EVALUATION OF MAIZE (*Zea mays L*) GENOTYPES FOR NITROGEN USE EFFICIENCY

By

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The University of Zambia

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DECLARATION

I, Brian Mutumwa Gondwe, hereby declare that all the work presented in this dissertation is my own and has not previously been submitted for a degree at this or any other university.

Signature.....

Date.....

APPROVAL

This dissertation of Mr Brian Mutumwa Gondwe is approved, fulfilling part of the requirements for the award of Master of Science degree in Integrated Soil Fertility Management by the University of Zambia.

Examiner's names and signatures Date

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ABSTRACT

Low soil Nitrogen (N) challenges of accessibility and high cost of N fertilisers in Zambia are the major constraints associated with maize production. Increased production can only be attained by the application of N fertilisers and by the use of improved germplasm with the ability to efficiently use applied N. Nitrogen Use Efficiency (NUE) technology has been used and remains a potential means of addressing some of the challenges associated with low available N in agricultural soils and the non-availability of inorganic nitrogen fertilisers to farmers. A study was carried at the National Irrigation Research Station, Nanga to evaluate and screen maize genotypes for NUE. To achieve this, the following were determined, NUE, N partitioning and secondary traits associated with low N tolerance. The experiment had two levels of N at 0 and 100 kg N ha^{-1} and was arranged in a Randomised Complete Block Design (RCBD). Nitrogen utilization efficiency, N uptake efficiency, N grain accumulation, N harvest index, harvest index, N uptake, grain yield and biomass yield varied significantly among genotypes at $p < 0.001$. The ranges for total grain N ha^{-1}, harvest index, N harvest index, and biomass and grain yields were 28.9 - 235 kg ha^{-1}, 0.2 - 0.45, 0.3 - 0.7, 5.7 - 27 tons $ha^{-1}$and 1.4 - 9 tons ha^{-1}, respectively. The NUE Partial Factor Productivity and NUE-AE exhibited significant differences at $p < 0.001$ and values ranged from 13 - 94 and 9.7 - 48.5 kg ha^{-1}, respectively for all genotypes. Anthesis silking interval ranged from 0 to 5 days. Earliest genotype was L 857 and the latest were 151 and 152. The evaluation of NUE showed differences among the 30 genotypes. The results of this study show that among the 30 genotypes evaluated, 6 lines (658, 2035, 2026, 2006, 2091 and L 727) produced NUE-AE values higher than the reference genotype and the recorded world average, of 14.8 and 33 kg grain per kg N, respectively. For each kg of N applied these genotypes produced more grain than the rest of the genotypes. The genotypes that partitioned more of the vegetative N to the grain included 917, 2091, 652, 1061, 650, 658 and 2026. Based on the correlation analysis, genotypes with higher N utilisation efficiency partitioned more N to the grain. Overall, the NUE was determined mainly by the individual genotype and its interaction with the soil and environmental conditions. Secondary traits such as delayed senescence (stay green) N uptake, N uptake efficiency and grain N accumulation contributed positively to NUE. Based on the NUE-AE, the genotypes 658, 2035, 2026, 2006, 2091 and L 727 can be included in the next stage of the breeding programmes that have the objectives of breeding for tolerance to low N.

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ACRONYMS

AGRA	Alliance for Green Revolution in Africa
CSO	Central Statistical Office
CIMMYT	International Maize and wheat Improvement Centre
DAP	Days after planting
FAO	Food and Agriculture Organization of the United Nations
GNACE	Grain nitrogen accumulation efficiency
Gw	Grain yield
HI	Harvest index
IPGRI	International Plant Genetic Resources Institute
IPNI	International Plant Nitrogen Institute
K	Potassium
MOP	Muriate of potash
N	Nitrogen
NHI	Nitrogen harvest index
Ng	Grain Nitrogen
Ns	Nitrogen supply (residual and fertiliser application)
Nt	Total plant Nitrogen
NUE-AE	Nitrogen Use Efficiency Agronomic Efficiency
NUE-PFP	Nitrogen Use Efficiency Partial Factor Productivity
NupE	Nitrogen uptake efficiency
NutE	Nitrogen utilisation efficiency
P	Phosphorus
SSP	Single Superphosphate
TGN	Total grain Nitrogen
ZARI	Zambia Agriculture Research Institute

CHAPTER ONE

INTRODUCTION

Maize (*Zea mays* L.), is one of the most important cereal crops in sub-Saharan Africa (Edmonds *et al.,* 2009). It is also a versatile crop; growing across a range of agro ecological zones (Kamara, 2013). In sub-Saharan Africa, maize is a staple food for an estimated 50% of the population. It is an important source of carbohydrates, protein, iron, vitamin B, and minerals. People consume maize as a starchy base in a wide variety of porridges, pastes, grits, and beer. Green maize (fresh on the cob) is eaten parched, baked, roasted or boiled; playing an important role in filling the hunger gap after the dry season. Every part of the maize plant has economic value: the grain, leaves, stalk, tassel, and cob can all be used to produce a large variety of food and non-food products. The calories contribution from maize consumed according to Banziger and Diallo (2002) on average is about 50 % in Southern Africa. Per capital consumption of maize grain in Zambia is estimated at 140 kg year per year according to Smale and Jayne (2003). In industrialised countries, maize is largely used as livestock feed and as a raw material for industrial products, while in developing countries, it is mainly used for human consumption.

1.1 Origin and Botany

Regarding the origin, many theories have sprung up but all indicate Central America or Mexico as the probable area of origin (Dowswell *et al.,* 1996). This area is considered to be the home of teosinte (*Euchleana Mexican schrad*) a near relative to maize. Maize is a monoecious plant with separate male and female flowers on the same plant. Male flowers are located at the top of the plant in the tassel while the female flowers are approximately half way up the plant. The separate male and female flowers make maize a cross pollinated crop.

1.2 Maize Production Areas

Throughout the tropics and subtropics, resource poor farmers cultivate maize, mostly for subsistence as part of agricultural systems that feature numerous crops and sometimes livestock production. In Zambia, the crop is grown in most areas excluding some extraordinarily wet, dry infertile places where sorghum, millets and

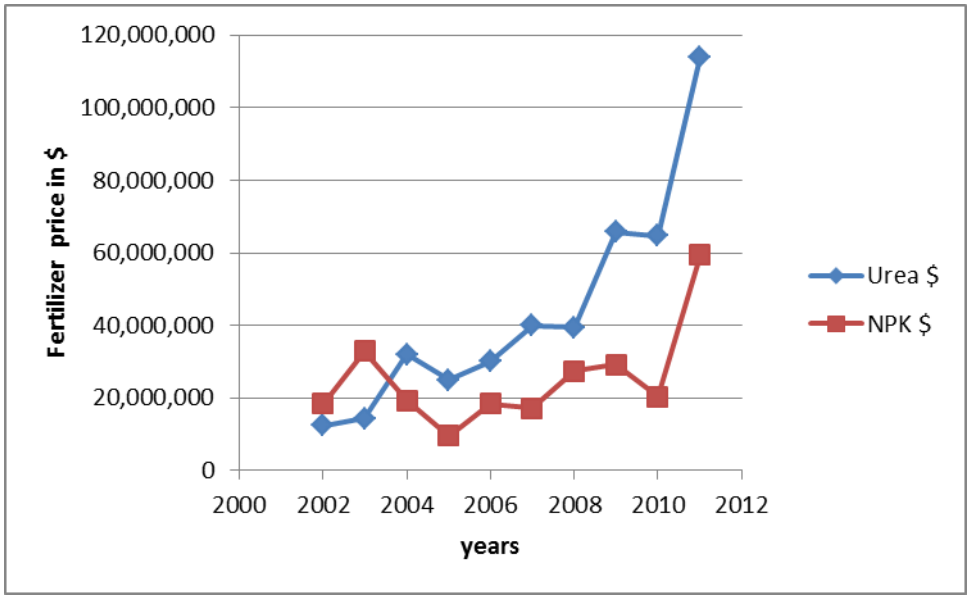
cassava are more adapted. It is well adapted to a varied range of soils, the most appropriate being the heavy textured soils which are associated with high fertility and water holding capacity. Unlike in the developed countries where hybrid maize varieties are normally grown using high inputs and mechanised operations, the production systems in sub-Saharan Africa often lack inputs such as fertiliser, improved seed, irrigation and labour. According to Edmonds *et al.* (2009), an increase in both production and production per unit area has been noted from 1994 to 2006 in sub Saharan Africa. Production per unit area increased from 1.2 to 1.6 tons ha^{-1}, while the average yield of maize in developed countries can reach up to 8.6 tons per hectare. Production per hectare is still very low (1.3 tons per hectare) in Zambia.

1.3 Constraints to maize production

According to Mosier *et al.* (2005) cited in Edmond *et al.* (2009), inadequate use of nitrogenous fertilisers and the deteriorating soil fertility are challenges for maize production in sub-Saharan Africa. While maize is a heavy feeder predominantly in terms of mineral nitrogen (N), Zambian soils are associated with low N and phosphorus (P) levels for plant uptake due to inherently low soil fertility. The full potential of improved varieties of maize is not realised without the application of some form of mineral and/or organic inputs. According to Presterl *et al*. (2002) and Gallais and Coque (2005) cited in Bukan *et al.* (2011), increase of nitrogen (N) fertiliser use in modern cereal production systems has been accompanied by steady increases in average crop yields.

Growing of N-efficient cultivars is an important prerequisite for integrated nutrient management strategies in both low- and high-input agriculture (Mi *et al.*, 2007). Concerns associated with wrong application methods are well documented. Currently, fertiliser use in sub-Saharan Africa averages 9 kilograms per hectare, the lowest of any developing region by far (FAO, 2005) and this has affected the national average yield which is 1.3 tons per hectare. One vital technology that can be used to accomplish high yields per hectare is the use of varieties which can efficiently exploit nitrogen from applied fertilisers and inherent soil N, i.e. by use of varieties that have higher nitrogen use efficiency. Attaining this output, growth is likely to encompass, among many other things, increased use of inorganic fertilizer and improved maize varieties.

In 2002, the Government of Zambia launched the Fertiliser Support Programme (FSP) as a temporary measure to provide subsidised input packages to smallholder farmers to address the high cost of compound fertilisers. From the year 2008, there was an increase in the importation of both urea and compound fertilizers probably due to an increase in the number of beneficiaries under the now Farmer Input Support Programme (FISP), (CSO, 2012).The cost of the fertiliser subsidy is too high compared to extension and research activities. Furthermore, other sectors of the economy like health and education compete for the same limited resources.



Source CSO 2012 report

Figure 1 Fertilizer importation trends per ton from 2002 to 2011 in Zambia

Fertiliser prices have been increasing over the years as shown in figure 1 from 2002 to 2011. Fertiliser prices after 2003, for urea, were generally higher than those of the compound fertilisers.

Focusing on maize, which is the most widely grown crop in Zambia, the yields increased from 2001 to 2010 according to the Food and Agriculture Organisation statistics as shown in table 1 (FAOSTAT, 2012). The results of an economic analysis conducted by the Food Security Research Project, to show the relationship between yield per hectare and fertiliser showed a positive correlation between fertilizer importation (urea) and maize yields. However, cultivated hectares and subsequently

maize yields reduced in 2009 due to the reduction in the number of fertiliser and seed input provision to farmers under the FSP (CSO, 2012).

Table 1 Trend of maize yield from 2001-2010 in Zambia

Year	2001	2002	2003	2004	2005	2006	2007	2008	2009	2010
Yield kg/ha	1378	1409	1725	1923	1859	1899	2334	2244	2069	2587

Source: FAO STAT, 2012

Even though production showed an increase in yield ha^{-1}, nitrogen use efficiency of maize is still very low. NUE- Partial Factor Productivity (PFP) is crop yield per unit of N applied and is indicative of the degree of economic and environmental efficiency in use of nutrient inputs (Doberman, 2005). The NUE-PFP answers the question, "How productive is this cropping system in comparison to its nutrient input?" (IPNI, 2012). Considering that Zambian farmers have been applying the blanket recommendation rate of 200 kg compound D ha^{-1}(20 kg N) and 200 kg urea ha^{-1}(92 kg N), this translates to 112 kg N ha^{-1} applied in the field. The average NUE (PFP kg kg^{-1}) has only been 12 kg kg^{-1} N compared to 33 kg grain kg^{-1}N worldwide (Raun and Johnson, 1999). There is need to bridge this gap by increasing productivity per unit area at reduced costs with appropriate inputs and application rates. Resource poor farmers are already hard hit with high costs of both organic and mineral nitrogen fertilisers currently prevailing in the country. Improving agronomic efficiency will be accompanied by both direct and indirect economic benefits.

1.4 Statement of the problem.

Low soil N fertility, high cost, limited supply of and accessibility to fertilisers are some of the most important limiting factors affecting maize production in Zambia (CSO, 2012). According to Waddington *et al*. (1998) limited use of fertilizers has led to low yields of maize. Additionally, Mineral N applied does not always correspond to an increase in yield; this indicates that not all applied N is utilised by the crop. Some of N applied to maize in most cases has not been accounted for meaning a substantial amount is lost through the system (Hirel, 2001).

Nitrogen use efficiency technology is worth exploring to address the challenges of limited accessibility to N fertilisers among the resource poor farmers. Nitrogen use efficiency (NUE) technology produces crops with yields comparable to conventional varieties which on the contrary require significantly less nitrogen. Even though breeders have sought to breed for N efficient genotypes to improve yields among small-scale farmers, specific information on particular N efficient genotypes is lacking. Additionally, availability of maize seed of nitrogen use efficient varieties is inadequate and the maize breeding programme of Zambia is trying to bulk seed for these genotypes (personal communication).

1.5 Objectives

The overall objective of the study was to screen and characterise maize genotype for nitrogen use efficiency.

Specific objectives were to:

I. Determine the NUE of different maize genotypes;
II. Determine N partitioning in maize genotypes between the biomass and grain;
III. Identify secondary traits associated with low-N tolerance.

1.6 Hypothesis.

It was hypothesised that:

- different maize genotypes have different nitrogen use efficiencies
- partitioning of N is different among maize genotypes with different nitrogen use efficiencies
- secondary traits can be used to predict nitrogen use efficient genotypes during the growing period.

1.7 Significance of study

This study will contribute to the identification of genotypes that are efficient at utilising nitrogen from fertilisers. These genotypes could be used as parental lines in maize breeding programme. Development of varieties capable of utilising nitrogen efficiently could mitigate the high cost of production due to the cost N fertilisers.

1.8 Justification

The use of genotypes capable of efficiently utilising N could contribute to the enhancement of food security in the country. It is vital that high yielding varieties be identified in order to reduce the cost of production associated with high fertiliser prices. Crops efficient in N utilisation will use less N to produce substantial yields per kilogram from N applied and this will reduce N losses through leaching and denitrification and this will address environmental concerns. According to Daberkow *et al*. (2000), improving N use efficiency by 15%-20% can increase food production by 2030 with 20 million tons less fertiliser than is currently being used and based on the current average fertiliser N recovery of 35%. Improved NUE among genotypes can reduce application rates of N fertilisers and subsequently, farmers' net returns from agriculture can increase. Farmers will not only optimise their use of N fertilisers but also maintain sufficient production profit margins with the NUE technology (IPNI, 2012).

The concept of fertiliser use efficiency implies not only the maximum uptake of the applied nutrient by the crop but also the availability of the applied nutrient under variable climatic, soil physical, chemical and biological conditions. Projections indicate that the climate in Zambia will be warmer with reduced rainfall in all Agro-ecological Regions (I, II and III) especially in Region I with rainfall less than 700 mm per year. This is expected to further reduce the productivity of maize in the country. In order to address this problem of low productivity there is need to increase and stabilise maize production in southern Africa by developing locally adapted maize cultivars with considerably improved tolerance to drought, N and P stresses. A study was carried out to screen and evaluate the ability of different genotypes to use applied N efficiently so as to identify those capable of utilising it judicially.

CHAPTER TWO

LITERATURE REVIEW

2.1 Importance of Nitrogen

Nitrogen (N) is the most vital plant nutrient for maize production. Nitrogen is a constituent of the building blocks of almost all plant structures and of proteins enzymes and chlorophyll. Nitrogen occupies a distinctive position as a plant nutrient because relatively high quantities are required compared to the other essential nutrients (Marschner, 1995). It encourages root growth and crop development as well as uptake of the other nutrients. Nitrogen combined with high concentrations of chlorophyll exploits the sunlight as an energy source to carry out essential plant functions including nutrient uptake. Chlorophyll is associated with the production of simple sugars from carbon, hydrogen, and oxygen. These sugars along with their conversion products play a role in stimulating plant growth and development. Therefore, crops, usually respond quickly to N applications. In legumes, high application rates are unfavourable to nitrogenase activity in N_2 fixing systems. (Hofman and Cleemput, 2004).

2.2 Effect of Nitrogen on Crop Yield

According to Presterl *et al.* (2002) cited in Bukan *et al.* (2011), increased use of nitrogen fertilizer in cereal production has been achieved through steady increases in crop yields. Its use may continue to increase substantially as global population and food requirements grow. Because nitrogen uptake, biomass production and grain yield are strongly correlated, the requirement for nitrogen by a maize crop is related to grain yield. The use of inorganic and inorganic nitrogen fertilizer with well managed agronomic practices can increase yields. To assure high yields, farmers usually apply recommended rates of N of about 200 to 400 kg N ha^{-1} yr^{-1} in a wheat-maize cropping system, however, the cost of applying these rates is too high for the resource poor farmers in Zambia (CSO, 2012).

2.3 Types of Nitrogen fertilizers

The manufacture of inorganic nitrogen fertilisers is founded on the artificial fixation of atmospheric N in the form of NH_3. The NH_3 produced is used for manufacturing inorganic fertilisers, containing either NH_4^+, NO_3^-, a combination of both, or the amide form ($-NH_2$). Apart from straight N fertilizers, compound N fertilisers mixed with other primary nutrients, such as phosphorous (P) and/or potassium (K), are widely used (FAO, 2006a). Ammonium (NH_4^+), nitrate (NO_3^-) and urea ($CO(NH_2)_2$) make up the main types of N fertilisers that are very common on the Zambian market. The positively charge, NH_4^+-N is adsorbed by the negatively charged soil colloids (clay and organic matter) and thus preserved from leaching,however,NH_4^+ is transformed to the negatively NO_3^- form by bacteria which is vulnerable to leaching(Wild, 1988).

Nitrate N also can be lost to the atmosphere through denitrification when soils become water saturated (Stevenson, 1982).

2.4 Other sources of inorganic Nitrogen in Soils.

2.4.1 Nitrogen mineralised from organic matter

Mineralisation results in the conversion of organic nitrogen to mineral nitrogen by soil microbes. Mineralisation of soil organic matter is usually of the order of less than 50 kg N ha^{-1} $year^{-1}$ for low organic matter content soils to more than 200 kg N ha^{-1} $year^{-1}$, dependent on climatic conditions, organic matter content and tillage practices. To preserve balanced state conditions, this N release has to be compensated for by inputs of organic N and/or immobilisation (Hofman and Cleemput, 2004).

2.4.2 Biological Nitrogen Fixation

Rhizobium species existing in symbiotic association in root nodules of legumes (e.g. soybean, alfalfa, peas, and beans) can convert atmospheric N_2 gas to NH_3, which is further transformed to amino acids and proteins. In exchange, the legumes make available energy to the *Rhizobium* species which they need to grow and to fix N_2. Photosynthetic cyanobacteria are also N-fixing organisms and are particularly important in rice paddies. The quantity of N fixed differs significantly from crop to

crop, ranging from a few kg to several hundred kg N ha^{-1} $year^{-1}$. The process is reduced once other available sources of N are abundant (Marschner, 1995).

2.4.3 Atmospheric Nitrogen Deposition

Total atmospheric N (NH_4^+ and NO_3^-) deposition by lightening is of the order of 10-40 kg N ha^{-1} $year^{-1}$ in much of Europe and North America. The quantity deposited during the growing season is considered as N being formed by mineralisation of organic matter. Furthermore, this deposition adds to acidification of agricultural soils, with potential influence on biodiversity (Brussaert *et al.*, 2001), and to eutrophication of delicate ecosystems.

2.4.4 Organic Nitrogen Sources

Apart from inorganic fertilizers, the use of organic N through animal manure, sludge or other N-containing secondary products is quite important in countries with intensive cattle, poultry and swine feeding. Organic N sources can be immensely important N fertilisers in countries with developing agriculture, specifically when inorganic fertilizers are not handy or not cheap. Organic manure can be of plant or animal origin or a mixture of both however, most of the organic manure comes from dung and urine from farm animals. It exists as farmyard manure, urine or slurry as well as compost. Plant residues and animal manure nutrient composition are not constant. Plant material (catch or cover crops, legumes) is habitually added freshly cut (green manure) to the soil. Crop residual nutrients available for the next crop range from less than 20 per cent to more than 50 per cent of what is applied (FAO, 2006a). Legumes and manure can release quite high amounts of N in a relatively short time. However, around 50 per cent of the total amount of N in slurry manures exists in the form of NH_4^+, which is volatilised to some extent, depending on the application procedure. Other organic N sources, like farmyard manure and some composts, release their N in the soil in a gradual manner as compared to mineral N fertilizers (FAO, 2006b)

2.4.5 Nitrogen Input by Irrigation Water

Irrigation water can contain NO_3^- originating from sewage or leached from agricultural land. This contribution ought to be taken into account when calculations

are made with regard to fertilization practices. For example, a total irrigation of 100 mm and a concentration of 20 mg NO_3 --N L^{-1} provide an input of 20 kg N ha^{-1} (Hofman and Cleemput, 2004).

2.5 Nitrogen losses

2.5.1 Leaching

Nitrates are soluble in the soil solution. Unlike ammonium, nitrates are not held on soil colloidal particles. Once the soil is completely saturated, nitrates may leach into field drains or sub-surface aquifers as drainage water moves through the soil. Leaching is faster on light sand soils compared to deep clay or silt soils which are less free draining and therefore more retentive of nitrate. The amount of rainfall has a significant effect on the quantities of nitrate leached (Stevenson, 1982; Van Straaten, 2007).

Even though ammonium-N can be intensely fixed to clay particles and is less at risk of leaching than nitrate, under normal situations ammonium-N in the soil is speedily converted to nitrate. In practice, sources of nitrogen comprising ammonium-N will have a comparable risk of leaching as sources containing nitrate once used in excess of the requirement of a crop (Unkovich *et al*., 2007).

2.5.2 Run-off

During and subsequent heavy rainfall, N in solution or in organic form can move over the soil surface through runoff and enter open water courses. The quantity of N lost from soil in this way will differ usually from field to field and season to season depending on the amount and timing and intensity of rainfall and nitrogen applications. The sloping ground, the proximity to surface water and application of slurry present particular risks of nitrogen loss in run-off (Hofman and Cleemput, 2004).

2.5.3 Denitrification

There are two types of denitrification: biological denitrification and chemodenitrification. Biological denitrification refers to biochemical reduction of NO_3^--N to gaseous compounds. During denitrification, NO_3^- and NO_2^- are reduced to

N oxides (NO, N_2O) and molecular N (N_2) by *Thiobacillus denitrificans, Micrococcus denitrificans and Paracoccuss* species micro-organisms. Chemodenitrification encompasses, as its term suggests, the chemical reduction of nitrite ion to N_2O by compounds such as amines present in soil organic matter, and by inorganic ions (Fe^{2+}, Cu^{2+}), predominantly in sub-soils. It is less significant than nitrification or biological denitrification as a source of N_2O from agricultural soils (Anderson and Levine, 1986).

In anaerobic soils, nitrate can be denitrified and lost to the atmosphere as the gases nitrous oxide, a green-house gas, and nitrogen (N_2). Denitrification is a biological process and remains the most significant process in wet and warm soils where there is a source of nitrate after harvest or where there has been a current nitrogen application and there is abundant organic matter for the microbes to feed on. Some nitrous oxide is formed during nitrification of ammonium-N to nitrate-N and some of this also can be lost to the atmosphere (Unkovich *et al.*, 2007).

2.5.4 Ammonia volatilisation

Urea is the leading fertilizer N source used in the world and volatilisation loss of NH_3 from urea is estimated in the range from 30- 50% of the urea-N applied to surfaces of alkaline soils (Tisdale, 1985). Volatilisation of ammonia from N fertilizers is projected at 18% in developing countries, based on N sources used and environmental conditions, while volatilisation losses in industrialized countries is estimated at 7% (Bouwman *et al.*, 2002). When urea-containing fertilizer sources or manure are applied on the soil surface and not incorporated, especially in humid environments, a large portion of the N can be lost to the air as NH_3.

Nitrogen may be lost to the atmosphere as ammonia gas. Significant losses commonly occur from livestock housing, livestock grazing and where organic manures are applied to fields and are not immediately incorporated by cultivation. There can also be significantly larger losses of ammonia when urea is applied compared to losses when other forms of nitrogen fertiliser, such as ammonium nitrate are used.

2.5.5 Environmental impact of N fertilisers

The availability of fertiliser N to plants is to some extent controlled by soil microbial processes. The N cycle in soils is complex and under certain conditions large amounts of plant available N can be lost from the soil to the atmosphere or in drainage water (Townsend *et al*., 2003).

In spite of using nitrogen (N) amendments in agriculture to enhance crop yield, excess application rates that outstrips crop demand leads to surface and groundwater pollution and contribute to shortages of fresh water resources (Scharf *et al.*, 2002). High application rates of inorganic nitrogen fertilisers in order to maximize crop yields combined with the high solubilities of these fertilisers leads to increased concentrations of nitrates in runoff into surface water as well as leaching into groundwater. Ammonium nitrate use is mostly detrimental to the environment where plants absorb ammonium ions preferentially over nitrate ions, excess nitrate ions which are not absorbed dissolve (by rain or irrigation) into runoff or groundwater (Wild, 1988)

The N lost to the atmosphere is in various forms of nitrous oxide gases (collectively referred to as NOx gases), whereas the N entering water bodies via run-off or leaching is in the form of nitrates (Harris, 1988). Nitrogen-containing inorganic and organic fertilizers can cause soil acidification when added. The process of nitrification transforms NH_4^+ to NO_3^-N. Nitrification is a major cause of soil acidification as two hydrogen ions are released to the soil solution for each nitrate ion produced. This is shown as:

$$NH_4^+ + 2O_2 \longrightarrow 2H^+ + NO_3^- + H_2O$$

This may lead to decreases in soil pH to acidic levels which further affects nutrient availability and this may be offset by liming.The use of fertilisers is a key factor in increasing soil productivity in terms of crop production amongst resource poor farmers(African union,2006). This is in line with the Abuja Declaration of 2006 that states: “Fertiliser is crucial for achieving an African Green Revolution in the face of rapid rising population and declining soil fertility (Abuja, 2006). The Abuja Summit Declaration indicates that Zambia uses 8 kg/ha of fertilizer and this indicates that most of the cultivated crop land is not fertilized to enhance yields.

Lack of cash and access to credit facilities make N fertilizers sometimes often beyond the reach of resource poor farmers who constitute the bulk of maize growers in Zambia. While there is a call for a new Green Revolution in Africa to increase production and the productivity at farm level by the small-scale farmers who make over 80% of the farming community in Zambia, fertilizer accessibility by small-scale farmers is a challenge (CSO, 2006). Mineral N fertilisers remain the major contributors as sources of N and hence their management should strike a balance between profitability and reduced pollution of the environment.

2.6 Nitrogen Use Efficiency

Nitrogen use efficiency (NUE) is defined as the ratio of grain yield to the N supply or the product of N utilization efficiency and N uptake efficiency (Moll *et al*., 1982). Nitrogen use efficiency can be expressed in several ways. Mosier *et al*. (2004) described four agronomic indices commonly used to describe nitrogen use efficiency: partial factor productivity (PFP, kg crop *yield* per kg nutrient applied); agronomic efficiency (AE, kg crop *yield increase* per kg nutrient applied); apparent recovery efficiency (RE, kg *nutrient taken up* per kg nutrient applied); and physiological efficiency (PE, kg *yield increase per kg nutrient taken up*). Crop removal efficiency (*removal of nutrient in harvested crop as % of nutrient applied*) is also commonly used to explain nutrient efficiency. Available data and objectives determine which term best describes nutrient use efficiency (Roberts, 2008). Nitrogen use efficiency (NUE) technology produces crop yields equivalent to conventional varieties at low N application rates (Davis, 2013). This tool has been used and still has potential as a means of addressing some of the challenges of low available N in agricultural soils and availability of inorganic nitrogen fertilizers to farmers (Hirel *et al*., 2012). The high cost of fertiliser compels the need for NUE to be maximised as it is vital to increase farmer profits derived from the application of fertiliser at the correct time in the correct amounts (Hirel *et al*., 2012)

2.6.1 Factors affecting Nitrogen use efficiency

2.6.1.1 Soil factors

The production potential of many soils in the world is impeded by the low supply of nutrients due to adverse soil physical and chemical constraints. Soil salinity, acidity, elemental deficiencies, toxicities and low organic matter have been cited as major chemical constraints while physical limits include high bulk density, poor soil structure, surface sealing, crusting and low water holding capacity. Amongst other nutrient dynamics, these aspects to some degree determine mineralisation and immobilisation, fixation by adsorption and precipitation mechanisms, leaching, runoff, and gaseous losses via denitrification and ammonia volatilisation in soils (Baligar *et al.,* 2001). Chemical and physical characteristics of soils also affect availability of nutrients in the soil solution, hence, affect nitrogen use efficiency.

Bulk density affects the pore space distribution. Soil compaction reduces the number of large pores (> 100 μm) and as these are the ones through which roots grow most easily, compaction can have an antagonistic consequence on root growth. Roots will be severely impeded if bulk densities exceed 1.55, 1.65, 1.80 and 1.85 g cm^3 on clay loams, silt loams, fine sandy loam and loamy fine sands, respectively (Gregory, 1988). The size, internal and external morphology of roots due to the adverse soil physical situations will impact the root's capability to explore larger soil volume and decrease nutrient and water accessibility and uptake, leading to low NUE and inferior yields.

Soil temperature and moisture greatly influence nutrient transformation (release), their uptake by roots and their subsequent translocation and utilisation by plants (Archer, 1988). Soil reaction significantly influences the availability of several plant nutrients nitrogen (N), phosphorus (P), potassium (K), calcium (Ca), magnesium (Mg), molybdenum Mo), and zinc (Zn) and phytotoxic levels aluminium (Al),manganese (Mn), iron (Fe), and hydrogen (H) (Baligar *et al.,* 2001; Rowell, 1988). Low pH and deficiencies of N, P, K, Ca, Mg, Mo, and Zn are mainly accountable for reduced growth and lower NUE (Marschner, 1995). Liming is an effective technique to ameliorate soil acidity constraints in the soils.

The soil organic matter (SOM) helps to preserve good aggregation and increase water holding capacity and exchangeable K, Ca, and Mg. It also decreases P fixation,

leaching of nutrients and decreases toxicities of Al and Mn. Increasing levels of SOM contribute to the sustainability of the cropping systems and add to higher NUE (FAO, 2005).

2.6.1.2 Fertiliser factor and Innovative N products

2.6.1.2.1 Nitrification and urease inhibitors

The inhibition of urease and nitrification activity reduces N losses owing to volatilisation and leaching; these are dependent on soil properties and the efficacy of the modified fertilisers. Urease inhibitor, NBPT [n-butylthiophosphoric triamide]), has been reported to considerably lower volatilisation losses on a varied range of soils **(**Singh *et al*., 2010**).**

Sulphur coated urea fertilisers are ordinary soluble fertiliser substances with readily available nutrients which after granulation, prilling or crystallization are given a protective (water-insoluble) coating to regulate water penetration and the rate of dissolution and nutrient release (Trenkel, 1997; Shaviv, 2005).

2.6.1.3 Times of application

Synchronising N availability and uptake with applications is cardinal to NUE as inorganic N is very susceptible to losses. One dose application technique does not support uptake by plants and it results in the waste of resources. Split application of N fertilisers rather than sole application is a recommended management practice that matches availability and uptake (Cassman *et al.*, 2002*;* Wild, 1988).

Application method has always been critical in ensuring fertiliser nutrients are used efficiently. Determining the right placement is as important as determining the right application rate. Various placements are available, but most generally involve surface or sub-surface applications before or after planting. Placement decisions hinge on the crop and soil conditions, which interrelate to influence nutrient uptake and availability (Robert, 2008).

Soil testing remains one of the most powerful tools available for determining the nutrient contributing capacity of the soil, but to be useful for making appropriate fertiliser recommendations good calibration data is also necessary (IPNI, 2012).

2.6.1.4 Tillage

In terms of tillage, certain types of systems have been documented to be associated with improved NUE. Conservation tillage is associated with increased soil organic matter and reduced soil bulk density which further improves physical and chemical characteristics of the soil over a long period of time. Soil organic matter and reduced bulk density also improve soil water and nutrient availability (FAO, 2005). These improved soil physical and chemical characteristics have a bearing on root growth and, hence, nutrient absorption from the soil.

2.6.1.5 Plant factor

Development of improved genotypes adaptable to a wide range of climatic changes has been a major contributor to the overall gain in crop productivity. Improved varieties have a higher harvest index as most biomass is converted into economic yield. Genetic variability has been reported to explain the differences in NUE and the parameters of nutrient uptake (Raun and Johnson, 1999).

Plant health is influenced by diseases, insects and weeds that compete for nutrients and water resources and lower NUE. Infections by diseases and insects also reduce crop yields and consequently NUE. Soil borne pathogens such as actinomycetes, bacteria, fungi, nematodes, and viruses present in the soil around roots lead to pathogenic stress and bring profound changes in the morphology and physiology of roots and shoots that reduces plants ability to absorb and use nutrients effectively (Baligar *et al.,* 2001).

Genetic improvement in tolerance to toxicities of Al, Mn, H, Na, trace elements, and salts; and to deficiencies of nutrients, drought, temperature extremes, aeration and high soil bulk density, will enhance the plants' ability to absorb and utilise nutrients more effectively (Baligar *et al.,* 2001; Fairhurst, 2012).Increasing the capability of leaves to carry on photosynthesis under N stress is an important avenue for improving NUE in agriculture. In some stay-green genotypes of maize, longer maintenance of leaf chlorophyll resulted in a 10–12% increase in grain weight (Spano *et al.,* 2003).

Interactions of N, irrigation, and climate variations have substantial effects on crop yield, N and water use efficiency and N leaching (Onsoy *et al.,* 2005). Provided there are adequate supplies of water and other nutrients, N usually has a significant effect

on crop growth, yield and quality. To lessen the over reliance on fertiliser, high costs of inputs and environmental pollution, it is imperative that plant breeders come up with varieties tolerant to water deficit and N stress (O'Neill *et al.,* 2004).

2.7 Benefits of NUE technology

NUE technology has the potential to decrease the quantity of N fertiliser that is lost by farmers every year due to leaching into the soil and waterways. Resource- poor small- scale farmers may efficiently reduce costs by making use of varieties that are tolerant to low soil N. Higher NUE in economic terms can reduce fertiliser input costs, lessen the rate of nutrient losses and enhance crop yields. NUE technology benefits the environment in that reducing application rates lowers the amount of volatilization in form of N_2O since less N fertilisers will be used as they are sources of Green House Gases (GHGs). Furthermore, N_2O has a very high global warming potential, reducing the amount of N fertilisers applied to the soil will mean less contribution to climate change (Davis, 2013).

2.8 Tools used in improving NUE

Tissue testing and chlorophyll meter methods are used as diagnostic tools to assess N status of growing crops and have proven useful in perfecting in- season N management. Leaf colour charts have been highly successful in guiding split N applications in rice and now maize production in Asia (Witt *et al*., 2005).

Precision farming technologies have introduced, and now commercialised, on-the-go N sensors that can be coupled with variable rate fertiliser applicators to automatically correct crop N deficiencies on a site-specific basis (Roberts, 2008).

Soil testing remains one of the most powerful tools available for determining the nutrient supply capacity of the soil, but to be useful for making appropriate fertiliser recommendations good calibration data is also necessary (IPNI, 2012).

2.9 Methods used to assess nitrogen use efficiency

The evaluation of NUE is cardinal to discriminate plant species, genotypes and cultivars for their ability to absorb and utilise nutrients for maximum yields. The NUE is affected by uptake efficiency and this involves the processes of obtaining N from soil, influx rate into roots, influx kinetics, and radial transport in roots which

hinge on root parameters. Uptake is correspondingly linked to the amounts of a specific nutrient applied or present in the soil, incorporation efficiency and utilisation efficiency which involves root and shoot parameters. According to Baligar *et al.* (2001), nitrogen use efficiency is fundamentally a function of capability of a soil to stock sufficient levels of nutrients and ability of plant to acquire, transport in roots and shoots and to remobilise to other parts of the plant. Further, the environmental aspects such as solar radiation, rainfall, temperature and response to diseases, insects allelophathy and root microbes have a great impact on NUE in plants. In field studies, nitrogen uptake efficiency (NupE), nitrogen utilization efficiency (NutE), nitrogen harvest index (NHI), harvest index (HI), NUE agronomic efficiency (NUE-AE and NUE partial factor productivity (NUE-PFP) are either calculated based on differences in crop yield and total N uptake with above ground biomass between fertilised plots and an unfertilised control ('difference method'), or by using N^{-15} labelled fertilisers to estimate crop and soil recovery of applied N (Dobermann,2005). The Isotope method provides the only direct way of determining the uptake of nutrients from the applied fertilisers and it further separates sources of N which include the soil and fertiliser. Labeling with ^{15}N allows more accurate evaluation of contribution of fertiliser under scrutiny than yield or total N of the crop. However, the costs implications are high for the method (IAEA, 2008). Employing a ^{15}N approach, Ma and Dwyer (1998) discovered that prior genetic improvement for NUE can be explained by greater N uptake and improved dry matter production.

In spite of drought tolerance, insect resistance and high yield receiving much attention, classifying genotypes according to their ability to use N fertilisers either efficiently or not has not been done. Therefore, the focus of this study was to screen and evaluate maize genotypes for NUE in field experiments.

CHAPTER THREE

MATERIALS AND METHODS

3.1 Study site

The experiment was conducted at the National Irrigation Research Station (NIRS), Nanga situated at 15° 45' S and 27° 56' E south of Kafue River. The site is located in agro-ecological region II and the ecological sub zone 9 (Veldkamp and Muchinda, 1984) of Zambia and on a plateau with an elevation of more than 600 m above sea level. The area receives between 600 mm - 1000 mm of rainfall per year. The research site has a well-established low N portion with a good irrigation system and a functional weather station where climatic data was collected on a daily basis. The length of the growing period for the plants is about 120 days with the early planting dates starting in November between the 20th and the 30th. The normal planting time in the region is between 1st and 10th of December with the harvesting starting in the following year between 10th and 20th of March.

3.1.1 Climatic data for Nanga area

The general climatic data indicate that from March to October, evaporation outstrips precipitation, representing a net moisture deficit during this time of the year and that is why irrigation was used during this period of the year to supply moisture.

3.1.2 General soil properties

The National Soil survey team of Zambia (ZARI, 2010) classified the soils in the region of the study area as Alfisols according to USDA soil taxonomy. These are somewhat well drained, deep, yellowish red to strong (5-7.5 YR), slightly hard to hard and friable to slightly firm sandy clayey soil with medium activity clay, medium base saturation, with a loamy top soil and a silt/clay ratio of less than 0.9. In the FAO/UNESCO world Soil Map legend classification, these soils are equivalently known as ferric luvisols or chronic luvisols. According to USDA soil taxonomy; this series is a typic kanhaplustalf, clayey, kaolinitic, isohyperthermic (Soil Survey, 1992).

3.1.3 Soil analysis and characterisation

3.1.3.1 Preparation of soil samples

The method for soil sampling was adapted from Lim and Jackson (1998). Soil samples were taken from different points of the experimental field and, thereafter, composited. Compositing involved the gathering and mixing of a series of individual samples, typically from a series of sampling points across the landscape. Reduction in clump size was required so that both compositing and subsampling for analysis represented uniform material. The samples were dried by spreading them out on trays and air dried for two days.

After drying, the samples were passed through a 2 mm sieve. Thereafter, the samples were analysed for selected elements and pH. Soil texture was determined by the Pippete method and soil pH by soil: calcium chloride extraction ratio of 1:2.5 (McLean, 1982). Total organic carbon (C) was determined by Walkey – Black wet oxidation using a solution of potassium dichromate and concentrated sulphuric acid. Total inorganic N (NH_4^+ and NO_3^-) were determined by steam distillation methods (Keeney and Nelson, 1982).Total N was determined by the micro – Kjeldahl procedure (Bremner and Mulvaney, 1982). Cation exchange capacity and exchangeable bases were determined by ammonium acetate method at pH 7 (Thomas, 1982). Available P determination was done by Bray 1 method (Olsen and Sommer, 1982). Bulk density was determined by core ring method (Blake and Bartge, 1986)

3.2 Experimental design and treatments

The experiment was set up in a randomised complete block design (RCBD) replicated 3 times with N levels at 0 and 100 kg N for each genotype. The 100 kg N plot was subdivided into a yield plot from which all yield parameters were determined which included seed and biomass yields and micro plot were biomass and grain N were determined. The ^{15}N labeled urea was applied as basal dressing in the micro plot and was sprayed in opened furrows, covered and, thereafter, followed by irrigation. The ^{15}N of 2 atom % was applied in micro plots at the rate of 20 kg N ha^{-1} while top dressing with 2 atom % N-15 urea was applied at the rate of 80 kg N ha^{-1}. Nitrogen fertiliser in form of granular urea was applied at 100 kg N ha^{-1}yield plot. Top dressing with 80 kg N ha^{-1} urea was applied at six weeks after planting

using banding method. The control plots received no fertiliser application. In this direct approach, a control or check treatment (without fertiliser N application) was included even though in principle it is not needed because fertiliser N uptake is measured directly using the ^{15}N labelled fertiliser. It was included in order to gather additional information (e.g. comparing difference-method data), for economic and productivity evaluations.

3.3 Planting Materials

The origins, genetic background, breeding emphasis of the maize parents were mainly on nitrogen use efficiency, drought tolerance and ecological adaptation to all agro ecological regions of Zambia. Planting of the 30 inbred lines was carried out during the off season under irrigation on 12th September, 2012. SeedCO and ZARI provided the planting materials for the experiment. Each plot had three-rows, at spacing of 0.75 m between and 0.25 m within the rows. Each station was planted to one seed to give a plant population of approximately 53,333 per hectare.

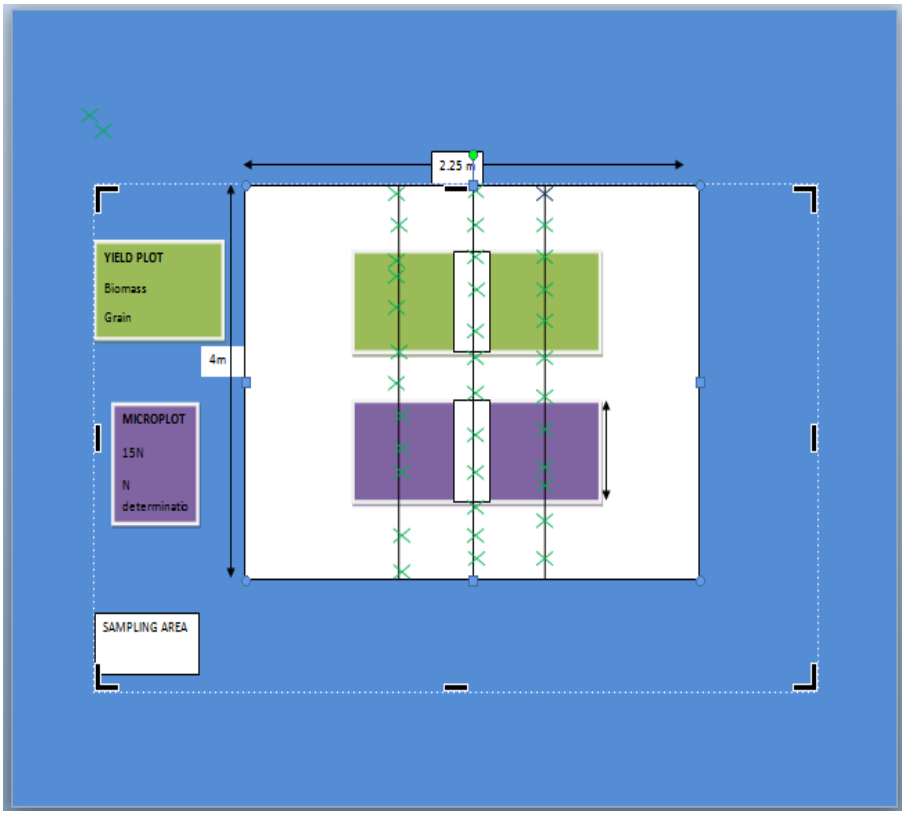


Figure 2: Experimental plot layout

3.4 Cultural practices

Phosphorus and potassium obtained from single super phosphate (SSP) and muriate of potash (MOP) were applied at 80 kg P_20_5 and 60 kg K_20 ha^{-1}. All the SSP and MOP fertilizers were applied once at planting using the banding method with 20 kg N ha^{-1}. Weed control was done mechanically using hand hoes. The stem borers (*Chilo partellus*) were controlled using phorate and Malathion spray every fortnight. Surrounding areas were also weeded to minimise encroachment by insects and rodents.

3.5 Data collection

Data were collected from the middle row in each plot. Agronomic parameters recorded were establishment plant count (EPC), plant and ear heights were measured as the distance from the ground level to the flag leaf and the node bearing the uppermost ear, respectively using a measuring tape. Days to 50% tasselling (TS) and silking (SK) were taken as the date when 50% of the plants in a plot had tasselled and extruded silk, respectively. Anthesis Silking Interval (ASI) which is the difference between Silking date and Anthesis date was also computed. Leaf width and length were also measured.

3.6 Plant tissue preparation and element determination.

Sample treatment was adapted from Campbell and Plank (1998). At harvest, plant materials were cleaned and freed from extraneous substances including soil and dust particles, and foliar spray residues that could influence analytical results and placed in paper bags. Fresh plant tissue samples were examined to determine the physical condition and extent of contamination. Decontamination procedures involved washing and rinsing plant materials in distilled water. After decontamination, samples were dried immediately to stabilise the tissue and to stop enzymatic reactions in a Forced-air oven. Removal of combined water also facilitated particle size reduction, homogenisation, and weighing. The drying was done at 80^oC for 48 hours. Drying temperatures above 80^oC were avoided in order to prevent thermal decomposition and reduction in dry weight.

3.6.1 Particle-size reduction

Plant tissue samples were reduced to 0.5 to 1.0 mm particle size to ensure homogeneity and to facilitate organic matter destruction. The harvested samples were first passed through a shredder to reduce their sizes so as to facilitate their passing through the mill. The samples were ground to pass through a 1.0 mm screen (20 mesh) using the Wiley Mill. Each sample was thoroughly mixed after grinding and a 5 to 8 g aliquot withdrawn for analyses and storage. To avoid contaminating samples during grinding, the grinding apparatus was cleaned after grinding each sample with a brush.

3.6.2 Moisture content determination

The samples at harvest were first weighed to obtain initial weight, followed by drying to a constant weight in the oven at 80^{o}C. The difference between the two weights was recorded as moisture content for stover and expressed as a percentage. A grain moisture sample was drawn by removing several rows of corn kernels the full length of randomly selected ears from each row sampled and thoroughly mixed. The grain was placed in moisture proof container to avoid moisture loss. Moisture content of grain was determined using the Dickey John moisture meter.

3.6.3 Organic matter destruction

Dry ashing was done according to Miller (1998). Dry ashing was conducted in a muffle furnace at a temperature of 500^{o}C for 8 hours. A sample weighing 1.0 g previously dried, ground (0.5 to 1.0 mm) at 80^{o}C and thoroughly homogenised was placed into a high-form 30-mL porcelain crucible.

3.6.4 Determination of total nitrogen in plant samples and grain

Total N in plant samples and grain was determined by the Kjeldahl method (Horneck and Miller, 1998).

3.7 Other parameters determined

Parameters measured were the components of yield and these include, total biomass, grain weight, dry matter yield, moisture content and grain yield. The other

parameters were computed after generation of the data above. Grain protein yield was considered as the product of grain yield and grain protein content and was correlated to nitrogen use efficiency for all the genotypes in the trial.

3.7.1 Parameters computed after plant analysis

3.7.1.1 Nitrogen harvest index (NHI)

An additional important parameter is the N harvest index (NHI). NHI is the ratio of N present in grain to total plant N content (Ng/Nt), analogous to the harvest index (HI). It is a measure of N translocation efficiency. Although not directly related to grain weight, it has significance for maximising grain protein content for a given amount of plant N as it relates to the amount of N in the grain.

3.7.1.2 Nitrogen utilization efficiency (NutE)

N utilisation efficiency (NutE) measures the response of grain yield to the total N in the plant, where total N in plants is based on above ground plant N (Dawson *et al.*, 2008).

3.7.1.3 Grain N accumulation efficiency (Ng/Ns)

Grain N accumulation efficiency (GNACE), which is the amount of N in the grain divided by the N supply Ng/Ns. This measure indicated the overall efficiency with which plants extract N from the soil and accumulate it in the grain at harvest time.

3.7.1.4 Harvest Index

This was measured as a ratio of the economical yield to the biological yield of the plant.

3.7.1.5 Nitrogen uptake efficiency (Nt/Ns)

N uptake efficiency was defined as total above ground plant N at harvest divided by the total N supply (Moll *et al.*, 1982). Uptake efficiency is a measure of how much N the plant absorbs in proportion to the N supply.

3.7.1.6 Nitrogen use efficiency Partial factor Productivity (NUE- PFP)

This is cardinal for farmers because it integrates the use efficiency of both indigenous and applied N resources according to Doberman (2005).

$PFPN = Y_N/F_N$

F_N = amount of (fertiliser) N applied (kg ha^{-1})

Y_N = crop yield with applied N (kg ha^{-1})

3.7.1.7 Nitrogen use efficiency Agronomic Efficiency (NUE- AE)

Agronomic efficiency of applied N (kg yield increase per kg N applied)

$AE_N = (Y_N - Y_0)/F_N$

F_N = amount of (fertiliser) N applied (kg ha^{-1})

Y_N = crop yield with applied N (kg ha^{-1})

Y_0= crop yield ha^{-1} in control plot without N application.

AE_N is the product of the efficiency of N recovery from applied N and the efficiency with which the plant uses each additional unit of N acquired. Agronomic NUE indices only offer precise calculation of NUE for systems that exist at a somewhat steady-state with regard to soil organic N content and where dissimilarities in root systems between unfertilised and fertilised crops are relatively small. This parameter is associated with a range of 10–30 kg grain kg^{-1} N and values greater than 30 kg kg^{-1} N designate a well-managed systems or at low levels of N use or low soil N supply.

3.8 Fifty Percent 50% tasselling and 50% silking

To determine 50 % tasselling**,** the number of days from sowing until 50% of the plants had extruded anthers (anthesis date, AD) were counted. In the same manner, 50 % silking was determined by counting the number of days from sowing until 50% of the plants showed silks (silking date, SD).

3.8.1 Anthesis Silking Interval

Calculating anthesis silking interval was done by subtracting the anthesis date at 50 % from the silking date at 50 % (SD – AD).

3.9 Data analysis

Data collected and computed were analysed using GenStat version 14 to determine differences among genotypes with respect to various parameters and NTSys- pc software version 2.2 was used to cluster the genotypes and execute the Principal Component Analysis based on NUE parameters determined in the experiment. Means of the genotypes that exhibited significant differences were separated using the standard error difference.

CHAPTER FOUR

RESULTS AND DISCUSSION

4.1 Soil chemical parameters of study site

Exchangeable Calcium (Ca), Magnesium (Mg) and potassium (K) were above the critical values as shown in Table 2. Therefore, deficiencies were not expected in this soil. The levels of organic C were below the critical level and this meant that the soil has poor nutrient retention ability, poor inherent N supply and poor moisture availability and workability. The level of total N was below the critical level and this meant that this soil had a poor indigenous N supply and that deficiencies were likely and N input was required in form of fertilisers. The level of P was adequate and above critical levels meaning deficient symptoms would be very unlikely during the growing period. However, P application was done so as to maintain levels for the current crop.

The soil had a pH of 6.34 which is moderately acidic and was within the recommended range for crop production. At this pH level Aluminium (Al), manganese (Mn) and iron (Fe) toxicities are expected to be minimal. Availability of micronutrients and P could be greatest in this pH range. The pH of this soil indicated that soil microorganism responsible for N mineralisation would be in abundance and active as they function best at soil pH range 5.5 – 6.5 (Fairhurst, 2012). The site was suitable for maize production.

Table 2. Selected chemical and physical characteristics of soils at experimental site

Soil pH	Ca^{2+} (cmol/kg)	Mg^{2+} (cmol/kg)	Na^{+} (cmol/kg)	K^{+} (cmol/kg)	N (%)	Org C (%)	P (mg/kg)	CEC (cmol/kg)	Zn (ppm)	NH_4-N (ppm)	NO_3-N (ppm)	Bulk D g/cm^3
6.34	9.98	6.65	0.01	1.18	0.08	0.85	14.46	7.2	BDL	2	7.3	1.46
Critical value	0.1	0.1	0.1	0.1	0.1	2	10	14				

BDL= below detectable level

4.2 Soil profile description for Nanga

The soils of this mapping unit occur in the middle upper slopes of the survey area. They are deep, well drained, dark reddish brown to reddish brown, sandy clay top soil with clayey subsoil. The color variations mainly differ with the depth. The slope is generally gentle to flat. These soils are compact when dry and firm when moist. They are sticky and very plastic below 70 cm. They are moderately porous, fine and medium roots penetrate to at least 200 cm where profiles are very deep. Pitholitic coarse nodules and concretion of black iron manganese throughout the deeper layers from the depth of 150 cm in the profile. This profile lacked mottles (Appendix xix).

Acidity in the profile sampled is medium with pH values ranging between 6.37 and 7.00 in the subsoil a slight increase can be seen from 6.37 to 7.00, or acidity conditions decreasing from medium to neutral (Table 3).

Available Phosphorous is critically deficient in the subsoil and probably high in the topsoil. This can be attributed to decaying organic matter; leaves, grasses and other plant debris gathered. The soils in this mapping unit (m.u Nka-e) have a medium base saturation. These soils are rated as inherently of good arable agriculture potential on account of high fertility status. In terms of classification, to the family level (International Classification) the soils consists of Typic Kanhaplustalf, fine, kaolinitic, iso-hyperthermic (USDA, 1992). The main physical properties are sandy clay, sandy clay loam and clay textures, with a moderate to strong structure. Soil water permeability is medium, with a well-drained condition. Plant root penetration is moderate to good. Main chemical characteristics are dominated by a medium acid soil reaction state, combined with high ability to hold plant nutrients and moderate high soil fertility status. Both the physical and chemical properties are considered as attributes from the geological contribution of the limestone dolomite parent materials prevalent in the area. The profile has six (6) horizons

Table 3. Chemical characteristics of the soil profile

			Exchangeable bases (cmol/kg)							
Horizon (layer)	Depth (cm)	pH $CaCl_2$	Ca	Mg	Na	K	N	Oc	Av. P-1 mg Kg	CEC (cmol/kg)
Ap	0-10	6.37	5.68	7.20	0.01	1.33	0.16	1.12	7.21	9.20
BA	10-27	6.49	5.91	7.28	0.02	1.21	0.15	0.67	2.45	10.53
Bt	27-60	7.00	5.37	7.94	0.02	0.61				11.73
Btz_1	60-83	6.71	5.22	8.07	0.03	0.46				12.27
Btz_2	83-120	6.78	5.19	8.02	0.02	0.38				12.40
BC	120-190	6.69	5.91	8.39	0.03	0.44				11.47
Critical value										

Source Nanga soil survey report 2010

4.3 Genotype characteristics

The genotypes comprised early to medium maturing white cultivars with maturity period of 90 to 100 days. The plants were grouped into two categories namely maize and sorghum growth type. This classification was based on the pigmentation of the leaves, the sorghum type being plants with very shiny light green while the maize type plants had the normal green exhibited by most maize plants. Plant leaf arrangements of the inbred lines were grouped into three categories namely: normal, vertical and very vertical. The vertical plant leaf types were very susceptible to stem borers while the normal ones (IPGRI, 1991) were very resistant to stem borer attack

4.3.1 Diseases

Maize streak virus, Grey leaf spot and Rust were observed on plants in the experimental plots. Plant diseases and pests retard normal plant growth and effective absorption of plant nutrients from the soil.

4.3.2 Days to silking 50%

Significant differences were observed among genotypes at $p < 0.05$ (Appendix I).The number of days to 50% silking ranged from 61 to 80. The genotypes were grouped according to days to 50% silking and ranged from 63 to 80 days as shown in Table 4. The average number of days to 50 % silking for all the genotypes was 68 days. The first group silked earlier (between 63 to 69 days) and it comprised the genotypes L353, L375, L428, L512, L857, L652,L3080, L727, L1061, L2091, L2006, L2035, L2026, L654, L665 and L666. The second group silked later (after 70 days) and comprised the genotypes L1212 , L10 , L151, L152, L60, L645, L658, L666, L695, L7, L713 and L917. Among all the genotypes, the earliest and the latest were L857 and L 152, respectively.

4.3.3 Days to 50% tasselling (DAP)

Significant differences were observed among genotypes at $p < 0.05$ (Appendix II). The days to 50% tasseling for genotypes range was from 61 to 80 days (Table 4). The first group tasselled between 63 to 68 days and comprised the genotypes L353, L375, L428, L512, L857, L652, L654, L665, L666, 2026, 2006, 2091, 1061, 727, 2035 and 3080. The second group which tasselled after 70 days comprised the genotypes L1212, L10, L151, L152, L60, L645, L658, L666, L695, L7, L713 and L917. Among all the genotypes, the earliest and latest to tassel were L857, and L152 and L151, respectively.

4 .3.4 Tassel size

Significant differences were observed ($p < 0.001$) (Appendix VI) for tassel size. The tassel size according to the scale in the IPGRI maize descriptor is 1 to 5 as shown (Table 4). Genotypes 2026, 151, 152 and 713 had the smallest tassels while 375, 654, 658, 353 and 666 had the largest tassel. Genotypes 10, 60, 645, 649, 650, 652 and 665 had medium tassel size of 2.5. Lines 1212, 640, and 512 had tassel size of slightly above 1. The small tassel size is correlated with high tolerance to low soil N.

Table 4. Tassel size, 50% tasseling and silking

Genotype	Tassel 50%	Silk 50%	Tassel size
L857	61a	61.33a	1.333ab
L2026	63ab	63.33ab	1a
L353	63.33ab	63.33ab	3d
L652	63.33ab	65ab	2abcd
L1061	64.33abc	65ab	1.333ab
L2091	65abc	64ab	1.667abc
L2006	65.67abc	64ab	1.333ab
L3080	65.67abc	66.33ab	2abcd
L727	65.67abc	65ab	2abcd
L375	66abc	66ab	2.667cd
L645	66abc	69.67abc	2.333bcd
L428	66.67abc	66.33ab	1.667abc
L512	66.67abc	66.33ab	1.333ab
L650	66.67abc	68.33ab	2abcd
L649	67abcd	68.67ab	2.333bcd
L2035	68abcd	66.33ab	1.667abc
L654	68.33abcd	69ab	2.667cd
L10	68.67abcd	70abc	2abcd
L665	68.67abcd	69ab	2.333bcd
L7	68.67abcd	70.67abc	2.333bcd
L666	70abcd	71.33abc	3d
L713	70abcd	70abc	1a
L60	70.33abcd	70abc	2.333bcd
L658	70.67abcde	70.33abc	2.667cd
L640	72bcde	66.67ab	1.333ab
L1212	72.33bcde	72abc	1.333ab
L695	72.33bcde	72abc	1.667abc
L917	73.67cde	74bc	2abcd
L151	76.39de	79.45c	1a
L152	79.67e	79.67c	1a
p	< 0.007	< 0.0016	<0.001
sed	3.9	3.9	0.48
lsd	7.8	8.7	0.96
cv	7.1	7.8	31.2
mean	68	68	1.9

Values with same letters are not significantly different

4.3.5 Anthesis -Silking Interval (days after planting DAP)

Based on the Days to 50% silking and tasseling discussed above, the Anthesis-Silking Interval ranged from- 5 to 4 days among the genotypes (Table 5). In some instances, the number of days to 50 % silking and 50 % tasselling were the same. Genotypes 1212, 152, 2026, 353, 375, 428, 512, 60, 658, 665, 713, 857 and 917 had the same number of days to 50 % silking and 50 % tasselling hence zero anthesis-silking intervals (Table 5).

In this study, a number of genotypes exhibited a scenario were silking was earlier than tasselling. Delay in anthesis was a less common feature and this in turn increased the anthesis silking interval. The ASI in this case ranged from -4 to 5 days. D'Andrea *et al*. (2006) working with inbreds at 0 and 400 kg N ha^{-1}recorded ASI of 7- 10 days contrary to this study. Shortened anthesis-silking interval, reduced barrenness and delayed leaf senescence, measured under low N have been shown to be useful indicators for low N tolerance, when included with grain yield in a cultivar improvement program (Lafitte and Banziger, 1997).

Table 5. Anthesis – Silking interval of genotypes

Genotype	ASI
L640	-5 a
L2006	-2 b
L2035	-2 b
L2091	-1 bc
L727	-1 bc
L1212	0 bc
L428	0 bc
L512	0 bc
L60	0bc
L658	0bc
L695	0bc
L152	0bc
L353	0bc
L375	0bc
L713	0bc
L2026	0bc
L665	0bc
L857	0bc
L917	0bc
L1061	1bcd
L3080	1bcd
L654	1bcd
L666	1bcd
L10	1bcd
L649	2cd
L650	2cd
L652	2cd
L151	2cd
L7	2cd
L645	4d
p	<0.001
sed	1.3
lsd	2.6
cv	753
mean	0.21

values with same letters are not significantly different

4.3.6 Plant height

The genotypes exhibited significant differences in plant height at $p < 0.001$ (Appendix III).The plant height at the flag leaf stage (72 DAP) ranged from 91.7 cm to 210 cm. The average height of all the genotypes was 154 cm. The tallest and shortest genotypes had the highest and lowest biomass yields, respectively. Lines 640 and 727 were the shortest and tallest, respectively as shown in Table 6.

4.3.7 Leaf Length

The leaf lengths at the Flag Leaf stage ranged from 57 to 95 cm (Table 6) among all genotypes and were statistically different at $p < 0.001$(appendix IV). The leaf size had a bearing on the amount of light trapped by each genotype and on the final canopy cover. Lines 2091 and 353 had the longest and shortest leaves, respectively. The leaf length ranged from 56.7 cm to 95.3 cm.

4.3.8 Leaf width

Significant differences were observed (Appendix V) at $p < 0.001$.The leaf width ranged from 6.3 cm to 12.1 cm (Table 6). This parameter was highly correlated to the biomass yield of the genotypes of $r = 0.6$. The genotypes with wide leaves had a higher corresponding biomass yield (Tables 8).

Table 6. Plant height, leaf length and leaf width of genotypes

Genotype	plant height	Leaf length	Leaf width
L640	91.7a	60.96ab	6.296a
L857	91.7a	63.59abc	7.185abc
L151	96.1a	63.63abc	7.37abc
L353	110ab	56.7a	8.407bcdefg
L152	124.4bc	73.04bcde	7.556abcd
L375	130bcd	74.78cdef	8bcde
L428	136.1bcd	77.78defg	9.444efghi
L1212	136.7bcde	73.63cdef	9.556fghi
L917	136.7bcdef	72.56bcd	8.148bcdef
L713	139.4cdefg	80.04defg	9.667ghi
L695	140.6cdefg	73.07bcde	7.185abc
L645	142.8cdefgh	69.85bcd	7.593abcd
L665	150.6cdefghi	81.85defgh	7.259abc
L666	151.7cdefghi	73.48cdef	6.926ab
L60	152.8cdefghi	86.26fghi	8.963defgh
L658	154.4defghi	76.81defg	7.148abc
L649	155defghi	76.81defg	7.741abcd
L654	157.2defghij	79.89defg	7.815abcd
L512	158.9defghij	78.93defg	8bcde
L650	165.6eghij	79.04defg	7.185abc
L652	169.4hijk	82.15defgh	8.667cdefg
L7	171.7ijkl	79.41defg	9.333efghi
L2006	175.6ijklm	94.04hi	9.815ghi
L10	183.9jklmn	71.3bcd	9.741ghi
L2091	195klmn	95.3i	10.852ij
L3080	195.6klmn	88.96ghi	12.111j
L2026	197.8lmn	85.85efghi	10.407hi
L2035	200.6mn	89.19ghi	10.407hi
L1061	201.1mn	88.07ghi	10.667i
L727	210n	93.04hi	10.333hi
p	<0.001	<0.001	<0.001
sed	12.1	5.3	0.64
lsd	24.2	10.6	1.29
cv	9.6	8.3	9.1
mean	154.1	78	8.7

Values with same letters are not significantly different

In summary, lines 650, 652, 7, 2006, 10, 2091, 3080, 2026, 2035, 1061 and 727 were taller, had longer leaf length and broader leaves compared to others. The traits mentioned translated to higher biomass yields in genotypes lines 2006, 2091, 3080, 2026, 2035, 1061 and 727.

4.3.9 Moisture content of grain

The moisture content of grain ranged from 9.1% to 14.6% with a mean of 12.08% (data not shown). Genotypes with larger kennels had considerably higher moisture content than those with smaller kennels.

4.4 Nitrogen Use Efficiency parameters

Genotype 151 was used as a check genotype for the parameters associated with NUE. This genotype has been associated with both low N and drought tolerance in the maize breeding programme (Chanda personal communication, 2012). The NUE parameters investigated in this study included nitrogen use efficiency, Grain Nitrogen Yield per Hectare, Harvest Index, Nitrogen Harvest Index, Biomass Yield, Grain Yield, Nitrogen Utilization Efficiency, Grain Nitrogen Accumulation Efficiency, N uptake efficiency and Nitrogen Uptake. Each of these components is discussed below.

4.4.1 Nitrogen use efficiency

This component of NUE is composed of two parameters, the Agronomic efficiency (NUE-AE) and the partial factor productivity (NUE-PFP). Agronomic efficiency (NUE-AE) is the *relative yield increase per unit of N applied* and partial factor productivity (NUE-PFP) is *crop yield per unit of N applied*. They are indicative of the degree of economic and environmental efficiency in use of nutrient inputs (Doberman, 2005). The NUE-PFP answers the question, "How productive is this cropping system in comparison to its nutrient input?" The NUE- AE answers a more direct question: "how much productivity improvement was gained by the use of this nutrient input?" (IPNI, 2012).

4.4.1.1 Nitrogen use efficiency (Partial factor productivity)

The partial factor productivity (NUE-PFP) was determined as a product of N uptake efficiency and N utilization efficiency and significant differences across genotypes were observed ($p < 0.001$) as shown in appendix VII .The NUE-PFP among all genotypes ranged from 13 to 94 kg grain/kg N (table 6). The check genotype had NUE PFP of 21.5 kg grain kg N^{-1}. Lines 2006, 2035, 2091 and 727 were superior to

the check line by 272, 215, 307 and 326%, respectively. Other lines 353, 645, 652, 1212, 1061 and 658 were also superior to the check by 42,55,64,75,125 and 138 %, respectively. Lines 60, 640, 10, 151, 695, 7 and 152 had NUE of less than 30 kg grain per kg N^{-1} while lines 375, 654, 650, 512, 353, 917, 645, 652 and 1212 had NUE between 30 and 38 kg grain per kg N^{-1}. Common values for this parameter range from 40–70 kg grain kg^{-1} N in most systems and values greater than 70 kg grain per kg N^{-1} are associated with low rates of N or with very efficiently managed systems (Doberman, 2005; Baligar *et al.,* 2001). N uptake efficiency had a greater bearing on NUE for all genotypes and strong correlation of r = 0.89 as shown in table 24.Genotypes with high NupE had higher NUE values.

4.4.1.2 Nitrogen Use efficiency (Agronomic efficiency)

The NUE Agronomic efficiency was significantly different ($p < 0.001$) (Appendix VIII) and ranged from 10 to 49 kg grain/kg N among the 30 genotypes (Table 7). The NUE - AE for the check was 14.8 kg grain kg N^{-1}. The genotypes 857,428 and 713 were superior to the check by 13, 15 and 17 % respectively; while the genotypes 375, 665,654, 512 and 353 were superior by 28, 29, 30, 34 and 36 % respectively; and the genotypes 917, 645,652,1212, 3080 and 1061 were superior by 42, 48, 63, 67, 77 and 84 %, respectively. Genotypes 2035, 658, 2026, 2006, 2091 and 727 were superior to the check by 122, 124, 154, 176, 182 and 203 % respectively. The NUE-AE is associated with a range of 10–30 kg grain kg^{-1} N and values greater than 30 kg grain kg^{-1} indicate well managed systems or at low levels of N use or low soil N supply (Doberman, 2005; Baligar *et al*., 2001). Data analysis shows that lines 658, 2006, 2091, 2035 and 727 had higher NUE-PFP and NUE-AE implying that they produced more grain yield for each kg of N applied compared to other genotypes.

Table 7. Nitrogen use efficiency Agronomic efficiency and Nitrogen use efficiency Partial factor productivity

Genotype	NUE-AE	NUE-PFP
L60	9.69a	13.51a
L640	11.46ab	15.93ab
L10	14.11abc	19.99abc
L151	14.82abcd	21.48abcd
L695	15.52abcd	21.74abcd
L7	16.09abcd	22.72abcde
L152	16.18abcd	23.28abcde
L666	16.29abcd	23.36abcde
L649	16.72abcd	23.44abcde
L857	18.06abcde	25.34abcdef
L428	18.43abcde	26.36abcdef
L713	18.76bcde	26.73abcdef
L375	20.49cdef	29.63bcdef
L665	20.56cdef	28.81bcdef
L654	20.81cdef	29.65bcdef
L650	20.86cdef	30.46cdef
L512	21.43cdef	31.13cdef
L353	21.71cdef	31.23def
L917	22.65cdef	32.15cdef
L645	23.71def	34.16def
L652	26.14ef	36.14efg
L1212	26.64ef	38.47fgh
L3080	28.33fg	47.17ghi
L1061	29.37fg	49.4hi
L2035	35.53gh	69.3j
L658	35.81gh	52.38i
L2026	40.64hi	81.78k
L2006	44.19i	81.87k
L2091	45.1i	89.55kl
L727	48.5i	94.42l
p	<0.001	<0.001
cv	19.2	18.4
lsd(0.05)	7.5	11.6
sed	3.8	5.8
mean	24	38.4

NUE-AE=nitrogen use efficiency agronomic efficiency, NUE-PFP= nitrogen use efficiency partial factor productivity. Values with same letters are not significantly different

4.4.2 Grain Nitrogen yield per hectare

The grain % N ranged from 1.3 to 2.5% and differences among genotypes were statistically significant ($p < 0.001$) (Appendix IX). Lines 60 and 727 had the highest and lowest levels of grain N respectively. The total grain % N ha^{-1} ranged from 31.6 to 268 kg /ha (Table 8). Genotypes 640,151 and 428 on average yielded grain N in yield per hectare of above 40 kg ha^{-1}.Genotypes 152, 713, 649, 695, 10, 857 and 7 had an average grain N yield between 55 and 59 kg/ha. Lines which had grain N ha^{-1} between 61 and 68 kg ha included Lines 7, 512, 857, 713 and 666. Lines 654, 665, 650, 917, 645, 353, 652, 1212 and 3080 had yields between 76 and 99 kg grain N ha^{-1}. Genotypes 353, 1212, 652, 3080, 1061, 658, 2035, 2006, 2026, 2091 and 727 yielded more grain N ha^{-1} by 68, 80, 80, 95, 165, 190, 225, 282, 302, 336 and 434 % respectively as compared to the check genotype 151 which had 50 kg grain N ha^{-1}.

4.4.3 Harvest Index (HI)

HI is a good indicator of high yields among genotypes. Significant differences were observed among the genotypes ($p < 0.001$) (Appendix X).The ratio of grain yield to biological yield ranged from 0.2 to 0.45 for the genotypes under evaluation (Table 8). Line 60 had the lowest HI while Line 2091 had the highest index. Lines with high HI were associated with high grain yielding ability. The genotypes with highest NHI had the highest efficiency of translocting N to the grain rather than to the stover. Lines with HI between 0.2 and 0.24 included 60, 7 and 10. Lines with HI from 0.26 to 0.31 include 512, 695, 654, 665, 713, 640, 1212, 666, 428, 645, 152, 649, 917 and 650.Genotypes with HI between 0.32 to 0.39 are 857, 353, 727, 2035,1061, 2026 and 375. Lines 3080, 2006, 652, 658 and 2091 had HI of 0.41, 0.42, 0.43, 0.44 and 0.45 respectively. The increases in HI was for genotypes Lines 652, 2006, 375, and 2091 were 37, 41, 44 and 66% more than the check line 151 genotype respectively which had a HI of 0.26 (Table 8).

4.4.4 Nitrogen Harvest Index (NHI)

Differences among the genotypes were highly significant at $p < 0.001$(Appendix XI). NHI is the ratio of N present in grain to total plant N content (Ng/Nt), analogous to the harvest index (HI), which is the ratio of grain to total biomass. It is a measure of

N translocation efficiency. The nitrogen harvest index ranged from 0.26 to 0.71 with line 60 having the lowest and Line 2091 the highest (Table 8). The pattern of data on NHI grouped genotypes in four classes. Lines 60, 7 and 512 were in the first class with NHI between 0.26 and 0.29. The second group comprised Lines375, 151, 428, 713, 1212, 695, 152, 645, 10 and 2035. This class had NHI values from 0.30 to 0.39. The third class comprised lines 3080, 654, 857, 666, 917, 640, 353, 727, 649, 665, 2006 and 2026. This group had NHI values from 0.41 to 0.49. The fourth and final group comprised Lines 658, 650, 1061, 652 and 2091. This group had NHI values from 0.50 to 0.71. Although not directly related to grain weight, NHI has significance for maximising grain protein content for a given amount of plant N (Hefyn and Aly, 2008). Furthermore, the genotypes with high N utilization efficiency had the higher NHI. Genotypes 2026, 658, 650, 1061, 652 and 2091 were at 60, 66, 66, 87, 87 and 140 % higher than the check 151 which had 0.30 values for NHI (Table 8).

Table 8. Grain N yield, Nitrogen harvest index and harvest index of genotypes

Genotype	Grain N ha^{-1}	NHI	HI
L60	31.62a	0.26a	0.2048a
L375	42.33ab	0.30abc	0.3889gh
L428	46.42abc	0.30abc	0.2732abcdef
L640	46.47abc	0.45abcdef	0.2781abcdef
L151	50.45abcd	0.30abc	0.2606abcde
L10	55.62abcd	0.38abcdef	0.2169ab
L152	57.75abcd	0.36abcdef	0.2775abcdef
L649	58.22abcd	0.47bcdef	0.2805abcdefg
L695	59.39abcde	0.35abcde	0.2476abcd
L7	60.8abcde	0.29ab	0.2149ab
L512	62.91abcde	0.28ab	0.2428abc
L857	64.83abcde	0.41abcdef	0.3238bcdefg
L713	66.36abcde	0.34abcd	0.2986abcdefg
L666	66.91abcde	0.42abcdef	0.2667abcde
L654	75.75bcde	0.41abcdef	0.295abcdefg
L665	77.06bcde	0.47bcdef	0.2701abcde
L650	77.63bcde	0.51def	0.3324cdefg
L917	79.73bcde	0.43abcdef	0.3109abcdefg
L645	82.18bcde	0.36abcdef	0.2794abcdefg
L353	83.45cde	0.46bcdef	0.31abcdefg
L652	89.83de	0.56fg	0.3709efgh
L1212	89.98de	0.35abcde	0.2884abcdefg
L3080	98.5e	0.41abcdef	0.3652efgh
L1061	134.91f	0.55ef	0.3591efgh
L658	146.71f	0.50cdef	0.3511cdefgh
L2035	163.99fg	0.39abcdef	0.3484cdefgh
L2006	195.29gh	0.48bcdef	0.3808fgh
L2026	203.89h	0.48bcdef	0.3661efgh
L2091	219.5h	0.72g	0.4501h
L727	268.09i	0.46bcdef	0.3563defgh
p	<0.001	<0.001	<0.001
cv	21.2	24	17.9
lsd(0.05)	32.9	0.16	0.1
sed	16.5	0.08	0.04
mean	95.2	0.42	0.3

NHI=nitrogen harvest index, HI= harvest index .Values with same letters are not significantly different

4.4.5 Biomass yield

Biomass yields exhibited significant differences statistically ($p < 0.001$) (Appendix XII). Biomass yield per hectare ranged from 5.7 t ha^{-1} for line L 640 to 26.6 tons $ha^{-1}$for Line 727 (Table 9). Differences were very significant statistically among genotypes. Five genotypes 151, 649,152, 666 and 695 produced biomass greater than 8 tons but less than 9 tons ha^{-1}. Lines that produced more 9 tons but less than 12 ton ha^{-1} were 713, 650, 428, 652, 10, 353, 654, 917, 7 and 665. Lines 645, 512, 3080, 1061, 1212 and 658 produced biomass above 12 tons ha^{-1} but less than15 tons ha^{-1}. Lines 2035, 2091, 2006, 2026 and 727 produced biomass between 20 tons and 26.6 tons ha^{-1} and these stayed green for a longer time than the others and this could have increased photosynthetic activities (not measured). In prolonged photosynthetic period, sufficient carbohydrates could have been translocated to the roots thereby maintaining their activities and eventually contributing to an increase in N uptake and increased biomass production.

4.4.6 Grain yield

Differences observed among genotypes were highly significant ($p < 0.001$) (Appendix XIII). Grain yield of genotypes ranged from 1.3 to 9 tons ha^{-1} (Tables 9). The lowest yielding genotype was line 60 and the highest yielding was 727. Half of the total genotypes had grain yields less than 3 tons ha^{-1} in the first grouping. The second grouping of genotypes which included lines 650, 512, 353, 917, 645, 652 and 1212 produced yields above 3 tons ha^{-1}.The third group produced yields above 4 tons ha^{-1} and lines associated with this group included 3080, 1061, 658, 2035, 2026, 2006, 2091 and 727. Lines 2026, 2006, 2091,727 and 2035 had grain yield greater than 6 tons/ha. For all genotypes, higher yields were mostly attributed to their considerable high harvest indices, more for dry-matter and for nitrogen. Lower yields could be associated with lower HI for the lines in the study. Grain yields for genotypes 727, 2091, 2006, 2026, 2035, and 658 were higher by 339, 316, 281, 280, 222 and 144% than the check genotype 151, respectively. Grain yield for the check was 2.1 tons ha^{-1}.

Table 9. Grain and biomass yield of genotypes

Genotype	Grain yield ha^{-1}	biomass yield ha^{-1}
L60	1351a	6622ab
L640	1593ab	5733a
L10	1999abc	9689abcdef
L151	2148abcd	8237abcd
L695	2174abcd	8756abcde
L7	2272abcde	10622bcdefg
L152	2328abcde	8400abcd
L666	2336abcde	8682abcde
L649	2344abcde	8267abcd
L857	2534abcdef	7956abc
L428	2636abcdef	9600abcdef
L713	2673abcdef	9126abcdef
L665	2881bcdef	10889cdefgh
L375	2963bcdef	7704abc
L654	2965bcdef	10104bcdefg
L650	3046cdef	9156abcdef
L512	3113cdef	12800efgh
L353	3123cdef	10000bcdefg
L917	3215cdef	10311bcdefg
L645	3416def	12222defgh
L652	3614efg	9600abcdef
L1212	3847fgh	13778gh
L3080	4717ghi	12948fgh
L1061	4940hi	13734gh
L658	5238i	14800h
L2035	6930j	20119i
L2026	8178k	22400i
L2006	8187k	21422i
L2091	8955kl	21334i
L727	9442l	26578j
p	<0.001	<0.001
cv	18.4	17
lsd(0.05)	1156.5	3428
sed	577.8	1712.7
mean	3839	12053

Values with same letters are not significantly different

4.4.7 Nitrogen utilization efficiency (in kg^{-1} N^{-})

The differences among genotypes in N utilization efficiency were very significant statistically ($p < 0.001$) (Appendix XIV). N utilization efficiency (ratio of grain yield to total N in plant (Nt) measures the response of grain yield to the total N in the

plant. Since total plant N is difficult to measure, the study considered only aboveground plant N. This parameter contributes to the overall NUE of plants. The genotypes had a wide range varying from 12 to 33 kg $^{-1}N^{-1}$ (Table 10). Lines 375, 2006, 1061, 652 and 2091 were efficient at utilising N more than the check lines by 53, 54, 55, 73 and 120 % respectively (Tables 10). The check NutE was 15 kg grain per kg N of total plant N. Four groups emerged, first comprising genotypes having values from 12 to 20 $kg^{-1}N^{-1}$. This included the following: 7, 60, 695, 151, 10, 512, 713, 666, 1212, 152, 645, 640, 654, 857, 727, 2035, 428, 353 and 917. The second group had efficiencies ranging from 20 - 24 $kg^{-1}N^{-1}$. Included in this category were 665, 658, 649, 650, 2026, 30080, 1061, 375 and 2006. The final group which had efficiencies between 26 to 30 $kg^{-1}N^{-1}$ included were the following, 652 and 2091.

4.4.8 Grain N accumulation efficiency

The grain N accumulation efficiency exhibited statistical differences among genotypes ($p < 0.001$) (Appendix XV). This assessed plant N extracting capabilities from the soil and subsequent accumulation in the grain. The grain N accumulation efficiency was an overall efficiency, indicating which plants extracted N from the soil and fertilizer and accumulated it in the grain by harvest. The overall efficiency of extracting N from soil and fertilizer ranged from 0.23 to 1.9 Ng/Ns (Table 10). Lines 512, 857, 713 and 666 had higher efficiencies of 20% more than check, Line 151. Lines 654, 665, and 650 were higher by 40%. Lines 917 and 645 accumulated more N than the check by 50%. Lines 353, 1212 and 652 had a higher value of 60% more than the check. The check line had 0.35 meaning that for every gram of available N absorbed from the soil and fertiliser, the plant translocated 0.35 to the grain. Lines 1061, 658, 2035, 2006, 2026, 2091 and 727 accumulated N in grain by 150,161,193,250,250,300 and 375% respectively more than the check (Table 10).

4.4.9 N uptake

N uptake among genotypes exhibited significant differences statistically at $p < 0.001$ (Appendix XVI). N uptake was higher in lines L645, 3080, L512, 1212, L658, L2006, L2091, L2026, 2035 and L727. N uptake ranged from 92 kg ha^{-1} to 250 kg ha^{-1} among genotypes. Lines 640 and 60 had the least N uptake and produced low biomass production. High N uptake in some genotypes could also be attributed to the

longer maturity and period of absorption of mineral-N from the soil as well as the higher root density (data not shown). According to the study of Jansson and Persson (1982), application of fertilizer N leads to an increase in the net mineralisation of soil N with subsequent consumption of the mineralised N by the crop. Singh and Arora (2001) working on wheat genotypes, attributed the levels of N uptake to demand by the plants. Furthermore, they concluded that growth and uptake were dependent on the amount of N being supplied to the genotype during the growing period. Borrell *et al*,(2001) cited in Hefyn and Aly (2008) studying sorghum established that roots of the stay green sorghum varieties maintained a greater capacity to extract N from soil as compared to non-stay green genotypes during kernel filling stage. Borass *et al*. (2003) cited in the same publication indicated that delayed senescence during grain filling was connected to amount of light interception and N availability through remobilization to actively growing kernels of maize. Bänziger *et al,* (2002) also have reported that stay green genotypes exhibit enhanced water and nutrient uptake.

4.5 N uptake efficiency

Significant differences were observed amongst the genotypes ($p < 0.001$) (Appendix XVII). This ratio ranged from 0.92 – 5.097 (Table 10) and only nine genotypes were above the check whose N uptake efficiency value was 1.44. Genotypes Lines 658, 2091, 2006, 2035, 2026 and 727 had a much higher N uptake efficiency ratio than the check value of 1.44 by 41, 60, 73, 93, 147 and 240 % respectively (Table 10). Many authors have attributed differences in NUE to differences in N uptake efficiency. Employing a ^{15}N approach, Ma and Dwyer (1998) discovered that prior genetic improvement for N use Efficiency (NUE) can be explained by greater N uptake and improved dry matter production. Van Sanford and Mac Kown (1986) and Dhugga and Waines (1989) working with wheat reported that genetic variation plays a role in varying N uptake efficiency.

Table 10. Grain nitrogen accumulation efficiency, Nitrogen uptake efficiency, Nitrogen utilisation efficiency and Nitrogen biomass yield of genotypes

Genotype	GNACE	NupE	NutE	N B yield ha^{-1}
L60	0.224a	1.095ab	12.57a	109.5ab
L375	0.304ab	1.282abc	23.28cd	128.2abc
L640	0.332abc	0.92a	17.73abcd	92a
L428	0.337abc	1.381abcd	19.03abcd	138.1abcd
L151	0.355abc	1.438abcd	14.95abc	143.8abcd
L10	0.399abcd	1.38abcd	15.27abc	138abcd
L152	0.413abcd	1.379abcd	16.94abc	137.9abcd
L649	0.416abcd	1.105ab	21.61bcd	110.5ab
L695	0.432abcde	1.51abcd	14.38ab	151abcd
L7	0.441abcde	1.871cde	12.23a	187.1cde
L512	0.453abcde	1.983cdef	15.76abc	198.3cdef
L857	0.464abcde	1.386abcd	18.52abcd	138.6abcd
L713	0.469abcde	1.721bcde	15.85abc	172.1bcde
L666	0.478abcde	1.413abcd	16.39abc	141.3abcd
L654	0.540bcde	1.633abcd	18.23abcd	163.3abcd
L665	0.554bcde	1.517abcd	20.32abcd	151.7abcd
L650	0.558bcde	1.413abcd	22.03bcd	141.3abcd
L917	0.574bcde	1.653abcd	19.39abcd	165.3abcd
L645	0.587bcde	2.016cdef	17.18abcd	201.6cdef
L353	0.606cde	1.608abcd	19.29abcd	160.8abcd
L1212	0.641de	2.378efg	16.85abc	237.8efg
L652	0.643de	1.39abcd	25.72d	139abcd
L3080	0.703e	2.118def	22.36bcd	211.8def
L1061	0.956f	2.122def	23.23cd	212.2def
L658	1.045f	2.586fg	20.56abcd	258.6fg
L2035	1.166fg	3.695i	18.9abcd	369.5i
L2006	1.372gh	3.53hi	23.35cd	353hi
L2026	1.439h	3.661i	22.34bcd	366.1i
L2091	1.562h	2.954gh	33.2e	295.4gh
L727	1.913i	5.097j	18.63abcd	509.7j
p	<0.001	<0.001	<0.001	<0.001
cv	21.1	19.3	22.8	19.3
lsd(0.05)	0.23	0.62	7.2	62.4
sed	0.12	0.31	3.6	31.2
mean	0.7	1.98	19.2	197.5

GNACE= Grain nitrogen accumulation efficiency, NupE= Nitrogen uptake efficiency, NutE= nitrogen utilisation efficiency, N B yield = Nitrogen biomass yield. Values with same letters are not significantly different

The ranges of different nitrogen use efficiency components are shown in table 11 and in addition, the lowest and highest genotypes for each component are indicated.

Table 11. Combined summary ranges for NUE Parameters

NutE genotype			HI genotype			NUE - AE genotype		
Range	Lowest	Highest	Range	Lowest	Highest	Range	Lowest	Highest
12-33	L7	L727	0.2-0.45	L60	L2091	10-49	L60	L727

NUE - PFP genotype			**NHI genotype**			**Grain N Accum genotype**			**NupE genotype**		
Range	Lowest	highest	Range	Lowest	Highest	Range	lowest	highest	Range	lowest	highest
13-77	L60	L727	0.26-0.7	L60	L2091	0.22-1.91	L60	L727	0.9-5.1	L640	L727

4.6 Cluster Analysis

The general cluster analysis was done according to Rohlf (1998) to group genotypes that were closely related based on NUE parameters. The cluster (Figure 3) shows that the genotypes were grouped in four categories when Nitrogen use efficiency (PFP and AE), N utilisation efficiency, N uptake efficiency, harvest index, Nitrogen harvest index, grain yield and biomass yield were considered. Lines 917, 353, 654, 857, 649, 665, 650, 640, 151, 695, 10, 152, 666, 713, 428, 7, 512, 1212, 645, 60 and 375 belong to the first cluster. This cluster comprised genotypes with very low NUE meaning that they produced less than 28 kg of grain per kg N applied to the crop.

The second cluster comprised Lines 652, 658, 1061 and 3080 and these have a superior NUE compared to the previous cluster and these accumulated 28-35 kg grain per kg N applied. The third cluster had only 2091. This genotype had superior N utilisation efficiency and NUE, hence, it being clustered alone. The fourth cluster had Lines 2006, 2026, 2035 and 727 with superior NUE. Clusters three and four in the whole group of genotypes accumulated 44- 48 kg grain per N applied.

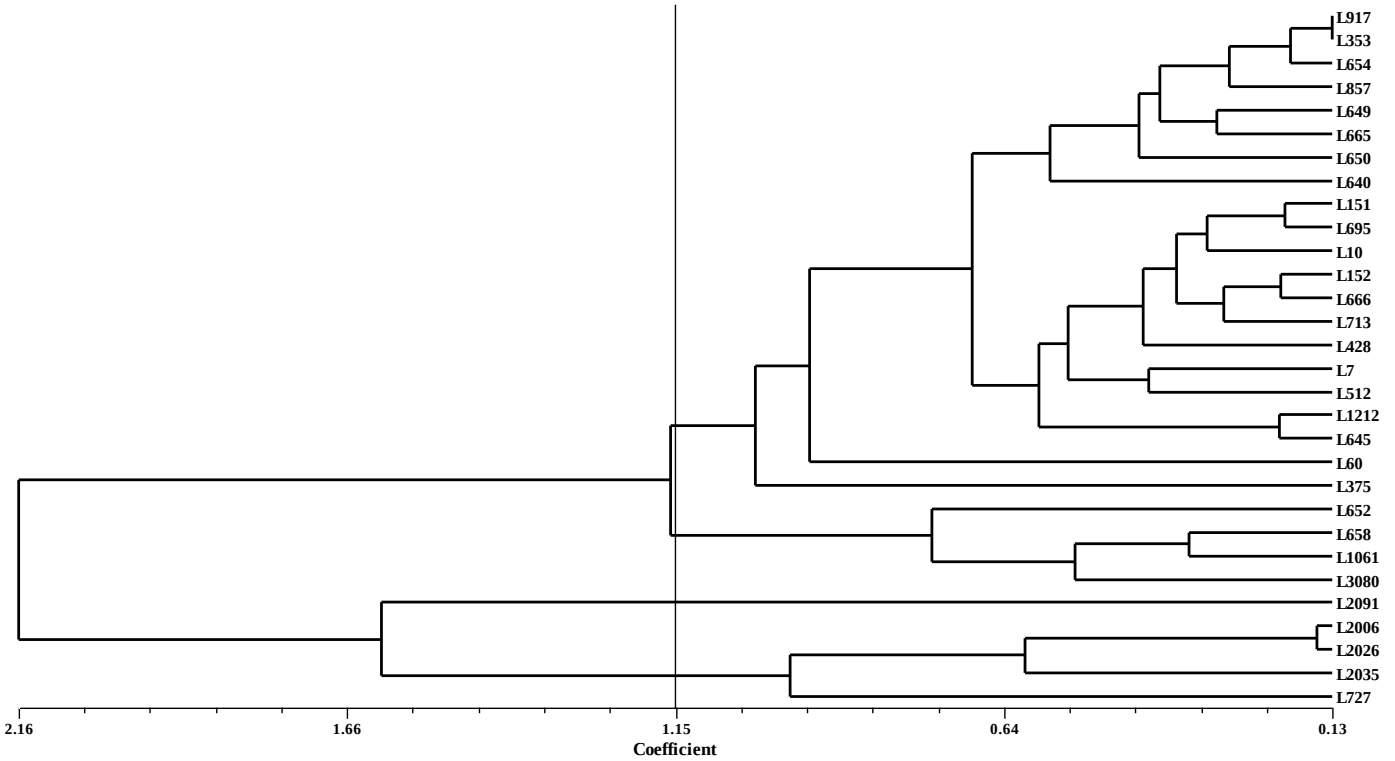


Figure 3 Dendogram showing clusters of genotypes in the experiment based on nitrogen use efficiency parameters

Correlation matrix of NUE parameters and yield components was developed to show the relationships as shown in Table 12. Nitrogen use efficiency, NHI and HI were positively correlated to grain yield at r = 0.97, 0.47 and 0.65 respectively. Total grain % N /ha and biomass yield /ha were also highly correlated to NUE at 0.92. The correlation between NUE and grain N accumulation was positive and very high. The r value was 0.95 and this is in agreement with Hefyn and Aly (2008) who showed that grain N accumulation and NUE in his study were strongly related statistically.

Nitrogen utilisation efficiency was positively correlated to grain yield at r = 0.50, NUE-PFP and NUE-AE at 0.5 and 0.52, respectively. Nitrogen use efficiency was positively correlated to N utilization, N uptake, biomass N, grain N accumulation, NHI and grain yield at r = 0.5, 0.87, 0.1, 0.9, 0.5 and 0.9, respectively.

Grain yield was positively correlated to NupE, N grain accumulation, NHI, TGN, NUE-AE, NUE-PFP, HI, and NutE at 0.89, 0.97, 0.47, 0.97, 0.97, 1.00, 0.65 and 0.50, respectively. NutE was highly negatively correlated to % N biomass at r = - 0.5 and positively correlated to the harvest index at r = 0.89. Genotypes with relatively high HI had better N utilisation efficiency.

Harvest index was highly correlated with NUE- AE and NUE-PFP at 0.66 and 0.65, respectively, in this study. The secondary traits namely: plant height, leaf length and leaf width correlated positively to NUE with a very weak relationship. The same traits were highly and positively correlated to biomass yield of the genotypes in the experiment.

Table 12. Correlation matrix for Nitrogen use efficiency parameters

	%_N_BM	%_N Grain	BY ha	GY ha	Grain N ha	NutE	HI	NUE PFP	NUE AE	NupE	N yield BM	Grain N acc	NHI	TGN ha
% N BM	-													
% N Grain	0.12ns	-												
BY ha^{-1}	0.17ns	0.01ns	-											
GY ha^{-1}	0.11ns	-0.03ns	0.93**	-										
Grain N ha^{-1}	0.16ns	0.18 ns	0.92**	0.97**	-									
NutE	-0.50**	-0.19ns	0.22**	0.50**	0.44**	-								
HI	-0.08ns	-0.18ns	0.35**	0.65**	0.60**	0.89**	-							
NUE PFP	0.11ns	-0.03ns	0.93**	1.00**	0.97**	0.50**	0.65**	-						
NUE-AE	0.10ns	-0.05ns	0.91**	0.97**	0.95**	0.52**	0.66**	0.97**	-					
NupE	0.39**	0.05ns	0.97**	0.89**	0.89**	0.09ns	0.31ns	0.89**	0.87**	-				
N yield BM	0.39**	0.05ns	0.97**	0.89**	0.89**	0.09ns	0.31ns	0.89**	0.87**	1**	-			
Grain N acc	0.16ns	0.18ns	0.92**	0.97**	1.00**	0.44**	0.60**	0.97**	0.95**	0.89**	0.89**	-		
NHI	-0.42**	0.33ns	0.23**	0.47**	0.53**	0.86**	0.76**	0.47**	0.48**	0.12ns	0.12ns	0.53**	-	
TGN_ha	0.16ns	0.18ns	0.92**	0.97**	1.00**	0.44**	0.60**	0.97	0.95**	0.89**	0.89**	1**	0.53**	-

%N BM - Nitrogen content in Biomass,**% N Grain**- N in grain as %, **BY ha^{-1}** -biomass yield, **GY ha^{-1}** -grain yield per ha, **Grain N ha^{-1}** -total grain N per ha, **NutE**- N utilisation efficiency, **HI**- harvest index**, NUE**- Nitrogen use efficiency(**AE** agronomic efficiency, **PFP** partial factor productivity), **NupE**- N uptake efficiency, **N yield BM** - Nitrogen yield in biomass per ha**, Grain N acc**- grain N accumulation, **NHI**- Nitrogen harvest index, **TGN/ ha**- total grain N per ha.
ns- not significant, **- highly significant ($p < 0.05$)

4.7 Relationships among plant genotypic attributes and NUE parameter.

Identifying genotypes that utilise applied N more efficiently is a potential way of reducing N losses through leaching and denitrification (Davis, 2013). The NUE technology has been used and still has potential as a means of addressing challenges of low available N in agricultural soils and availability of inorganic N fertilisers to farmers.

Nitrogen use efficiency in these genotypes was mostly explained by the level of efficiency of N uptake. This means genotypes that absorbed more N from the soil and transformed it into biomass N were associated with high NUE (PFP and AE).The strong correlation between NUE-AE and NUE-PFP are indicative of the degree of economic and environmental efficiency in the use of nutrients. Most N efficient genotypes had higher values of the NupE component and correspondingly high values for the determined NUE and this was in agreement with studies done by Dhugga and Waines (1989) who showed that N uptake efficiency accounted for a larger portion of NUE. Hefyn and Aly (2008) evaluating the yielding ability and NUE in maize inbreds and their crosses found out that NupE was responsible for high NUE. Raun and Johnson (1999) advocated for improving NupE as a way of increasing NUE.

Nitrogen utilisation efficiency for genotypes showed varying association with other NUE parameters but not as high as NupE in this study. Its correlation with grain yield and NUE was at $r = 0.5$ for both parameters and significant. NutE accounted for a less proportion of NUE as compared to NupE. Bertin and Gallais (2000) in their study with maize suggested that both NupE and NutE were the main components of NUE. They reported that NUE increased with increased NupE at high N application rates while NutE at low N application. Nitrogen utilisation efficiency indicated the ability of the plant to translocate N uptakes into grain. The correlation between NutE and grain yield suggested that there was remobilization and partitioning of N from vegetative plant parts to the grain. Nitrogen utilisation and N uptake efficiencies played a substantial role in promoting grain yield in this study contrary to the results of Laffitte and Edmeades (1994) who established that N utilisation efficiency alone played a leading role in determining grain yield.

In this study, there was no correlation between NupE and NutE . Presterl *et al.* (2002) stated that a balanced combination of the two constituents was essential to accomplish greater grain yields; however, they established a negative correlation between the two components.

The harvest indices of the genotypes had a significant correlation with NUE and grain yield at r = 0.65. Harvest index affected NUE as it has a bearing on yield. Harvest index was a good indicator of grain yield in these genotypes in this study. Genotypes with high HI accumulated more assimilate above ground and translocated them to grain during kernel filling. This is in agreement with finding by Gallais and Hirel (2003) and Di Fonzo *et al*. (1982) in their studies with maize who found that genotypes with high harvest index were high yielding when compared to those with low harvest index. Genotypes with high HI also were associated with high NUE as this is a source – sink relationship (Coque and Gallais, 2007). Decline in HI has been reported at low N application in inbred lines and it has been suggested that N deficiency in plants is responsible for reduced growth rate and biomass production hence the low partitioning of assimilates to the grain. Inamullah *et al*. (2011) in their study found a correlation between HI, NUE and yield in maize hybrids.

In this study it was established that NHI was also significantly correlated to NUE and grain yield at r = 0.47. Previously, Di Fonzo *et al*. (1982) also showed low N input grain yield was related to NHI. Some genotypes like 658 and 2091 exhibited efficient partitioning of N assimilates to the grain and producing more grain yield per kg of N at this rate of N application. Both NUE and NHI in these genotypes were high. In a similar study with inbred lines, Hefyn and Aly (2008) showed that some of the genotypes had high NHI and NUE at low N application. High NHI is fundamental in better-quality nutrition as it reflects the grain protein content and thus the grain nutritional quality (Sinclair1998) who found that NHI was associated with high grain protein. These genotypes with high NHI could be used as sources of high quality grain for purposes of improved nutrition in rural communities in line with the millennium development goal. It was observed that genotypes with a high NutE value also had high N in grain. Nitrogen harvest index was positively and highly correlated to N utilisation efficiency at r = 0.86 in this study.

Genotypes with high grain N accumulation had the highest yield. This suggests that these genotypes were efficient at absorbing N from the soil and fertiliser and later translocating it to grain production and subsequently protein.

The N uptake in this study varied considerably among genotypes despite the same N application, water supply and management practices. Differences in N uptake could only be attributed to the genetics of the plant. Genotypes with high uptake correlated highly with grain yield at r = 0.89. It can be speculated that genotypes that had a high N uptake could have had an extensive and well developed root system (Baligar, *et al*. (2001). Ortiz-Monasterio *et al.* (1997) associated high N supply in the soil with uptake. Genotypes with higher N uptake produced more biomass in this study at this level of N application, suggesting a relationship between the two.

The length of the growing period also affected N uptake. Genotypes that showed late senescence had high N uptake compared to genotypes that senesced earlier. Prolonged photosynthesis increased N uptake in genotypes that had long growing period. In prolonged photosynthetic period it is speculated that sufficient carbohydrates were translocated to the roots thereby maintained their activities and eventually contributed to an increase in N uptake. Coque and Gallais (2007) in their study concerning N uptake in maize showed that prolonged photosynthesis in genotypes that stayed green for a long time enhanced N uptake. This is in agreement with this study.

Nitrogen use efficiency- Agronomic Efficiency (kg grain $kg^{1}N$) showed great variation among genotypes and this was greatly affected by the N uptake efficiency. Genotypes that absorbed more available N from the soil used it more efficiently to produce more grain than those that absorbed less. In this study, differences in NUE kg biomass kg^{-1} N in terms of biomass for the genotypes was not as wide as the NUE kg grain kg N^{-1} in terms of grain when compared. This could be attributed to some inability to translocate assimilates to the grain despite producing appreciable amounts of biomass during the growing period. There was high and significant correlation between yield and NUE at r =0.97. Other studies have also shown correlations between NUE and yield (Bertin and Gallais, 2000); Hirel *et al*., 2001) in maize. Genotypes with high NUE and NupE could further be evaluated to find out whether these two traits are highly heritable The study did not any correlation

between grain yield and N in the grain r = - 0.03.This is contrary to the findings of other authors who showed negative correlation between grain yield and N in grain or oil concentration in most cereals (Simmonds, 1995) including maize (Feil *et al.*, 1990) and oilseed rape (Jackson, 2000).

Tall plants were associated with higher biomass yield in this study. Information is limited and inconsistent about the relationships of maize yield and plant biomass with plant height. Yin *et al*. (2011) found that maize yield was, in general, strongly related to plant height during early (six-leaf growth stage, V6) to mid (V12) season under a wide range of N rate treatments and major cropping systems when the weather conditions were normal. To the contrary, Katsvairo *et al*. (2003) and Machado *et al*. (2002) found that the correlation of maize yield with plant height measured during early and mid-season was not consistent across locations and years.

Genotypes with high NupE and grain N accumulation efficiency also had high NUE suggesting that N partitioning had an effect on determining the overall NUE. Some genotypes like 658 and 2091 exhibited efficient partitioning of N assimilates to the grain and producing more grain yield per kg of N at this rate of N application. Both NUE and NHI in these genotypes were high. In a similar study with inbred lines Hefy and Aly (2008) discovered that some of the genotypes had high NHI and NUE at low N application.

Grain N accumulation was also correlated to N utilisation. Some genotypes like 658 and 2091 exhibited efficient partitioning of N assimilates to the grain and producing more grain yield per kg of N at this rate of N application. Both NUE and NHI in these genotypes were high. In a similar study with inbred lines Hefyn and Aly (2008) showed that some of the genotypes had high NHI and NUE at low N application.

Nitrogen use efficiency has been defined as the amount of grain produced per unit available soil N (Moll *et al.,* 1982) and was extended by Huggins and Pan (2003) to partition differences in yield and grain N between cropping systems into components associated with soil and plant processes. A maize genotype that uses N efficiently can be defined as a genotype that absorbs large amounts of N from the soil and from fertiliser, producing high grain yield per unit absorbed N and leaving only little N in the straw.

From a physiological point of view, the study recorded a negative relationship between grain yield and ASI. Previous studies by Gallais and Hirel (2003), Hefyn and Aly (2008) also indicate the similar result.

4.8 Principal component Analysis

To understand the degree of contribution of each NUE parameter to variation a Principal Component Analysis (PCA) was carried out to identify patterns and to highlight similarities and differences (Figure 4). From the analysis PCA results identified NUE and NutE as major contributors to the variation. Nitrogen use efficiency contributed 73% to the variation, while N utilisation efficiency contributed 18%. The two traits contributed a total of 91 % of the variation. The eigen value for the two were greater than one (1) eigenvalue threshold. Nitrogen uptake efficiency, grain N accumulation, HI, NHI, yield, and BMY had less than one (1) eigenvalue (Appendix xviii).

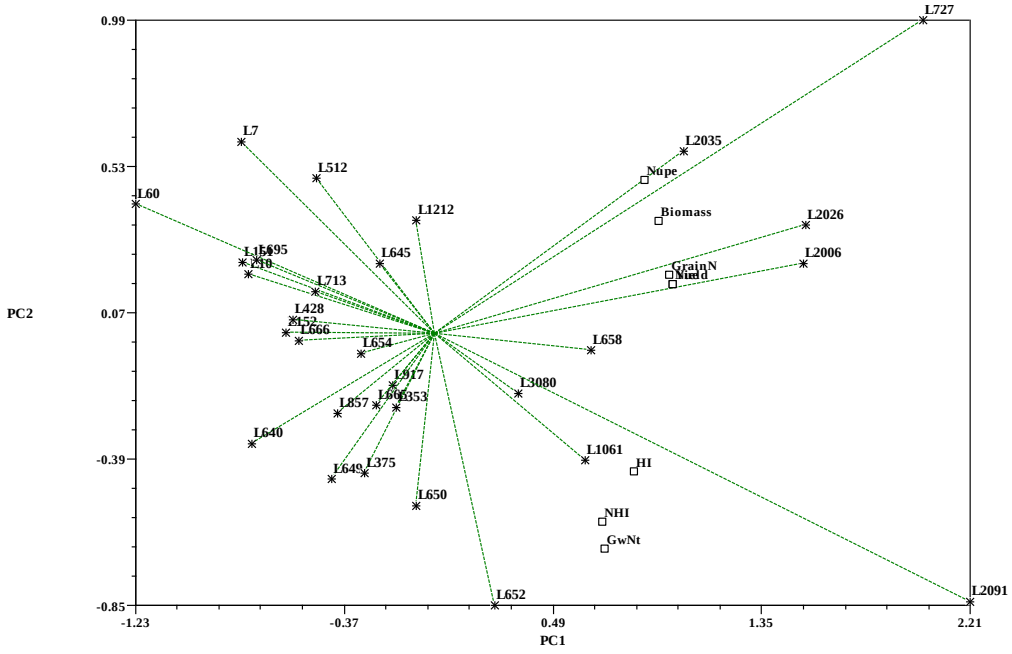


Figure 4 Grouping of genotypes based on NUE parameters. Square represent Nitrogen use efficiency parameters and the stars * represent 30 genotypes in study

CHAPTER FIVE

5.0 CONCLUSION

The study was conducted to evaluate and screen for nitrogen use efficiency with the specific objectives (i) to determine the NUE of different maize genotypes, (ii) to determine N partitioning in maize genotypes to grain and biomass and (iii) to identify secondary traits associated with nitrogen use efficiency.

The evaluation of NUE showed differences among the 30 genotypes. The results of this study show that among the 30 genotypes evaluated, 6 lines (658, 2035, 2026, 2006, 2091 and L 727) produced NUE-AE higher than the reference genotype L151 with 14.8 kg grain per kg N applied by 121. 124, 154, 176,182 and 203%, respectively. For each kg of N applied these genotypes produced more grain than the rest of the genotypes.

Nitrogen partitioning among genotypes evaluated using the Nitrogen harvest index ranged from 0.3 – 0.7. The genotypes that partitioned more of the vegetative N to the grain included 917, 2091, 652, 1061, 650, 658 and 2026. Based on the correlation analysis, genotypes with higher N utilisation efficiency partitioned more N to the grain.

Overall, the NUE was determined mainly by the individual genotype and its interaction with the soil and environmental conditions. Secondary traits such as delayed senescence (stay green) N uptake, N uptake efficiency and grain N accumulation positively contributed to NUE. Based on the NUE-AE, the genotypes 658, 2035, 2026, 2006, 2091 and L 727 can be included in the next stage of the breeding programme.

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CHAPTER SIX

6.0 RECOMMENDATIONS

In order to improve NUE one has to manipulate N uptake, thus a more in-depth study of genotypes root morphology, root functioning and root specific N enzymes and N transporters would be very valuable. Furthermore, there is need to probe whether the large plant size genotypes and greater growth rate is as a result of efficient recovery of fertiliser N or whether large plants are able to capture a greater amount of soil N due to the extent of root structure. In this regard, the N-15 be used to verify some N uptake and NupE values for all genotypes that were evaluated. Seed should be multiplied in advance to increase the quantities to avoid non availability as inbred lines are very sensitive to abiotic and biotic factors during plant growth. Genotypes with high NUE should further be evaluated in different soil types under rainfall conditions to assess if the performance will be the same as that under irrigation. Genotypes could further be evaluated to ascertain their partitioning ability under different N application rates of N and protein production.

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APPENDICES

Appendix I: Fifty percent (50%) Silking

Analysis of variance Variate: silking Source of variation	d.f.	(m.v.)	s.s.	m.s.	v.r.	F pr.
REP stratum	2		163.25	81.63	2.88	
REP.*Units* stratum						
GENOTYPE	29		1603.37	55.29	1.95	0.016
Residual	57		1615.98	28.35		
Total	88		3238.81			

Appendix II: Fifty percent (50 %) Tassel

Analysis of variance Variate: tassel Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
REP stratum	2	134.16	67.08	2.88	
REP.*Units* stratum					
GENOTYPE	29	1451.27	50.04	2.15	0.007
Residual	57	1327.43	23.29		
Total	88	2831.28			

Appendix III: Plant height

Accumulated analysis of variance

Variate: PH

Change	**d.f.**	**s.s.**	**m.s.**	**v.r.**	**F pr.**
REP	2	1866	933	4.31	0.018
GENOTYPE	29	92671.3	3195.6	14.76	<.001
Source	2	446.8	223.4	1.03	0.363
GENOTYPE.Source	1	316.9	316.9	1.46	0.232
Residual	55	11911.1	216.6		
Total	89	107212	1204.6		

Appendix IV: Leaf length

Variate: LL					
Change	d.f.	s.s.	m.s.	v.r.	F pr.
REP	2	3.47	1.73	0.04	0.962
GENOTYPE	29	8109.86	279.65	6.3	<.001
Source	2	2.24	1.12	0.03	0.975
GENOTYPE.Source	1	3.23	3.23	0.07	0.788
Residual	55	2440.34	44.37		
Total	89	10559.14	118.64		

Appendix V: Leaf width

Variate: LW					
Change	**d.f.**	**s.s.**	**m.s.**	**v.r.**	**F pr.**
REP	2	0.8453	0.4226	0.66	0.518
GENOTYPE	29	183.814	6.3384	9.97	<.001
Source	2	0.8574	0.4287	0.67	0.514
GENOTYPE.Source	1	0.1029	0.1029	0.16	0.689
Residual	55	34.9681	0.6358		
Total	89	220.5877	2.4785		

Appendix VI: Tassel size

Analysis of variance
Variate: tassel_size

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
REP stratum	2	0.1556	0.0778	0.23	
REP.*Units* stratum					
GENOTYPE	29	31.6556	1.0916	3.19	<.001
Residual	58	19.8444	0.3421		
Total	89	51.6556			

Appendix VII: Nitrogen Use Efficiency, Partial Factor Productivity

Analysis of variance

Variate: NUE-PFP

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
REP stratum	2	5.19	2.60	0.05	
REP.*Units* stratum					
GENOTYPE	29	44255.80	1526.06	30.48	<.001
Residual	58	2904.03	50.07		
Total	89	47165.02			

Variate: NUE-PFP

Grand mean	38.39
s.e.d.	5.778
l.s.d.	11.565

Variate: NUE- PFP

Stratum	d.f.	s.e.	cv%
REP	2	0.294	0.8
REP.*Units*	58	7.076	18.4

Appendix VIII: Nitrogen Use Efficiency AE

Analysis of variance

Variate: NUE-AE

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
REP stratum	2	4.00	2.00	0.09	
REP.*Units* stratum					
GENOTYPE	29	9196.02	317.10	14.95	<.001
Residual	58	1230.51	21.22		
Total	89	10430.53			

Variate: NUE-AE

Grand mean	23.95
s.e.d.	3.761
l.s.d.	7.528

Variate: NUE-AE

Stratum	d.f.	s.e.	cv%
REP	2	0.258	1.1
REP.*Units*	58	4.606	19.2

Appendix IX: Grain N ha^{-1}

Analysis of variance

Variate: Grain_N_ha

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
REP stratum	2	25.6	12.8	0.03	
REP.*Units* stratum					
GENOTYPE	29	305274.9	10526.7	25.92	<.001
Residual	58	23553.9	406.1		
Total	89	328854.4			

Variate: Grain_N_ha

Grand mean	95.2
s.e.d.	16.45
l.s.d.	32.94

Variate: Grain_N_ha

Stratum	d.f.	s.e.	cv%
REP	2	0.65	0.7
REP.*Units*	58	20.15	21.2

Appendix X: Harvest index

Analysis of variance

Variate: HI

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
REP stratum	2	0.001820	0.000910	0.30	
REP.*Units* stratum					
GENOTYPE	29	0.294190	0.010144	3.35	<.001
Residual	58	0.175703	0.003029		
Total	89	0.471713			

Variate: HI

Grand mean	0.3070
s.e.d.	0.04494
l.s.d.	0.08996

Variate: HI

Stratum	d.f.	s.e.	cv%
REP	2	0.00551	1.8
REP.*Units*	58	0.05504	17.9

Appendix XI: Nitrogen Harvest Index

Analysis of variance

Variate: NHI

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
REP stratum	2	0.025168	0.012584	1.26	
REP.*Units* stratum					
GENOTYPE	29	0.852406	0.029393	2.95	<.001
Residual	58	0.577358	0.009954		
Total	89	1.454931			

Variate: NHI

Grand mean	0.415
s.e.d.	0.0815
l.s.d.	0.1631

Variate: NHI

Stratum	d.f.	s.e.	cv%
REP	2	0.0205	4.9
REP.*Units*	58	0.0998	24.0

Appendix XII: Biomass yield/ ha

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
REP stratum	2	1.163E+07	5.813E+06	1.55	
REP.*Units* stratum					
GENOTYPE	29	2.405E+09	8.293E+07	22.11	<.001
Residual	58	2.175E+08	3.751E+06		
Total	89	2.634E+09			

Appendix XIII: Grain yield ha^{-1}

Analysis of variance

Variate: Grain yield ha^{-1}

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
REP stratum	2	51911.	25956.	0.05	
REP.*Units* stratum					
GENOTYPE	29	442558020.	15260621.	30.48	<.001
Residual	58	29040316.	500695.		
Total	89	471650247.			

Variate:Grain Yield ha^{-1}

Grand mean	3839.
s.e.d.	577.8
l.s.d.	1156.5

Variate: Grain Yield ha^{-1}

Stratum	d.f.	s.e.	cv%
REP	2	29.4	0.8
REP.*Units*	58	707.6	18.4

Appendix XIV: Nitrogen utilization efficiency

Analysis of variance

Variate: Nitrogen utilization efficiency

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
REP stratum	2	46.38	23.19	1.21	
REP.*Units* stratum					
GENOTYPE	29	1573.19	54.25	2.83	<.001
Residual	58	1113.34	19.20		
Total	89	2732.91			

Variate: Nitrogen utilization efficiency

Grand mean	19.20
s.e.d.	3.577
l.s.d.	7.161

Variate:NutE

Stratum	d.f.	s.e.	cv%
REP	2	0.879	4.6
REP.*Units*	58	4.381	22.8

Appendix XV: Grain N accumulation

Analysis of variance

Variate: Grain N accumulation

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
REP stratum	2	0.00075	0.00038	0.02	
REP.*Units* stratum					
GENOTYPE	29	15.28150	0.52695	25.58	<.001
Residual	58	1.19484	0.02060		
Total	89	16.47709			

Variate: Grain N accumulation

Grand mean	0.679
s.e.d.	0.1172
l.s.d.	0.2346

Variate: Grain N accumulation

Stratum	d.f.	s.e.	cv%
REP	2	0.0035	0.5
REP.*Units*	58	0.1435	21.1

Appendix XVI: Total Nitrogen yield in biomass

Analysis of variance

Variate: N yield biomass

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
REP stratum	2	2521.	1261.	0.87	
REP.*Units* stratum					
GENOTYPE	29	784659.	27057.	18.57	<.001
Residual	58	84491.	1457.		
Total	89	871671.			

Variate: N yield biomass

Grand mean	197.5
s.e.d.	31.16
l.s.d.	62.38

Variate: N yield biomass

Stratum	d.f.	s.e.	cv%
REP	2	6.48	3.3
REP.*Units*	58	38.17	19.3

Appendix XVII: Nitrogen Uptake efficiency

Analysis of variance

Variate: NUpE

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
REP stratum	2	0.2521	0.1261	0.87	
REP.*Units* stratum					
GENOTYPE	29	78.4659	2.7057	18.57	<.001
Residual	58	8.4491	0.1457		
Total	89	87.1671			

Variate: NUpE

Grand mean	1.975
s.e.d.	0.3116
l.s.d.	0.6238

Variate: NUpE

Stratum	d.f.	s.e.	cv%
REP	2	0.0648	3.3
REP.*Units*	58	0.3817	19.3

Appendix XVIII : Eigen values of NUE parameters

Parameter	Eigen value	percent	cumulative
NUE-AE	4.37	72.79	72.79
NutE	1.09	18.18	90.96
HI	0.32	5.38	96.35
NHI	0.21	3.54	99.89
BMY	0.01	0.11	100.00
Grain yield	0.00	0.00	100.00

Appendix: XIX .Soil profile

Soil Profile No.:	SP/2009/001
Location:	National Irrigation Research Station (NIRS) Nanga, Mazabuka
Latitude:	15º 46’51.6”S
Longitude:	27º 55’28.5”E
Elevation:	
Author:	
Date:	09/03/2009
Land use class and Code:	
Drainage:	Well drained
Permeability:	Medium
Land physiography:	Degraded Central African Plateau, interfluve
Slope:	Simple; 0-2% - middle slope depression
Parent material:	Limestone dolomite
Depth:	Deep
Agro-climatic zone:	IIa 9
Vegetation and land use:	Munga woodland, cropland
Soil Classification: Soil Taxonomy – USDA: FAO/UNESCO	Fine, kaolinitic, isohyperthermic typic kanhaplustalf

Horizon	Depth (cm)	Description
Ap	0 – 10	Yellowish red (5YR 5/6) dry, brown (7.5YR 4/4) moist, sandy clay; sub angular blocky-moderate medium, hard, firm, sticky, plastic; unspecified accumulations clay; few, medium, irregular, unfilled other insect holes in local concentrations; many fine, random and few medium, random roots; many, moderately porous, fine, inped, interstitial and common medium, exped, tubular pores; clear smooth boundary.
BA	10 – 27	Yellowish red (5YR 4/6)dry, dark reddish brown (5YR ¾) moist, sandy clay; strong, medium, sub angular blocky; hard, firm, sticky and plastic; few, faint, thin, dark reddish brown clay skins on both horizontal and vertical ped faces; unspecified accumulations of clay; common, medium, irregular and tubular unfilled termite holes and other insect holes in local concentrations; many, fine, random roots; many, moderately porous fine, inped, interstitial, many, exped, tubular and few, course,

Bt	27 – 60	exped, vesicular pores; course gritty fragments of 10% volume; gradual, smooth boundary.
Btz_1	60 – 82	Strong brown (7.5YR 4/6) dry, brown (7.5YR 4/4) moist, clay; strong, fine and strong, medium, sub angular blocky; hard, friable, very sticky, plastic; common, distinct, moderately thick, brown clay skins on both horizontal and vertical ped-faces; common, spherical, unspecified accumulations of clay in local concentration, many medium and coarse, spherical, irregular and tubular other insect holes with unfilled and partly filled soil material in local concentrations; common, fine, random roots; many, fine, moderately porous, inped; interstitial, many, medium, exped, tubular and common, coarse, exped, vesicular pores; course, gritty at 20% volume; abrupt, wavy boundary.
Btz_2	83 – 120	Strong brown (7.5YR 5/6) dry, strong brown (7.5YR 4/6) moist; gritty clay; weak, coarse sub angular blocky; slightly hard, very friable, slightly sticky, plastic; moderately weakly cemented, sesquioxides (Fe, Al, Mn) and carbonates, discontinues over 1m; clay bridges, carbonate coats and manganese coats; many medium, spherical, yellow nodules and concretions iron-manganese throughout the horizon; few, medium irregular other insect holes with unfilled and partly filled soil material in local concentrations; few, fine random roots; many fine, moderately porous, inped, interstitial and common, medium, exped, tabular pores; coarse, gravelly rock fragments at 50% volume; clear smooth boundary.
BC	120 - 190	Yellowish, red (5YR 4/8) dry, yellowish red (5 YR 4/6) moist; gritty clay; weak, coarse sub angular blocky; slightly hard, cemented sesquioxdes (Fe, Al, Mn) discontinues over 1m; clay bridges and manganese coats; many, irregular coarse nodules and concretions of black iron manganese throughout the horizon; very few, fine, ransom roots; many fine, moderately porous, inped, interstitial and common, medium, exped, tubular pores; coarse gravelly rock fragments at 80% volume, day wavy boundary.
		Brown (7.5YR 4/4) moist, gritty clay; strong, weak, medium, sub angular blocky; friable, very sticky, very plastic; cemented sesquioxides (Fe, Al, Mn), discontinues over 1m and local discontinues; clay bridges and manganese coats; common very coarse,

irregular, iron manganese with nodules, very few, fine, random roots; many, fine, moderate porous, inped, interstitial and common, medium, exped, tubular pores; Flaggy rock fragments with 80% volume.