1.29	0.279
Kakamega 2	Treatments	7	0.033918	0.004845	2.52	0.032
	Time	1	0.038536	0.038536	20.07	<.001
	Treatments*Time	7	0.013224	0.001889	0.98	0.458
Butere	Treatments	7	0.029125	0.004161	3.99	0.002
	Time	1	0.013382	0.013382	12.84	<.001
	Treatments*Time	7	0.021947	0.003135	3.01	0.013
Butula	Treatments	7	0.03029	0.004327	3.16	0.01
	Time	1	0.018372	0.018372	13.42	<.001
	Treatments*Time	7	0.013123	0.001875	1.37	0.246

Kakamega1 – Khwisero Sub-location, Kakamega2 – Shikhulu Sub-location

Appendix 9: ANOVA showing the soil differences in terms of Shoot dry weights and plant heights

Variate	Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Shoot dry weights	Soil	4	0.65404	0.16351	12.23	<.001
	Treatments	7	1.64594	0.23513	17.59	<.001
	Soil*Treatments	28	0.57634	0.02058	1.54	0.058
Plant heights	Soil	4	126.207	31.552	8.85	<.001
	Treatments	7	356.541	50.934	14.29	<.001
	Soil*Treatments	28	133.239	4.759	1.33	0.145

Appendix 10: ANOVA for the effects of the treatments on soil pH in soils from the different sites

Soils	Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Masaba central	Treatments	9	0.41851	0.0465	5.98	0.005
Kakamega 1	Treatments	9	0.14004	0.01556	1.33	0.33
Kakamega 2	Treatments	9	0.49038	0.05449	1.83	0.181
Butere	Treatments	9	0.68085	0.07565	7.98	0.002
Butula	Treatments	9	0.97398	0.10822	4.55	0.013

Kakamega1 – Khwisero Sub-location, Kakamega2 – Shikhulu Sub-location

2. FIELD EXPERIMENTS

Appendix 1: ANOVA for the effects of the treatments on selected chemical soil properties in Masaba central

Variate	Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Soil pH	Block stratum	2	0.04604	0.02302	1.38	
	Treatments	5	0.27743	0.05549	3.33	0.05
Soil P	Block stratum	2	5.792	2.896	2.83	
	Treatments	5	64.338	12.868	12.57	<.001
Soil N	Block stratum	2	0.00065	0.00032579	5.4	
	Treatments	5	0.00133	0.00026681	4.42	0.022

Appendix 2: ANOVA for the effects of the treatments on selected chemical soil properties in Butula

Variate	Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Soil pH	Treatments	5	0.29976	0.05995	2.94	0.058
Soil P	Block stratum	2	2.6133	1.3067	2.07	
	Treatments	5	70.0989	14.0198	22.21	<.001
Soil N	Block stratum	2	0.00237	0.001184	7.05	
	Treatments	5	0.0003	0.0000599	0.36	0.867

Appendix 3: ANOVA for the effects of the treatments on different parameters in Masaba Central site

Variates	Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Nodule number	Block stratum	2	140.46	70.23	2.33	
	Treatments	5	741.54	148.31	4.92	0.03
Nodule dry weight	Block stratum	2	0.7898	0.3949	3.85	
	Treatments	5	3.6977	0.7395	7.21	0.008
Number of active nodules	Block stratum	2	9.513	4.756	0.74	
	Treatments	5	351.308	70.262	10.98	0.002
Above ground biomass	Block stratum	2	569	284	0.02	
	Treatments	5	398101	79620	5.84	0.015
Grain yield	Block stratum	2	0.19661	0.09831	4.41	
	Treatments	5	0.38148	0.0763	3.42	0.046

Appendix 4: ANOVA for the effects of the treatments on different parameters in Butula site

Variates	Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Nodule number	Block stratum	2	215.41	107.7	2.79	
	Treatments	5	1408.25	281.65	7.31	0.005
Nodule dry weight	Block stratum	2	0.1677	0.0839	0.48	
	Treatments	5	10.9681	2.1936	12.64	<.001
Number of active nodules	Block stratum	2	14.08	7.04	0.24	
	Treatments	5	1245.29	249.06	8.63	0.003
Above ground biomass	Block stratum	2	70755	35377	1.8	
	Treatments	5	391856	78371	3.99	0.035
Grain yield	Block stratum	2	0.13662	0.06831	3.11	
	Treatments	5	0.52724	0.10545	4.79	0.017

Appendix 5: ANOVA for effects of lime on selected soil chemical properties in Masaba central site

Variates	Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Soil pH	Block stratum	2	0.27163	0.13581	4.94	
	Treatments	6	1.05143	0.17524	6.37	0.003
Soil P	Block stratum	2	3.0515	1.5258	2.51	
	Treatments	6	56.9081	9.4847	15.61	<.001
Soil N	Block stratum	2	0.00012	5.9E-05	1.27	
	Treatments	6	0.00207	0.00035	7.5	0.002

Appendix 6: ANOVA for the effects of lime on selected soil chemical properties in Butula

Variates	Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Soil pH	Block stratum	2	0.1679	0.08395	5.9	
	Treatments	6	2.37483	0.3958	27.81	<.001
Soil P	Block stratum	2	2.2063	1.1031	1.97	
	Treatments	6	52.1323	8.6887	15.48	<.001
Soil N	Block stratum	2	0.00316	0.00158	6.45	
	Treatments	6	0.00077	0.00013	0.52	0.782

Appendix 7: ANOVA for the effects of lime on soybean grain yields in Masaba central and Butula

Site	Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Masaba central	Block stratum	2	0.12197	0.06099	2.36	
	Treatments	6	1.18886	0.19814	7.68	0.001
Butula	Block stratum	2	0.13327	0.06664	3.78	
	Treatments	6	2.62208	0.43701	24.78	<.001