PHYSIOLOGICAL AND MORPHOLOGICAL BASIS OF YIELD DIFFERENCE IN UPLAND RICE VARIETIES IN RESPONSE TO LOW NITROGEN IN KIRINYAGA COUNTY, KENYA.

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DECLARATION

This thesis is my original work and has not been presented for award of a degree or any other award in any university.

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DEDICATION

This thesis is dedicated to my late father Benson Kagito and my late mother Margaret Muthoni who had great love for education. May the Lord God rest their souls in eternal peace.

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ABBREVIATIONS AND ACRONYMS

ANOVA:	Analysis of Variance
ASL:	Above sea level
FAO:	Food Agricultural Organization
GOK:	Government of Kenya
IITA:	International Institute for Tropical Agriculture
IRRI:	International Rice Research Institute
KALRO:	Kenya Agricultural and Livestock Research Organization
KARI:	Kenya Agricultural Research Institute
KES:	Kenya Shilling
N:	Nitrogen
NERICA:	New Rice for Africa
NCPB:	National Cereals and Produce Board
NIB:	National Irrigation Board
NUE:	Nutrient Use Efficiency
NRDS:	National Rice Development Strategy
NPK:	Nitrogen-Phosphorus-Potassium
LSD:	Least Significance Difference
MOA:	Ministry Of Agriculture
MOALF:	Ministry of Agriculture, Livestock and Fisheries
MT:	Metric Tones
MWUR:	Mwea Upland Rice
SAS:	Statistical Analysis System
WARDA:	West Africa Rice Development Association

ABSTRACT

Rice is the third most important cereal grain in Kenya after maize and wheat. It has been grown by smallholder farmers as a commercial food crop under irrigated and rain fed ecologies .Low soil N fertility, prevalent in smallholder farmers' fields is a major constraint to upland rice production mostly under rain fed ecology. The ever escalating fertilizer prices have made the input to be unaffordable to most smallholder farmers who are resource poor. Efficient use of nitrogen in agricultural practice can increase yields, decrease production costs and reduce the risk of environmental pollution. The response of rice crop to N-fertilizer have been documented, however the effects of low soil N on yield and quality of rice are lacking. The objective of this study was to evaluate growth and yield of upland rice varieties under different nitrogen fertilizer treatments in order to determine the efficiency of nitrogen fertilizer uptake and identify rice varieties adaptable to low soil nitrogen. A field research was carried out at the experimental farm of Kenya Agricultural and Livestock Research Organization (KALRO- Mwea), Kirinyaga County (0°39′S, 37°20′E).The experiment was split-plot in a randomized complete block design (RCBD), replicated three times. The main plot treatments were five upland rice varieties, MWUR1 (M1), MWUR4 ((M4), NERICA4 (N4), NERICA10 (N10) and IRAT 109. Four N rates, 0, 26, 52 and 78 kg N ha^{-1}, were randomly allocated to subplots. Calcium ammonium nitrate (26%N) was top-dressed in two equal splits at 21 and 45 days after sowing, without P and K application. Soil sampling was done before planting for analysis, to establish the soil nutrient status. The growth parameters determined included plant height, tiller numbers, leaf area, leaf chlorophyll content and stomatal conductance. At maturity, grain yield, shoots dry weight, root dry weight, culm length and panicle length were determined. Plant tissue and grain nitrogen content analysis were also determined by Kjeldahl method. Analysis of variance was performed using Statistical Analysis System version 9.00. Mean separation was done using least significance difference at a 5 % level of probability. Associations between variables were determined by polynomial functions in regression analysis. Although results revealed significant variations due to varieties and N treatments, the interaction between varieties and N treatments were not significant ($P \leq 0.05$) on grain yield. NERICA 4, MWUR 1 and MWUR 4 recorded higher plant height, higher filled grain ratio and higher yield components hence may be suitable for soils with low nitrogen. Across all varieties and N treatments, grain yield correlated positively (($P \leq 0.05$) with total plant shoot dry weight (R^2=0.95). In addition, root dry weight, culm length, number of tillers and panicle length positively correlated with grain yield. There were significant variations (($P \leq 0.05$) in nitrogen partitioning from the soil to the grains under nitrogen rates, unlike in the varieties. The results from this study revealed lack of interactions between varieties and nitrogen rates meaning that the rice varieties did not have synergetic effects on nitrogen uptake and utilization due to incremental nitrogen. Further studies are recommended on nitrogen use efficiency in upland rice under low soil fertility. In addition, cost benefits analysis of low soil nitrogen upland rice production need to be undertaken.

CHAPTER ONE: INTRODUCTION

1.1 Background Information

Rice was introduced in Kenya in 1907 and is currently the third most important cereal crop after maize and wheat (GOK, 2009). It has been traditionally grown by small-scale farmers as a commercial food crop within irrigation schemes, rain fed lowlands and upland ecologies. The main rice growing regions covering all rice production ecologies in Kenya include Mwea in Kirinyaga County, Ahero and Yala swamp in Kisumu and Siaya Counties respectively, Teso and Bunyala inBusia and Kakamega Counties, Kwale and Kilifi Counties, and Tana delta in Tana River County in the coast (Njinju *et al*., 2018). More than 90% of the rice produced in Kenya is cultivated under irrigated lowland ecosystem, while the proportion of rain fed upland rice production is negligible (Musila *et al.*, 2018).

The demand for rice in Kenya is increasing due to changes in eating habits and increasing urban population (Mati *et al*., 2011). Rice production is a strategic industry both as staple food for the country and also as source of employment and revenue earning hence, rice plays an important role as a food security crop in social-economic lives of people and the country as a whole (Shah *et al*., 2018). The demand for fragrance and high quality rice both in international and domestic market is expected to increase due to population growth, higher living standards and health conscious. The National rice consumption is estimated at 530,000 metric tons, compared to an annual production of about 129,000 metric tons (MOALF, 2012). This rice production meets only 26.8% of total domestic demand and is expected to rise with increasing population and change in eating habits (Onyango, 2014).The deficit is met through imports valued at over KES.9 billion per year (FAO, 2016b).

Rice consumption is increasing at a rate of 12% annually and therefore its production must increase to ensure self-sufficiency (Davis *et al*., 2016). Rice production in Kenya is constrained by both biotic and abiotic factors, while consumption can be explained by an increase in population and shift in eating habits making rice almost as frequent as `ugali' maize meal in diet particularly for urban population (Mati *et al*., 2011). To increase rice production, both planting area and productivity (grain yield) per unit area need to be improved. Most of Agricultural environments have low soil fertility and this situation is further complicated by the fact that few farmers can afford the chemical fertilizers whose cost has been rising steadily due to escalating oil prices, hence nitrogen fertilization should be done efficiently (Bowers and Cheshire, 2019). Crop production is highly dependent on supply of exogenous nitrogen fertilizers in most agricultural growing regions. The level of nutrient application by smallholders is low mainly due to the high costs of inorganic fertilizers (Otsuka and Kajirajan, 2006).This has led to dwindling yields as rice continues to be produced on same fields without/or with little soil N replenishment despite massive soil depletion and degradation. The challenge for rice research and development in the world, which includes improvement of the smallholder farmers' welfare and rural employment on a sustainable and economic basis, is to find ways and means of producing more food for the fast growing population with limited land, less water, less labour and even less chemical inputs, as well as to improve productivity (Msangya and Yihuan, 2016).The upland rice yield performance can be increased under ideal environmental conditions by using improved varieties, good management practices, adequate water and nutrient supply according to early research by IRRI (Haefele *et al*., 2016). Nitrogen holds the key to productivity of most cereal crops. It also plays a significant role in crop production and has structural and functional role in relation to plant growth and development (Mengel and Kirby, 1987; Epstein and Bloom, 2005; Taiz and Zeiger, 2010). It is an important component of numerous organic compounds such as amino acids, nucleic acids and proteins (Epstein, 1975). Photosynthetic organs contain a lot of nitrogen and synthesis of enzymes highly requires nitrogen. The low input use and low availability of soil nitrogen constrain rice productivity (Mghase *et al.,* 2010).

Nitrogen is indispensable to plant growth and development, however, its excessive application in rice production is not beneficial from the stand point of agronomy and environment (Leghari *et al*., 2016). Nitrogen fertilizer based pollution is becoming a serious issue for many regions where agriculture is concentrated. To minimize the nitrogen footprint in agricultural production, development of technologies which can allow economically viable production while using less applied nitrogen need to be addressed (Pikaar *et al.,* 2018). Therefore, a balanced and sustainable use of fertilizer is of utmost importance. In recognition of the rising demand for rice, the government is committed to increasing rice production in Kenya. This is in line with Sustainable development goals and Kenya Vision 2030 to provide adequate food and nutrition security and reduce poverty (Charlton, 2016). It is necessary to undertake evaluation studies on low nitrogen responses for different upland rice genotypes in order to determine the efficiency of nitrogen fertilizer uptake and identify upland rice genotypes with desirable characteristics (Leghari *et al*., 2016). This would help develop a sustainable production system that would ensure rice productivity is maintained with less nitrogen fertilization. This will bridge the rice deficit currently being experienced, contribute to food security and offer employment to participants in the rice value chain in Kenya.

1.2 Statement of problem

Global food security is at stake since the demand for rice is exceeding production (FAO 2016). Rice farming in Kenya offers great potential for food security and incomes to subsistence farmers and players along the rice value chain. Despite this, rice production is low and has stagnated for a couple of years due to constraints in its production and climate change related problems (Kundu *et al*., 2017). Rice (*Oryza sativa* L.) is a principal staple food in Kenya; however its production is hindered by abiotic stresses which include drought, low soil nitrogen fertility, lack of irrigation water and cold damage (Atera *et al*., 2018). Nitrogen is usually the nutrient limiting crop production, and the cost of mineral N-fertilizer accounts for a major portion of the total cost of rice production (Guo *et al*., 2018). Fertilizer use efficiency by rice plants is low due to the losses from ammonia volatilization, denitrification, leaching, ammonium fixation and runoff (Savant and De Datta. 1982), giving more importance to economic and environmental issues of nitrogen fertilization. This has resulted into food and nutritional insecurity and low living standards of the rural population.

The rising population and increased food demand require increased production of more rice to meet the food demands and curb food insecurities. There exists a vast potential for production of upland rice in most counties in Kenya. Upland rice varieties with high yield potential are available, though their productivity has not been well documented (MOA, 2011). Mineral fertilizers are expensive and not environment friendly such that when applied in excess, they cause eutrophication of fresh water bodies and coastal water ecosystems (Raven and Taylor, 2003) in addition to increased emissions of greenhouse gases, such as nitrous oxide (N_2O) (Matson *et al*., 1998).Whereas there is reported research in relation to low soil fertility in rice production, there is little work reported on influence of low soil nitrogen in upland rice production. Irrigated lowland rice is constrained by water shortage and Kenya has a potential for upland rice production in about one (1) million hectare of land (MOALF, 2012). Ironically only 449 ha of this land is being utilized at present leaving many areas still unexploited (Diagne *et al*., 2013)

1.3 Significance of the study

In Kenya, rice is the third most important staple food crop after maize and wheat in terms of consumption. However, annual rice production is low at only 129,000 metric tons (MT) against consumption of 530,000 metric tons. This is further compounded by the fact that, of the three grain staples, rice has the highest annual growth rate in per capita consumption at 12% followed by wheat at 4% and maize at 1%. This makes the country import 75% of total rice domestic requirements to meet the demand. Although nitrogen is a macronutrient that is often limiting to plant growth, the level of application by smallholders is low due to the perceived high cost of mineral fertilizers. Food production has to increase to meet the demand of a growing population. In light of the high energy costs and increasing scarce resources, future agricultural systems have to be more productive and more efficient in terms of inputs such as nitrogen fertilizers. Upland rice is unexploited despite the gigantic potential it has in terms of land availability and ease of cultivation which can unlock the current status of deficit rice production (MOALF, 2012).There is limited knowledge on nitrogen fertilizer use by upland rice in Kenya. This study therefore aims at filling scientific knowledge gaps regarding responses of rice to low soil nitrogen as a constraint in upland rice cultivation. The generated information will assist to develop a tailor-made low cost production, upland rice cultivation technology for Kenyan rice farmers while maintaining clean environment for sustainable agriculture, consequently improve food and nutritional security and alleviate poverty to our resource poor farmers.

1.4 Broad objective

The study aimed at evaluating upland rice varieties for adaptability to low soil nitrogen and their growth, development and grain yield in Mwea, Kirinyaga County.

1.4.1 Specific objectives.

i. To evaluate the effects of different rates of nitrogen fertilizer on morphological development and yield of five upland rice genotypes.
ii. To identify upland rice genotypes most adaptable to low nitrogen and morphological and physiological characteristics of upland rice that influence their production.
iii. To determine nitrogen partitioning in rice grains and above ground biomass under different nitrogen application levels.

1.5 Hypotheses

i. Morphological development and yield of upland rice varieties is not influenced by different nitrogen levels.

ii. Low soil nitrogen does not influence physiological processes in rice and hence decrease grain yield.

iii. There is no variation in partitioning of nitrogen between rice grain and above ground biomass under different nitrogen application levels.

1.6 Conceptual Framework

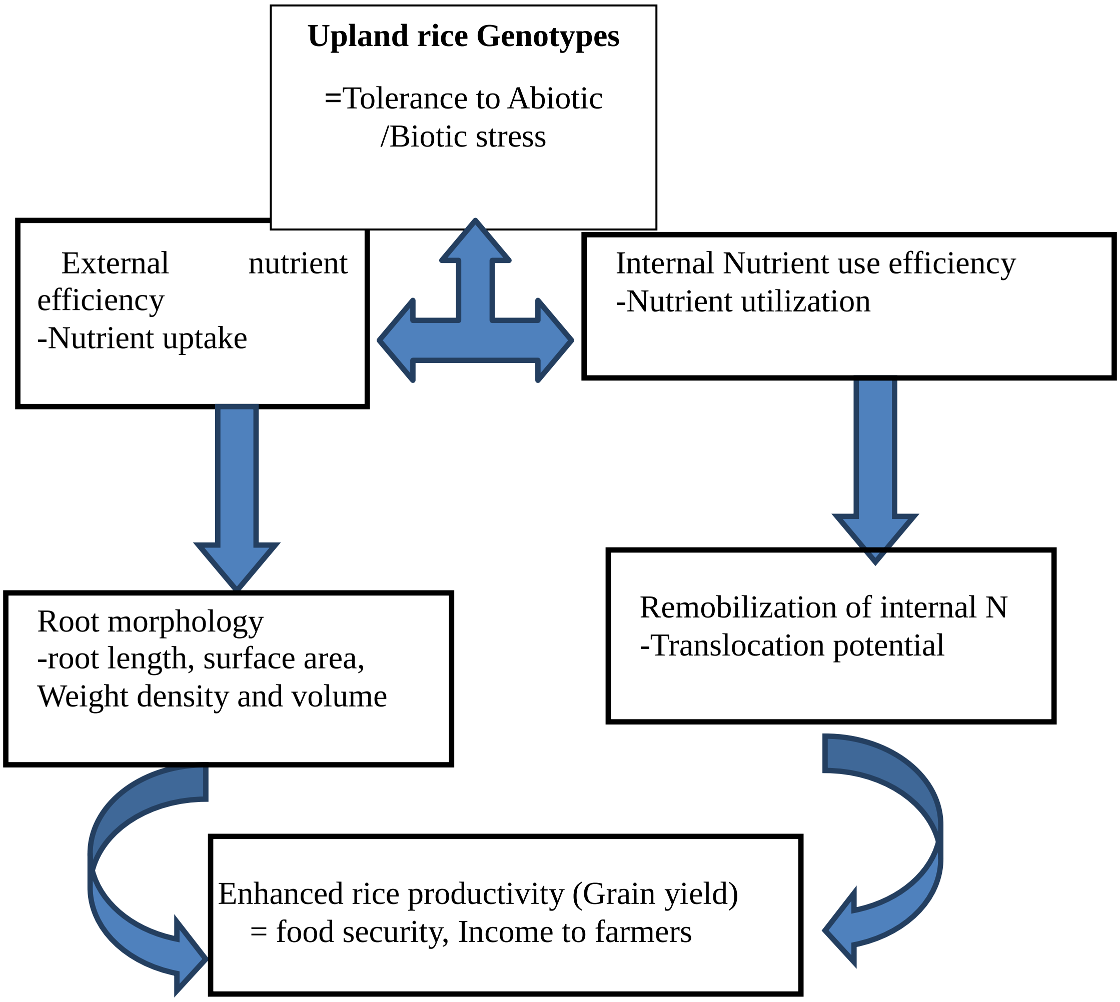


Figure 1.1: Conceptual framework

CHAPTER TWO: LITERATURE REVIEW

2.1 History, origin and distribution of rice

Rice belongs to the genus *Oryza* and family Poaceae tribe Oryzeae. It originated in Asia (*O.sativa* L.) and West Africa (*O. glaberrima* Steud). According to Vaughan *et al*., (2004) the genus Oryza is made up of 23 species, with two species being cultivated (*O. sativa* and *O.glaberrima*), while the other 21 are not domesticated. The *O. sativa* has further three subspecies; indica Kato, japonica Kato and javanica (Roschevicz; 1931). The subspecies japonica has two strains namely tropical and temperate which is commonly sticky rice due to high amylopectin content. Indica are found in tropical and sub-tropical regions while javanica are mainly grown in Indonesia and japonica are found in temperate regions (Chauhan *et al.,* 2017). The largest number of species (9) is found in Africa, which is considered to be the centre of origin of the genus. The two cultivated species are *O. sativa* and *O. glaberrima*.

Rice has been cultivated in South Eastern Asia for millennia and is one of the oldest food crops (Castillo *et al*., 2016). Archaeological excavations in the Indus Valley civilisation of India show evidence of rice grown as early as 2,300 BC and important cereals at Mohenjodero (Grist, 1975) in China. Specimens of rice have been discovered dating from the third millennium BC. However, the land rice is thought to originate from is Java, meaning Island of Rice. Rice has also had a long association with Indonesia, Ceylon and the Philippines (Chauhan *et al.,* 2017).

The most important *Oryza* species are *O. glaberrima,* Steud. and *O. sativa*. *O. glaberrima* was formerly considered to have arisen from the wild species, *O. breviligulata* *(*Nourollah, 2016). However, now, *O. glaberrima* is believed to the descendant of *Oryza* *perennis,* Moench with *O. breviligulata* being a collateral descendant (Grist, *op. cit.*). *O. perennis* was probably the precursor of *O. sativa* in Asia and *O. glaberrima* in Africa (Ren *et al*., 2018). Most modern taxonomists consider *O. sativa* and *O. glaberrima* to be monophyletic in origin and to have a common ancestry. The exact route through which *O. sativa* spread is not clear. Available evidence suggests that it moved from South East Asia, where it originated, to India and Indo-China, and then northwards through Asia, and south and east through the Malay Archipelago as humans migrated and resettled (Lu, *et al*., 2015). The subspecies japonica are commonly sticky rice due to high amylopectin content. Indica varieties have long grains, while japonica have short grains and javanica have broad grains.

2.2 Rice production in Kenya

Rice was introduced in Kenya in 1907 from Asia. Successful growth requires temperature above 21° C, rainfall over 2000 mm/year, flat land and fertile soil (Kimani, 2011). Most Kenyans living in the rural areas consume limited quantities of rice, but forms an important diet for the majority of urban dwellers (NRDS, 2015). According to National Cereals and Produce Board (NCPB), rice is Kenya’s third staple food after maize and wheat and consumption rate is likely to overtake wheat over time. Although Africa produces only 9% of the world’s total rice, it is nonetheless an important staple within individual countries (FAOSTAT, 2017).In Kenya and many other countries in the sub-Saharan Africa, the demand exceeds supply. Local production estimates at 129,000 metric tons while consumption is estimated at 530,000 metric tons (MOALF, 2012). The deficit has been due to increasing tastes, change from traditional foods to the level of commensurate production, increased population, climatic change, presence of blast disease and low yields. It is also gaining popularity due to its easiness and flexibility during preparation as it uses less fuel for cooking (FAO, 2006) especially for labour constrained households. Annual consumption is increasing at a rate of 12% as compared to 4% of wheat and 1% maize (MOA, 2011). To improve the enormous potential of the rice sub-sector, the MOA developed a comprehensive and all-inclusive NRDS, 2008-2018 and MOA strategic plan (2008-2012) both expected to ensure food security, food self-reliance and poverty reduction by employment creation.

Rice production is categorized into three classes depending on the source of water supply for the crop. Irrigated rice is currently the most important type of rice in Kenya. It is mainly produced under irrigation schemes managed by the Kenyan National Irrigation Board (NIB) (Kikuchi *et al*., 2019). About 40,000 hectares are under irrigation with an average national production of 4.5 tons ha^{-1} for the aromatic variety and 6 tons ha^{-1} for the non-aromatic varieties (FAO, 2016; NIB, 2008). The other two types are rain fed lowland rice and rain fed upland rice. There are 13,000 ha under rain fed rice production at Kwale, Kilifi, Tana River Counties at the Coast, Bunyala and Teso in Kakamega and Busia Counties respectively. The average yield for rain fed rice is below 2 tons ha^{-1} (GoK, 2003; Rosemary *et al.*, 2010). There is limited production of irrigated rice while upland rice production is marginally practiced despite its enormous potential in increasing natural production figures.

Production of rice in schemes has remained below projected levels mainly due to technical problems such as expensive maintenance of infrastructure, lack of improved crop varieties, declining soil fertility, pests and diseases (Kimani, 2011).

This in effect has led to reduced rice production causing serious rice deficit. Although there is glaring rice deficit to be made up, not much can be achieved unless the varieties production potential are exploited at low input costs. Little research has been conducted to determine performance of upland rice genotypes at low nitrogen fertility in Kenya. Therefore, future increases in rice production will rely heavily on use of potential high yielding rice varieties.

2.3 Nitrogen requirement of the upland rice

Plant nutrients which primarily come from chemical fertilizers are essential for crop production. Nitrogen holds the key to productivity of all cereal crops (Farooq *et al*., 2017). It is a basic constituent of chlorophyll, proteins and all enzymes are involved in photosynthesis, especially Rubisco which alone accounts for more than 75% of the total leaf N (Hak *et al*., 1993). In rice, up to two-thirds of the N absorbed by the crop, even in fertilized fields, comes from the soil. Natural sources of N, N transformations and availability of N, therefore, markedly influence the fertility of upland soils and the fertilizer N requirement for high yield (Kaur *et al.,* 2017). According to Vinod and Heuer (2012), nitrogen is required in large quantities by rice and N deficiency frequently limits rice grain yields hence nitrogen fertilizers are very important for increasing rice yields.

The availability of nitrogen in the soil varies with different soil types and conditions. Almost all upland rice soils have low N content (Kekulandara, *et al.,* 2019; De Datta, 1981). It help rice plants to grow vigorously, producing green leaves, tillers, and panicles while excessive application of N fertilizer to the soil will extend the vegetative growth; increased the non- productive tillers; lodging; and increased spikelet sterility (Pande, 1994). High nitrogen rates stimulate tillering and the formation of new leaves, causing shading, a condition that favor disease, lodging and reductions in productivity (Zhu *et al*., 2016; IRRI, 1980). The increase in number of upland rice stalks as a response to nitrogen fertilization and greater amount of nitrogen available for the upland rice, will increase tillering and number of panicles. Increasing nitrogen rates reduced the mass of 1,000 grains because the amount of carbohydrates was not sufficient to fill the greater number of spikelets produced (IRRI, 1974). Nitrogen fertilization increased the number of stems and panicles per square meter and the total number of spikelets, reflecting on grain productivity (IRRI, 1974).The cereal crop has different needs for N at different stage of growth. The differences in nitrogen concentration due to nitrogen levels were greatest at panicle initiation stage and started becoming narrower with the advancement in upland rice age (Behera *et al*., 2019). Plants accumulated nearly 15% of the total absorbed nitrogen up to tillering, 50% up to panicle initiation and 85–90% up to heading (FAO, 1994). The total number of spikelets is determined during the upland rice reproductive stage.High nitrogen rates induce the formation of large number of stalks and leaves, creating unfavorable conditions to yielding, such as shading and lodging (IRRI, 1993). Nitrogen applied at panicle initiation increases the number of filled spikelets per panicle (FAO, 1994). Arf, (1993) reported that, upland rice height increased with increasing nitrogen fertilization level.

2.4 Efficiency of N Fertilizer for Upland Rice

Nitrogen fertilizer is a major input for rice, but its uptake efficiency for upland rice is rather low (Amin *et al*., 2016). Upland rice is more likely to absorb nitrogen in the form of ammonium than nitrate (Yamagata, 1998). Nitrogen uptake efficiency of ammonium, aspartate and arginine in upland rice was higher because upland rice could directly take up amino acid nitrogen (Yamagata, 1998). When ammonium nitrate is applied, the upland rice absorbs the ammonium faster than the nitrate (IRRI, 1975). Among the important factors affecting the efficiency of N fertilizer are source, rate and time of application (Tsujimoto *et al*., 2019). The efficiency of N fertilizer is also determined by the forms applied and the agro-ecosystem in which they are used. Synchronizing N application with crop demand and soil N supply is one strategy for improving N use efficiency in upland rice (Getachew, and Nebiyu, 2018). Application of N which are not synchronized with the demand of the plant may result in considerable losses of N and low crop yield.

2.5 Ecological adaptation of rice

Climate directly influences the physiological processes that affect the rice plant's growth, development and grain formation (Arshad *et al*., 2017). Climatic factors which influence the physiological expression of the genetic potential of rice varieties in the tropics are mainly rainfall, temperature and solar radiation. Climate also affects the incidence of insect pests and diseases and hence grain yield (Arshad *et al*., 2017). Rice grows well between 0-2300m above sea level (ASL). Above 2300m, the low temperatures lead to slow vegetative growth and extended periods of flowering. Cold tolerant varieties are necessary for altitudes above 1500m above sea level. Rice requires relatively more water during the times of peak tillering activity, flowering and grain filling stages of growth than any other cereal (Dwivedi et *al*., 2016). Temperatures greatly influence the crop growth duration, pattern and gain yield. Critical temperatures at different growth stages of rice plant have been identified by Yoshida, (1978).

Air temperatures below 20˚ C and above 30˚C markedly affect the growth and yield of rice and the mean minimum temperature should not fall below 20˚C during the rice growing season (Dwivedi *et al*., 2016). Low temperatures at high altitudes injure rice plants, especially between June and July. This lead to poor germination, slow growth, stunted vegetative growth characterized by reduced height and tillering, prolonged flowering period and formation of abnormal grains.

Although there are varietal differences in response to high temperatures, varieties are rarely tolerant to temperatures above 35˚C -- 41˚C (Coast *et al*., 2016). However, such high temperatures are not experienced during rice growing season in most parts of Kenya. Drought is one of the major constraints in rain fed rice production and reduces the response to high levels of inputs and management. Daily rainfall distribution is critical, thus receiving about 200mm of rain over 2 or 3 days in 20 days period may lead to serious moisture stress in subsequent days whilst a rainfall of 100mm well distributed maybe more effective (De Datta and Vergana, 1975). Solar radiation and sunshine hour requirements of the rice crop differ from one growth stage to another (Yoshida and Parao, 1975) and has influence on protein content in the grain (Rao and Deb, 1978). Higher solar radiation is particularly important at the reproductive stage as it increases the number of spikelets and grain yield.

2.6 Role of nitrogen in growth and development of rice plant

The life cycle of the rice plant is completed within 80-120 days. The growth of the rice plant is divided into 3 main phases.

2.6.1 The vegetative phase

Nitrogen is essential for many processes and is crucial for any life on earth. In plants, much of nitrogen is used in chlorophyll molecules which are essential for photosynthesis and further growth (Smil, 2000). Nitrogen is applied to agricultural systems in large quantities and a deficiency leads to yield loss and triggers complex molecular and physiological responses (Banerjee and Roychoudhury, 2019).The vegetative phase consists of four stages: germination and emergence, seedling and tillering stages of growth. The vegetative phase is characterised by active tillering, gradual increase in plant height, and leaf emergence at regular intervals. Tillering may start when the main culm develops the 5th or 6th leaf (Kumar *et al*., 2017). Active tillering refers to a stage when tillering rate is high. The maximum tiller number stage follows active tillering. It is a stage when tiller number per plant or per square metre is maximum (Wang *et al.,* 2016). Tillers developed at early growth stages normally produce panicles (reproductive tillers), while those developed later may not (un-reproductive tillers).

2.6.2 The reproductive phase

The key period for nitrogen absorption by rice plant is from tillering to flowering. Most of the absorbed nitrogen is stored in the leaves and may be transported to the grains during grain filling (Jiang *et al.,* 2004). During plant vegetative stage, meristems and young developing organs need ample supply of nitrogen for synthesis and storage of their amino compounds, which are further incorporated in protein (Concenço *et al.*, 2016). The reproductive phase begins just before or after the maximum tillering stage depending on the variety and the environment. The reproductive stage is marked by the initiation of a panicle primordium of microscopic dimensions in the growing shoot. This takes place 40-60 days after emergence depending on the duration of the variety (Jagadish *et al*., 2015). The reproductive growth stage is characterised by culm elongation, decline in tiller number and emergence of the flag leaf, booting, heading and flowering. Initiation of panicle primordia usually dates back to about 30 days before heading. Panicle primordia can be recognised only under a microscope. Booting is the later part of the panicle development stage (Jagadish *et al*., 2015). About 16 days after visual panicle initiation, the sheath of the flag leaf swells. This swelling of the flag leaf sheath is called booting. The booting stage is followed by the emergence of the panicle (heading or exertion) out of the flag leaf sheath. Anthesis (blooming or flowering) begins with protrusions of the first dehiscing anthers in the terminal spikelets (Concenço *et al.*, 2016). Flowering begins at the top of the panicle. It takes 7-10 days for all spikelets on a panicle to complete anthesis. Anthesis normally occurs between 0800 and 1300 in tropical environments. Fertilization is completed within 5-6 hours.

2.6.3 Ripening phase

The ripening phase lasts from heading to maturity. A 120-day variety, when planted in a tropical environment, spends about 60 days in the vegetative stage, 30 days in the reproductive stage, and 30 days in the ripening period. Ripening follows fertilisation, and may be subdivided into milky, dough, yellow-ripe and maturity stages (Wang *et al.*., 2017). In the milk stage, the contents of the caryopsis are first watery, but later turn milky in consistency. In the dough grain stage, the milky portion of the grain turns first into soft and later hard dough. In the mature grain stage, grain colour in the panicles changes from green to yellow.

2.7 General nitrogen dynamics in rice

Nitrogen is an important component of numerous organic compounds such as amino acids, nucleic acids and proteins (Bloom, 2015; Epstein, 1975). It is one of the main factors involved in productivity and in improving the nutritional quality of rice grain (Fageria and Barbosa, 2001). This nutrient when over produced in the vegetative stage and between the neck-node differentiation and spikelet differentiation stages, absorbed and mobilized, induces increased levels of protein instead of carbohydrates, resulting in excessive growth of shoots, affecting negatively the root system, degree of lodging, spikelet fertility and lowering the resistance of plants to water deficiency (Marschner,1986; Matsushima, 1980).

The beneficial effect of nitrogen occur by influencing yield components such as number of panicles per unit area, number of spikelets per panicle, spikelet fertility and 1000 grain mass and panicle length (Fageria and Baligar, 2001; Fageria and Barbosa 2001), which are controlled by genetics factors of the plant and environmental factors (Freitas *et al.*, 2001). In rice, there are frequent variations within species, regarding the use and accumulation of nitrogen and its genetic control (Ferraz junior *et al.*, 1997; Freitas *et al.*, 2001; Fageria and Baligar, 2001).

These genotypic differences help in the adaptation of species and cultivars to various environmental stress conditions and form the basis for genetic improvement programs (Fageria and Baligar, 2001). The cultivation of rice genotypes with efficient nitrogen use in combination with correct nitrogen fertilization is a promising strategy for increasing yields of upland rice, reducing production costs and environmental impacts (Fageria and Baligar, 2005). The dynamic nature of nitrogen and its propensity for loss from soil-plant systems create a unique and challenging environment for its efficient management. Crop response to applied nitrogen and use efficiency are important criteria for evaluating crop nitrogen requirements for maximum economic yield (Dwivedi *et al.*, 2016).

Low recovery of nitrogen in annual crop is associated with its loss by volatilization, leaching, surface runoff, denitrification and plant canopy (Fageria and Baligar, 2005). Improving nitrogen use efficiency is desirable to improve crop yields and reduce cost of production. Development of nutrient-efficient rice varieties requires a holistic approach combining optimum fertilizer management with enhanced nutrient uptake through a vigorous root system (Maresma and Ketterings, 2017). Plant deficient with nitrogen have stunted growth, depending on severity of the deficiency. Leaf growth is inhibited, where young leaves are inhibited in particular. Plants have developed mechanisms to nitrogen deficiency which include hormonal up regulation of root growth and closing of aqua pores which result in shoot water stress and stunted shoot growth (Mitra, 2015). Nitrogen metabolism and cellular carbon must be tightly coordinated to sustain optimal growth and development for plants, and other cellular organisms (Jiang *et al.,* 2018). Plants are non-motile organisms and therefore through evolution they have developed the complex sensing and signaling mechanisms to robustly monitor and appropriately respond to the dynamic changes of their surrounding environments (Jubair, 2015).

2.7.1 Importance of Nitrogen in rice

Nitrogen is the most abundant mineral element in plant tissues which is derived from the soil and is generally needed in most rice soils (Sharma, 2016). The majority of plant usable nitrogen is consumed as nitrate (NO_3) and as ammonia (NH_4^+) (Huang *et al*., 2000). However, it is the most limiting nutrient for rice production (Helms and Slaton, 1996; Linscombe *et al*., 1999; Miller and Street, 2000). Nutrient efficiency has been widely used as a measure of the capacity of a plant to acquire and utilize nutrient for biological and grain yield (Kimani, 2011), however little information is available on the effects of deficiency and excess amounts of nitrogen on the shoot and root growth, biomass production and chemical composition in rice plants. Nitrogen-use efficiency (NUE) has been defined as the amount of biomass produced per unit of N applied. Fageria and Baligar (2003) and Singh *et al*.(1998) further defined nutrients use efficiency as the maximum economic yield produced per unit of nutrient applied, absorbed or utilized by the plant to produce grain and straw.

In cereals N recovery efficiency at global level is reported to be less than 40% (Raun and Johnson, 1999, Raun *et al*., 2002). High NUE is generally important under conditions of low soil N availability, as it entails high biomass production per unit of N uptake (Aerts and Chapin, 2000) and it is a useful index that combines both plant physiological and morphological responses along nutrient availability gradients. Unlike nutrients such as P, K, and zinc (Zn), no suitable soil test method has been established and implemented for determining the N-supplying capacity for soils used to produce rice (Dobermann and Fairhurst, 2000). Instead, numerous N rate and application timing studies are conducted on experiment stations and farms to determine the optimum N rate for the various cultivars that are grown in the rice-producing regions. Nitrogen is crucial for plants to perform the routine and fundamental cellular activities. The nitrogen nutrient includes inorganic compounds (Nitrate and Ammonium) and the organic compounds (all amino acids) which are synthesized by incorporating ammonium into C-skeletons (Erdal, 2019). Amino acids and the resulting proteins are the key building blocks of the cell. Both carbon and nitrogen nutrients are essential for various cellular functions and therefore adequate supply of these two nutrients are critical for plant growth, development and response to a wide array of stresses and ultimately for completion of life cycle and production of harvestable organs (Shahzad *et al*., 2018).

Nitrate (NO_3^-) and /or Ammonium (NH_4^+) are usually taken up by the root system and transported to the leaf while carbon assimilation and metabolism primarily occur in the leaf (Huang *et al*., 2018). Physiological and biological studies have concluded that when plants are deficient in nitrogen, the photosynthesis output was negatively affected which can then be recovered if nitrogen is provided back to the growth media or the soil (Coruzzi *et al*., 2001).Therefore, plants must develop a mechanism to sense the status of nitrogen in the root system and the surrounding soil environment and coordinate with the sensory machinery in the leaf where photosynthetic output will be determined.

Nitrogen increases plant height, panicle number, leaf size, spikelet number, and number of filled spikelets (Dobermann and Fairhurst, 2000), which largely determine the yield capacity of a rice plant. Panicle number is largely influenced by the number of tillers that develop during the vegetative stage. Spikelet number and number of filled spikelet are largely determined in the reproductive stage (DeDatta, 1981). Sufficient nitrogen during establishment and tillering ensures adequate tillers per unit area while sufficient N prior to and during panicle initiation ensures adequate panicle size. (MoA, JICA, 2011). Hence, Nitrogen is the most limiting nutrient for rice production in Kenya.

2.7.2 Enhancing Nitrogen use efficiency in rice.

Nitrogen is the most limiting nutrient for crop production in many of the world’s agricultural areas and its efficient use is important for economic sustainability of cropping systems (Sharma, 2016). Nitrogen is applied to agricultural systems in large quantities and deficiency leads to yield losses and triggers complex molecular and physiological responses (Kumar *et al*., 2019). Global food security is at stake since demand for rice has exceeded production (FAO 2017a). Poor and declining soil fertility remains the most important biophysical (abiotic) stress that accounts for the decline in agricultural productivity particularly in rice growing environments in sub-Saharan Africa (Abe *et al*., 2010).

Achieving and sustaining optimal yield in upland rice is a continuous challenge in agricultural systems which are already one of the major forces causing global environmental degradation (Foley *et al*., 2005).The dynamic nature of ‘N’ and its propensity for loss from soil-plant systems creates a unique and challenging environment for its efficient management. Low recovery of nitrogen in annual crops is associated with its loss by volatilization, leaching, surface run-off, denitrification and plant canopy (Abe *et al*., 2010). This is not only responsible for higher cost of crop production but also for environmental pollution, hence improving ‘N’ use efficiency (NUE) is desirable to improve crop yields, reducing cost of production and maintaining environmental quality (Pothstein, 2007). There is need to increase crop productivity in order to meet the growing population. The use of agricultural inputs particularly fertilizers are costly to farmers and the environment (Abbasi *et al.,* 2012). Addition of N fertilizer is typically the single highest input cost for many crops and since its production is energy intensive, the cost is dependent on the price of energy (Pothstein, 2007). From the standpoint of agronomy, nitrogen use efficiency is defined as the ratio of grain yield to nitrogen applied (Chen *et al.,* 2013; Abbasi *et al*., 2012).Today, nitrogen and other nutrients are used inefficiently in most of the world’s agricultural systems resulting in enormous and largely unnecessary losses to the environment. As nitrogen moves through the environment, the same nitrogen atom can contribute to multiple negative effects in the air, on land, in fresh water and marine systems and on human health (Abbasi *et al.,* 2012). Nitrogen emissions to the air, notably those of nitrous oxide are contributing to climate change. The application of nitrogen fertilizer in excess levels reduces nutrient use efficiency (NUE), resulting in reduction in crop yield per unit fertilizer applied. Since nitrogen is prone to leaching, water management in rice production is also critical for high nitrogen use efficiency (Zabarth *et al*., 2009). Rationalizing fertilizer application is an important aspect for sustainable agriculture because it can reduce the negative effects of farming on the surrounding environment (Zabarth *et al*., 2009). Therefore, to minimize the loss of nitrogen, reduce environmental pollution and decrease input cost, it is crucial to develop crop varieties with higher nutrient use efficiency.

Plant breeders, are developing rice cultivars that can exploit nitrogen more efficiently and make more economic use of the absorbed nitrogen (Cormier *et al.,* 2016). Nitrogen use efficiency can be defined as the ratio of grain yield to nitrogen supplied, which is affected by both nitrogen uptake efficiency and physiological nitrogen use efficiency (PE) (van Bueren *et al*., 2017). Nitrogen uptake efficiency is N uptake relative to the supply and physiological N use efficiency represents grain yield or plant biomass relative to N accumulation (Moll *et al*., 1982). Since carbon and nitrogen are required as key substrates during the grain filling period, there must be certain relationships among grain yield, physiological nitrogen use efficiency (PE), accumulation and redistribution of carbon (C) and nitrogen (N) during different development stages in plants (Cormier *et al.,* 2016).. Lodging is a major constraint limiting rice yield and quality due to bending and/or breaking of stems on rice (*Oryza sativa* L) production.

According to Zhang, (2016) excess nitrogen fertilization causes lodging. Nitrogen management therefore, is one of the most common and efficient methods applied to mitigate the negative effect (Foulkes *et al.,* 2011) Plant breeders have reduced lodging risk by introducing dwarfing genes to decrease plant height which is negatively correlated with lodging resistance; however height reduction could reduce the photosynthetic capacity resulting in yield reduction (Allan, R.E., 1986).According to Fageria *et al*. (1997) and Fageria (2007), rice yield is determined by yield components and associated characters and was highly correlated with panicle number and grain harvest index. The number of tillers produced ascertains panicle number and is an important factor in high grain yield (Garba *et al*., 2013). Moore *et al*. (1981) reported that biomass production and nitrogen uptake followed similar patterns, nitrogen was remobilized from the straw to grain until grain formation was complete. Moe and Ohira (1981) also reported that a major proportion of nitrogen was redistributed from vegetative organs to panicles during grain filling.

2.7.3 Soil conditions and Nitrogen availability

Nitrogen is one of the most widely distributed elements in nature and atmosphere. The soil accounts for only a small fraction of the lithosphere N and of this N, only a very small proportion is directly available to plants in the form of NO_3^- and NH_4^+ ions (Zabarth *et al*., 2009). Nitrogen is a very mobile element circulating between the atmosphere, the soil and living organisms (Mengel and Kirkby, 1996). Inorganic N exists in the form of NO_3^-, NH_4^+, NO_2^-, NO and elemental nitrogen, while the organic form includes protein, amino acids, amino sugars and other NO_3^- and NO_2^- , which is produced from aerobic decomposition of organic matter or addition of fertilizers (Garba *et al*. 2013). Gaseous N_2, N_2O and NO are forms of N lost through denitrification (Tisdale *et al*. 1993) thus making Nitrogen unique. Plants absorb N as both NO_3^- and NH_4^+. The age and type of plant, the environment and other factors determine preference of plants for either NO_3^- or NH_4^+. (Gicheru, 2012)

2.7.4 The role of N in plants and its availability in the soils

Nitrogen is an essential element for rice growth and metabolic processes (Noor, 2017). Photosynthesis is the most important source of energy for plant growth (Baker, 2008) because chlorophyll represents an important pigment for photosynthesis. Leaf chlorophyll content provides valuable information about physiological status of plants. The optimum temperature of general plant chlorophyll synthesis is 30^0C. Precipitation might affect the photochemical activity of chloroplasts (Zhou, 2003), with water being the medium used for transporting nutrients in plants. Consequently, chlorophyll synthesis and water are closely related. Plants inevitably adjust their own traits to adapt to different environments.Thus, climate and soils play important roles in regulating chlorophyll.

Nitrogen is very dynamic in soil-plant systems and changes with time and space (Garba *et al*. 2013). In well aerated soils of agricultural and natural ecosystems, most of the nitrogen is typically present as nitrate (NO_3) (Sabir *et al.,* 2007). Usually the two N sources co-exist in soils at various ratios and therefore it is not surprising that most plants appear to be able to acquire both NO_3 and NH_4. According to Boxman *et al.,* 1991, oversupply of both NO_3 and NH_4 can significantly reduce the extent of mycorrhizal associations. Nitrogen is the main plant nutrient which limits plant growth (Sabir *et al.,* 2007). It plays a pivotal role in several physiological processes inside the plant. It is fundamental to establish the plant’s photosynthetic capacity (Hageman and Below, 1984), it prolongs the effective leaf area duration, delaying senescence (Earl and Tollenaar, 1997). With the increasing use of fertilizer globally (Brown, 2001), the impact of excess nitrogen in agricultural and natural environments has become a subject of considerable attention (Galloway *et al.,* 2002).

Nitrogen plays a major role in plant biochemistry. It is an essential constituent of cell wall, cytoplastic protein, nucleic acids, chlorophyll and a vast array of other cell components (Sabir *et al.,* 2007). It is involved in all major processes of plant development and yield formation. Besides a good supply of N to plants stimulates root growth and development as well as uptake of other nutrients (FAO, 2000, Brady and Wiel, 2002). Nitrogen affects plant growth and productivity by helping the crop to have a better root growth and establish vigorous root system enabling the plant to mobilze soil moisture and nutrients more efficiently, alter leaf area photosynthetic capacity through increased plant height and girth growth and secure better canopy structure (Devi *et al.*, 2001). Nitrogen also increase shoot dry weight, which is positively associated with grain yield in cereals and legumes (Fageria, 2007). Grain harvest index and N harvest index are also reported to be improved by addition of N to crop plants (Fageria *et al*., 2006). Nutrient uptake in the soil is achieved by cation exchange, where root hairs pump hydrogen ions into the soil through proton pumps. These hydrogen ions displace cations attached to negatively charged soil particles so that the cations are available and taken by roots.

2.7.5 Varietal differences in yield and yield components

Crop genotypes play an important role in crop production systems. They affect crop productivity by their higher yield potentials, resistance against insect pests and diseases under different climatic conditions (Haggag, *et al.*, 2015). Upland rice varieties differ in yields and yield components. As reported by Kimani (2011), there exists genetic variability for N harvest index within the small grain genotypes while a high N harvest index in the genotypes is associated with efficient utilization of nitrogen. Variation in the N harvest index is a characteristic of genotype and this is a useful variable for selecting rice genotypes for higher grain yield (Haggag, *et al.*, 2015).

Genotypes have been observed to vary in their ability to utilize N efficiently and partition N between various rice plant components. Zhi-You *et al*. (2006) and Singh *et al.*(1998) observed great variability in rice for grain yield, N-uptake efficiency and partitioning parameters such as; physiological N-use efficiency (PNUE), agronomic N-use efficiency (ANUE), apparent recovery (AR), partial factor productivity of applied N (PFPN), N productivity index (NPI), and N harvest index (NHI) (Sharma, 2016). They reported the existence of significant differences among genotypes and N-levels for rice total biomass, total N-uptake, tillering ability, panicle numbers and total number of spikelets (m^{-2}). Straw and grain N concentration also vary within rice genotypes. Singh *et al*. (1998) on the other hand, observed that differences in physiological efficiency may occur because of differences in critical concentrations (internal N requirement) for expansion, growth, mass accumulation, organ formation and differences in the ability to translocate, distribute and redistribute the absorbed N in various organs, and the efficiency of N-use in photosynthesis (Sharma, 2016). N-use efficiency though largely dependent on interactions between nutrient balance, water availability, light intensity, disease pressures, and genotypes; can be improved through appropriate genetic manipulation. However, selection to improve the rice crop's N-use efficiency remains viable and one to be exploited because it is sustainable and environmentally friendly.

Saito *et al.*(2006) observed that traditional cultivars were less responsive to N application, where grain yield was 16% with N application (a 0.1–0.4 t ha^{-1} increase in grain yield), while grain yields of improved cultivars were increased by 37% with N application (a 0.9–1.2 t ha^{-1} increase in grain yield). Genotypes have been shown to differ in their nitrogen use-efficiency. Socolow (1999) reported that although N application increases yields, its use is known to have negative impacts resulting from N-compounds on the atmosphere, the ground water, and other segments of the ecosystems. Lawlor (2002) strongly argued that there is need to increase production of food without expansion of agricultural land and with less fertilizer in order to feed the growing population and save fossil fuel energy used in its production. Nitrogen deficiency reduces the % N as leaf: shoot ratio decreases with crops development. Under low N-conditions, rice plants attempt to acquire more N by increasing the root to shoot ratio (Marschner *et al*., 1986). In rice, yield production depends on the number of plants per unit area, tillers per plant, number of spikelets per tiller, grains per panicle and 1000 grains weight. However some of these traits have negative correlation with one another and hence their breeding is not straight forward. Baligar *et al*. (2001) argued that when N-supply is limiting to achieve genetic potential, then it is N-uptake that must be enhanced to obtain greater biomass.

CHAPTER THREE: MATERIALS AND METHODS

3.1 Location and study area description

The study was conducted at the Kenya Agricultural and Livestock Research Organization (KALRO-Mwea) Kirogo farm in Kirinyaga County (Fig 3.1). Kirogo farm is situated at latitude 0^{0} 37’S and longitude 37^{0} 20’ E at an elevation of 1159 m above sea level.

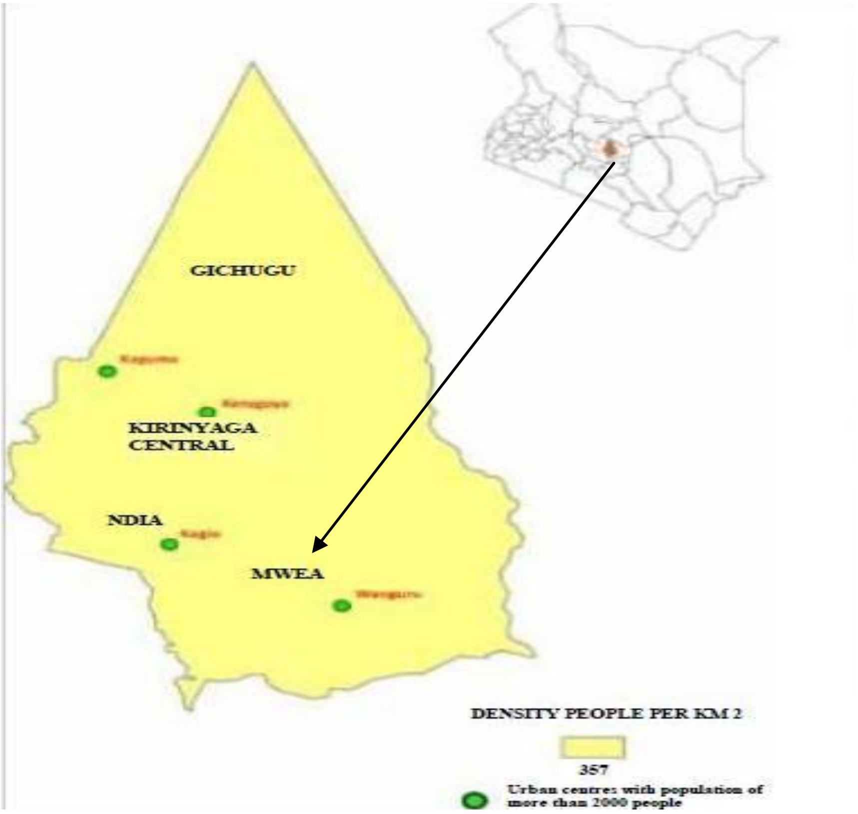


Figure 3.1: Location of the study area

The area receives an average rainfall of about 850mm per annum which is divided into long rains (March to June with an average of 450mm) and short rains (mid October to December with an average of 350mm).The rain is characterized by uneven distribution in total amount, time and space. The area temperature ranges between 15.4^{o} C and 29.5^{o}C with a mean of about 22^{o}C. The soil in kirogo farm, experimental site is a nitosol, which is deep, well drained dusky-red to dark reddish-brown, friable clay with low fertility. The study area is within the Agro Ecological Zone, Lower Medium 3 (AEZ LM3) and experiences a bimodal pattern of rainfall. The soil from kirogo farm, experimental site was sampled at a depth of 0 to 20 cm and analyzed for pH, macronutrients and micronutrients at the National Agricultural Research Laboratories Kabete before planting.

Table 3.1: Soil chemical characteristics at KALRO-Mwea, Kirogo field trial site

Soil Attributes	**Levels**	**Class**
pH	5.84	Slight acid
Total Nitrogen	0.11	Low
Total organic Carbon%	1.09	Low
Phosphorus, P_2O_5 (mg/kg)	310	High
Potassium me%	0.98	Adequate
Calcium me%	2.7	Adequate
Magnesium me%	8.06	High
Manganese me%	0.11	Adequate
Copper ppm	2.65	Adequate
Iron ppm	18.3	Adequate
Zinc ppm	2.27	Low
Sodium me%	0.30	Adequate

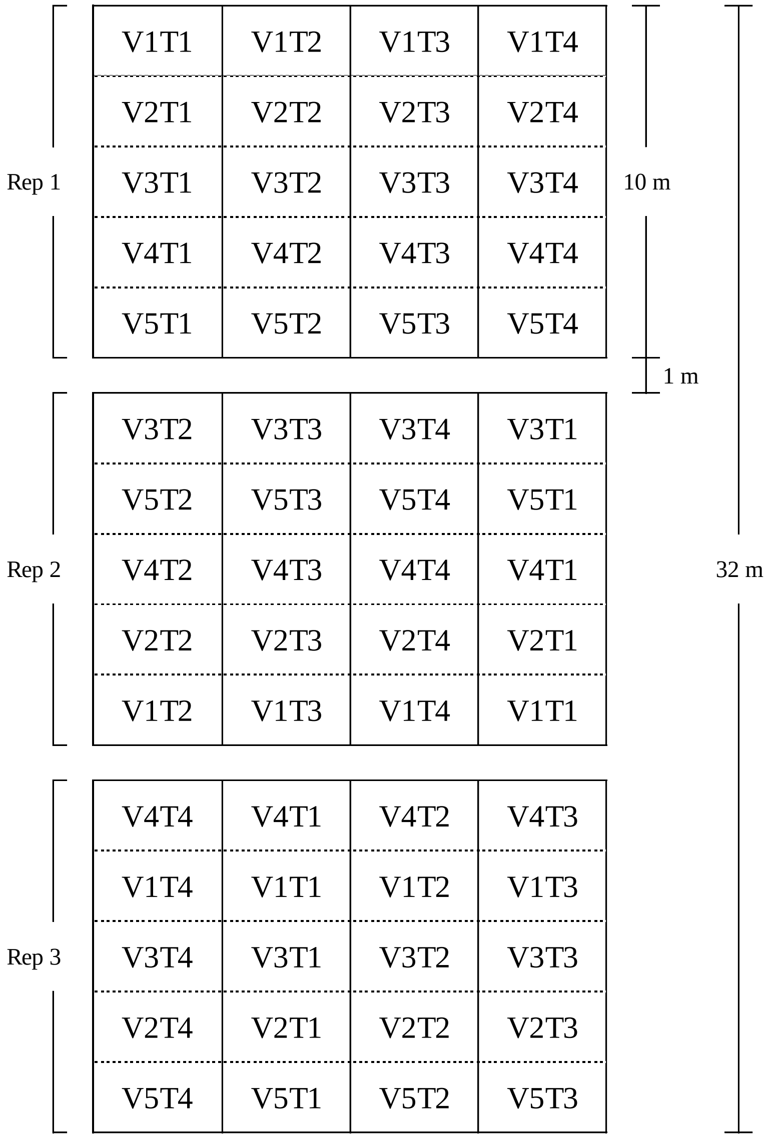
The soil in the experimental field was Nitisol containing 1.09% C determined by wet oxidation chronic acid digestion method, 0.11 total N determined by micro-Kjeldahl method, 310 phosphorus pentoxide (P_2O_5 mg kg^{-1}), 0.98 me% K determined by extraction with 1M ammonium acetate (Ph 7.0) using flame photometer.

3.2 Experiment design and layout

The trial was laid out as Randomized Complete Block Design (RCBD) with spilt-plot arrangement and the treatments were randomly assigned and replicated three times. The main plot treatments were the rice varieties and the sub- plots had the nitrogen rates. Plots measuring 2 m × 1.5 m were marked for planting each of the five upland rice genotypes. The soil was sampled in the experimental field at random for soil analysis. Sunflower which is deep rooted and it extracts soil nitrogen in large quantities from the ecosystem was planted for one season in the experimental field to deplete nitrogen nutrient. Generally sunflower seeds are sinks for plant nitrogen which is accumulated in the endosperm (Krishna, 2010). Another soil sampling was done and analysis performed before planting the trial. The seeds were directly planted in rows. During the laboratory analysis, soil pH and solution were determined using a glass calomel electrode system as described by Crockford and Nowell (1956) while organic matter was determined by wet oxidation chromic acid digestion method adopted from Walkey and Black (1934). The soil N was determined by the micro-Kjedahl method (AOAC, 1995). The soil K, Ca, Mg and Na was extracted with a 1M NH_4AC (pH 7) solution and analyzed with a flame photometer. The soil Mg was determined with an atomic absorption spectrophotometer (Ogunwale and Undo., 1978). The exchangeable acidity (H^+ and Al^{3+}) were measured from 0.1M HCL extractant by titrating with 0.1 M Na OH (Mclean, 1965). The micronutrients Cu, Zn, Mn and Fe were extracted with 0.1 M HCL (Ogunwale and Undo, 1978) and read on a Perkins Klimer atomic absorption spectrophotometer.

Seeds were direct planted at a spacing of 20 cm by 15cm by dibbling (MOA, 2012). Each plot had five rows. Fertilizer (CAN) treatments were randomized in each block. Zinc, a micro-nutrient was blanket applied as zinc sulphate at a rate of 25kg $ZnS0_4$/ha (Africa Rice Centre, 2002). Five upland rice varieties with similar growth patterns, two varieties being popular with farmers were selected and they were chosen at random after a preliminary experiment. Four nitrogen treatments, 0kgN ha^{-1} (control), 26kgNha^{-1}(low rate), 52kgNha^{-1}(standard rate) and 78kgNha^{-1}(high rate) were used. The routine agronomical practices of weeding, pest control and periodic irrigation were applied. The fertilizer treatments were applied to the soil in two equal splits with the first split being applied at tillering stage, 21 days after sowing (DAS) while the second split was applied after 45 days at panicle initiation stage. Nets were installed across the experimental field just before heading stage to prevent rice grains from bird damage.

FIELD LAYOUT



Rep 1

V1T1	V1T2	V1T3	V1T4
V2T1	V2T2	V2T3	V2T4
V3T1	V3T2	V3T3	V3T4
V4T1	V4T2	V4T3	V4T4
V5T1	V5T2	V5T3	V5T4

Rep 2

V3T2	V3T3	V3T4	V3T1
V5T2	V5T3	V5T4	V5T1
V4T2	V4T3	V4T4	V4T1
V2T2	V2T3	V2T4	V2T1
V1T2	V1T3	V1T4	V1T1

Rep 3

V4T4	V4T1	V4T2	V4T3
V1T4	V1T1	V1T2	V1T3
V3T4	V3T1	V3T2	V3T3
V2T4	V2T1	V2T2	V2T3
V5T4	V5T1	V5T2	V5T3

Key:

V1: MWUR1	T1: 0KgN
V2: MWUR4	T2: 26KgN
V3: NERICA4	T3: 52KgN
V4: NERICA10	T4: 78KgN
V5: IRAT 109	

3.3 Genotype description

Table 3.2: Upland rice genotypes description

Rice genotypes	Characteristics/ Attributes
MWUR 1	A Cross between Duorando precose × CT16317-CA-4-M (CIAT upland rice) (Kimani, 2011) -high yielding, drought tolerant, early maturing, tolerant to low soil fertility, tolerant to pests and diseases (Kimani,2011)
MWUR 4	A cross between NERICA1× WAB880-1-38-20-17-P1-HB(WARDA) (Kimani, 2011) -high yielding, drought tolerant, early maturing, tolerant to pests and diseases
NERICA 4	Developed by the Africa Rice Centre to improve the yields of Africa rice varieties (Moseley et al; 2010). Cultivar line-WAB450-1-B-P91-HB. - Most suitable for most areas in Kenya including Mwea and have wide potential for cultivation in Africa. - Drought tolerant and early maturing.
NERICA 10	Developed by the Africa Rice Centre to improve the yields of Africa rice varieties. Cultivar line-WAB450-11-1-1-P41-HB. (Okeleye *et al.* 2006) - Most suitable for most areas in Kenya including Mwea and have wide potential for cultivation in Africa. - Drought tolerant and early maturing.
IRAT 109	A derivative of IRAT 13and IRAT 10 with medium-long grains. - It is moderately resistant to blast, early maturing and has well developed rooting system.

Nerica's are high yielding and stress tolerant upland rice varieties developed for Africa to address the continental-wide rice cereal challenge, poverty and food insecurity (FAO/SAA 2008). The NERICA varieties have good agronomic performance and resistance to Africa's harsh conditions Nitrogen fertilizer treatments used in the experiment were 0 kg ha^{-1}, 26 kg ha^{-1}, 52 k gha^{-1} and 78 kgha^{-1}

3.4 Soil sampling and analysis

Soils were randomly sampled at 0-20 cm depth using an auger, mixed thoroughly and sub sampled. The sub-sample was air-dried, ground and sieved using a 2.0 mm sieve. The samples were thereafter kept in well labeled plastic container for further analysis. The soil pH was determined using pH water (ASTM, 1995). The pH water samples were shaken for 10 minutes and the samples were left for 24 hours before measured using standard pH meter. Organic matter was determined by wet oxidation chromic acid digestion method (Walkey and Black, 1934).The total nitrogen in the soil was determined using Kjedahl method (Bremmer, 1960). The soil sample was ground and sieved through a 1.0 mm sieve sieze and 10 g of soil was weight and place into digestion tube. 7 ml of concentrated Salicylic acid and 3 ml Sulfiric acid added to soil sample in digestion tube. The nitrogen in the solution was determined using manual method.

Soil potassium (K), calcium (Ca), magnesium (Mg) and sodium (Na) was extracted with a 1M NH_40 pH7 solution, and analyzed with a flame photometer. Mg was determined with an atomic spectrophotometer (Jackson, 1978).The exchangeable acidity (H+ and Al^{3+}) were measured from 0.1M HCl extractant by titrating with 0.1 M Na0H (Mclean, 1965). Micronutrients Cu, Zn, Mn and Fe were extracted with 0.1M HCl (Ogunwale and Undo, 1978) and read on a Perkins klimer atomic absorption spectrophotometer.

3.5 Sampling and Data collection

3.5.1 Plants sampling and procedure

Plants for data collection were selected and marked randomly. Within the five rows in a plot, the outer rows were not considered while sampling. The random sampling method was followed where five plants were selected in the sampling area and they were tagged in each plot for field data collection.

3.5.2 Data collection

Data collection commenced one month after sowing and continued after every two week until the crop attained full maturity. The data was recorded on a designed data sheet.

3.5.2.1. Tiller counts

The number of tillers per plant was determined by counting with a tally counter.. Tiller counts were carried out at an interval of 30, 45, 60 and75 days after sowing.

3.5.2.2 Plant height

The plant height (from the ground to the tip of the panicle) was taken by measuring the main tiller from the base to the tallest fully formed leaf using a meter rule at interval of 30, 45,60 and 75 days after planting.

3.5.2.3 Stomatal conductance

Stomatal conductance was measured using leaf porometer. It is measured in mmol per square meter per second and it measures the rate of passage of carbon dioxide

(CO_2) entering or water vapor exiting through the stomata of leaf. A measure of between 300-700 mmol m^{-2} s^{-1} is appropriate in irrigated trials. Stomatal conductance is an important trait responsible for the genotypic difference in gas diffusion for photosynthesis and transpiration in rice (*Oryza sativa* L.) Stomatal functioning plays important roles for two plant processes, transpiration and photosynthesis. Stomatal conductance is measured in mmol m^2 s^{-1}, which is a measure of rate of postage of carbon dioxide (CO_2) entering or water vapour exiting through the stomata of the leaf. A measure of between 300-700 mmol m^2 s^{-1} is appropriate for irrigated rice trial while a conductance measurement of between 80-300 mmol m^2 s^{-1} indicate stressed trials. Stomatal pores in the epidermis provide gates for two very important plant processes, photosynthesis and transpiration. Since the majority of leaf nitrogen is contained in chlorophyll molecules, there is close link between leaf chlorophyll content and leaf nitrogen.

In rice (*Oryza sativa* L) leaf photosynthesis is known to be highly correlated with stomatal conductance, however, it remains unclear whether stomatal conductance dominantly limits the photosynthetic rate. Leaf photosynthetic potential is a major determinant of yield potential in rice. In principle, increase in stomatal conductance regulates gas exchange (CO_2 and water) allowing plants to increase their carbon dioxide uptake and subsequently enhancing photosynthesis. Stomatal conductance for gas diffusion and transpiration is closely correlated with leaf photosynthesis in rice (Kuroda and Kumura, 1990, Miah *et al*., 1997, Kanemura *et al*., 2005). High stomatal conductance is responsible for high rate of leaf photosynthesis according to Taylaram *et al*. (2011).

3.5.2.4 SPAD reading

SPAD value indicates chlorophyll contents and color of the crop. The leaf chlorophyll content provides valuable information about physiological status of plants. The Chlorophyll levels were established by recording readings from the leaf blade of the main tiller at three points of the top most fully opened leaves using a SPAD meter.

3.5.2.5 Leaf area

Leaf area was taken by measuring the length and width of the first, middle and last leaves of the sample plants and their average used to calculate leaf area index (LAI). Leaf area index is a measure of the size of photosynthetic machinery of a plant.

LAI=LxWxNx0.72/A, where;

L=Length, W=Width, N=Number of leaves/plant, A=Area coved by plant, 0.72=Constant for determination of LAI of rice (Watson, 1995). This was to understand the radiation captured along the plant stand and canopy under different nitrogen rates and variety. Since Leaf Area Index is the efficiency of photosynthetic process and photosynthetic surface (Lockhart and Wiseman, 1988), it is therefore important in determining plant productivity.

3.5.2.6 Yield and yield components

Harvesting was done at the appropriate time (40 days after heading). This was when 80% of the grain had reached hard dough stage. At maturity, all panicles per hill of the selected plants were harvested and threshed. Filled grains were separated from unfilled grains and counted. The filled grains were weighed in grams and one thousand grain weight was measured. Data on yield and yield related parameters were recorded during and after harvesting. Measurements of the main tiller from the ground to the neck and then to the tip of the panicle was taken using a ruler to obtain the culm and panicle lengths respectively. The number of hills and panicles harvested in each plot were counted, recorded and put together for processing. Panicle weight, thousand grain weight and grain yield were adjusted to 14% moisture content. Flowering dates were recorded when 50% plants started to flower.

3.5.2.7 Shoot biomass

The straw from the harvested plant samples were tied together and oven dried for 24 hours at 72^{o}C. The above ground biomass was determined by weighing the oven dried straw of the samples and the weight recorded.

3.5.2.8 Roots measurement

Roots play important functional role in plant adaptation to stress. Root lengths and mass after harvest was collected from the five selected plants in each plot. The root system was extracted using a monolith stainless open ended metallic cylinder of 40 cm length and 15 cm diameter. The collected roots were washed free of soil using clean running water. The cleaned root samples were stored in FAA (Formalin: Acetic acid 70% Ethanol 1:1:18 ratio by volume) put in plastic bags and well labeled. Each sample was then cut into small pieces using a pair of scissors and spread evenly on a glass tray. Distilled water was applied to the cut root pieces and spread evenly. For total root length measurements, root samples were spread on transparent glass tray without overlapping. Digital images were taken using an Epson scanner (ES 2200) at 300dpi resolution. The total length of root samples was measured using WinRHIZO software V 200 7d (Reagent Instruments Quebec Canada). The root sample was there after dried in an oven at 72^0C for 24 hours. Using an electric weighing balance, the dried root sample was weighed and the root dry weight recorded.

3.6.8 Seed and plant tissue analysis

Five plants (hills) were sampled from each plot, pooled and dried at 70^{ρ} c for 24 hours. The dried plant parts were chopped and ground to powder using a Heiko vibrating mill. For uniformity of sampling, only leaves and stems were included in plant tissue sample. N content (%) was separately analyzed from the straw and seeds in each treatment for all genotypes after harvest.

To determine nitrogen content from plant tissue, protein analysis procedure (Kjeldahl Method) AOAC-12th Edition was used. The sample was digested with sulphuric acid and a powdered catalyst. The digest was diluted with distilled water and the acid neutralized by an alkali containing sodium hydroxide. The ammonia formed was distilled into boric acid solution containing a mixed indicator (methyl blue, methyl red and thymo blue). Thereafter, the liberated ammonia was titrated with standard hydrochloric acid.

$$\%N = NHCL \times \frac{\text{corrected acid volume}}{\text{Gm of sample}} \times \frac{\text{14gmN}}{\text{mole}} \times 100$$

Since most proteins contain 16%N, a conversion factor of 6.25 was used to convert percent nitrogen to percent crude protein. To determine nitrogen partitioned to stem, leaves and grain the N content obtained was divided by the total amount of N in the whole plant and later converted to a percentage through multiplying by 100.

3.7 Meteorological data

The temperature, rainfall, radiation, sunshine and other weather data was recorded daily using instruments placed at the weather station near the trial site. Annual rainfall for upland rice growth is between 200-400mm and temperature range of between 25^{o} c to 33^{o}c is optimum for normal development of rice. Temperatures greatly influence the crop growth duration pattern and grain yield. Temperature increase of 1^{0} c shortens number of days from sowing to heading by 4 -5 days for some genotypes (Nakagawa *et a*l., 2001). Critical temperatures at different growth stages of rice plant have been identified by Yoshida, 1978. Air temperatures below 20℃ and above 30℃ markedly affect the growth and yield of rice and the mean minimum temperature should not fall below 20℃ during the rice growing season. Low temperatures at high altitudes injure rice plants, especially between June and July. This lead to poor germination, slow growth, stunted vegetative growth characterized by reduced height and tillering, prolonged flowering period and formation of abnormal grains. Although there are varietal differences in response to high temperatures, varieties are rarely tolerant to temperatures above 35˚C – 41˚C. High temperatures during grain filling period has been reported to decrease the grain filling duration, leading to lower grain weight and yield of rice (Chung *et al*., 2006). However, such high temperatures are not experienced during rice growing season in most parts of Kenya.

Table 3.3: Weather data for Kirogo experimental site 2015/16 season

Month	**Rainfall** (mm)	**Rain days**	**Temp. (^{0}C)**	
			Max	**Min**
May,**2015**	84.4	8	27.6	18.0
June	17.4	5	26.3	16.7
July	10.2	1	25.9	15.5
August	9.8	2	26.7	16.0
September	0	0	28.5	16.5
October	70.2	13	29.5	17.6
November	375.3	22	27.3	17.1
December	86.5	8	28.3	15.7
January,**2016**	84.8	7	29.0	16.3
February	23.8	1	31.1	15.4
March	47.3	5	31.2	17.4
April	331.4	16	29.1	16.9

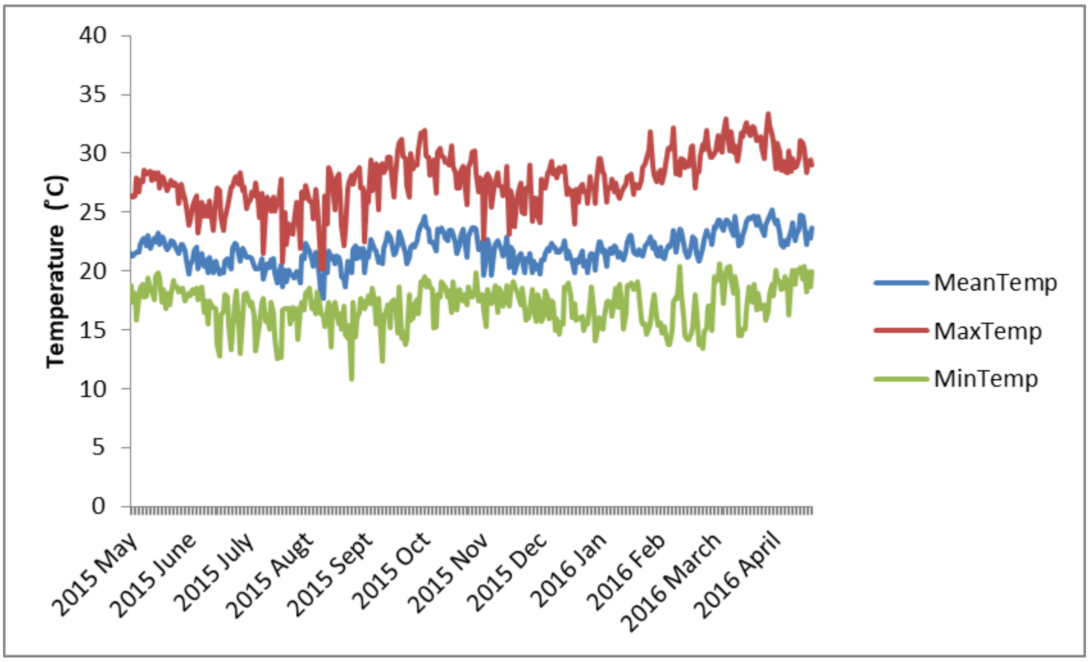


Figure 3.2: Changes in temperature at the experimental site

Temperatures greatly influence the crop growth duration pattern and grain yield. Critical temperatures at different growth stages of rice plant have been identified by Yoshida (1978). Air temperatures below 20℃ and above 30℃ markedly affect the growth and yield of rice and the mean minimum temperature should not fall below 20℃ during the rice growing season. Low temperatures at high altitudes injure rice plants, especially between June and July. This lead to poor germination, slow growth, stunted vegetative growth characterized by reduced height and tillering, prolonged flowering period and formation of abnormal grains.

3.7.2 Solar radiation at the trial site

Although there are varietal differences in response to high temperatures, varieties are rarely tolerant to temperatures above 35˚C – 41˚C. High temperatures during grain filling period has been reported to decrease the grain filling duration, leading to lower grain weight and yield of rice (Chung *et al*., 2006).

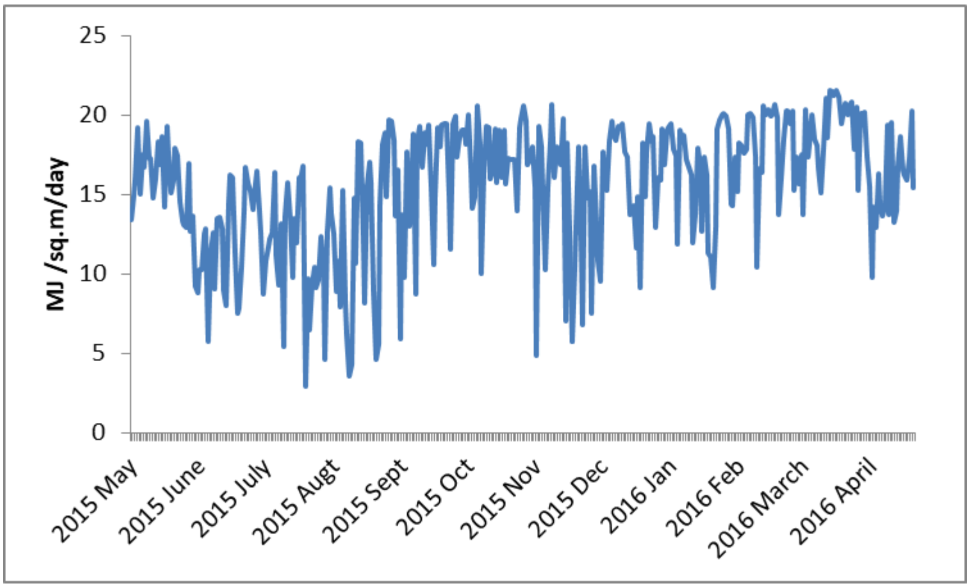


Figure 3.3: Solar radiation at the trial site

However, such high temperatures are not experienced during rice growing season in most parts of Kenya. Solar radiation and sunshine hours requirements of the rice crop differs from one growth stage to another (Yoshida and Parao, 1976) and has influence on protein content in the grain (Rao and Deb, 1978). Higher solar radiation is particularly important at the reproductive stage as it increases the number of spikelets and grain yield.

3.8 Data Analysis

The data collected for different variables measured was arranged and subjected to statistical analysis. Analysis of variance (ANOVA) was performed using general linear model (GLM) of SAS statistical computer package version 9.0 to determine the significance of the effects of nitrogen fertilizer levels on the upland rice varieties. Significantly different means were separated using least significant difference (LSD) at 5 % level of significance. Associations between variables were determined by regression analyses.

CHAPTER FOUR: RESULTS AND DISCUSSION

4.1. Effect of N-level on SPAD values

The effects of N application on leaf chlorophyll content was positively significant ($P \leq 0.05$) with SPAD values increasing with increase in nitrogen level in all the tested rice varieties (Table 4.1).

Table 4.1: Effect of N-level and variety on SPAD value and conductance

	Season 1		**Season 2**	
	Conductance (mmol m^{-2}s^{-1})	**SPAD value**	**Conductance (mmol m-^{2}s^{-1})**	**SPAD value**
Varieties				
MWUR1	333.0^{a}	30.78^{b}	340.1^{a}	38.53^{a}
MWUR4	411.3^{a}	32.75^{ab}	435.1^{a}	38.58^{a}
NERICA4	402.5^{a}	37.01^{a}	350.7^{a}	49.76^{a}
NERICA 10	377.5^{a}	29.82^{b}	402.5^{a}	43.13^{a}
IRAT	385.2^{a}	34.49^{ab}	385.2^{a}	36.62^{a}
LSD $_{0.05}$	**88.1**	**2.50**	**83.3**	**11.3**
N rates kgha^{-1}				
Control	320.3^{b}	30.46^{b}	374.4^{b}	39.68^{a}
26	381.8^{b}	31.82^{b}	381.8^{b}	40.95^{a}
52	348.8^{b}	33.80^{ab}	354.5^{b}	41.85^{a}
78	476.8^{a}	35.80^{a}	474.4^{a}	42.81^{a}
LSD $_{0.05}$	66.81	2.54	64.8	10.48
VxNR	NS	NS	NS	NS

Values with the same letter(s) within the column are not significantly different

This observation was pronounced in season one. The highest SPAD value was recorded in NERICA 4 (37.01 and 49.76 in season one and season two respectively (Table 4.1).In season two the same trend was repeated with NERICA 4 recording higher SPAD value. Under low nitrogen level of 26kgNha^{-1}rice variety NERICA 4 recorded the highest SPAD reading of 38.6 and 40.36 during the first and second season respectively (Appendix 4). Among the different N-levels, significant variation was observed in the SPAD values with the highest at 78kgN ha^{-1} followed by 52kgN, 26kgN and 0kgN ha^{-1} in season one. Although the same trend was observed in season two where N-level of 78kgN ha-1 recorded higher value compared with other treatments, the N-treatments had no significant differences (Table 4.1). Soil-plant analysis development (SPAD) is a tool for predicting grain yield response to nitrogen fertilization. This is a plant based diagnosis which is an important method of determining nitrogen contents of crops.

Photosynthesis is the most important source of energy for plant growth. According to Prasertsak *et al*. (1997) lower chlorophyll content is a symptom of insufficient N supply which is a critical nutrient in crop production. The leaf color of a plant can be used to identify stress level due to its adaptation to environmental change (Singh *et al*., 2002). Several factors can affect chlorophyll synthesis. These include light, carbon dioxide concentration, level of other nutrients, water stress, air temperature and plant density among others.

4.2. Effect of nitrogen levels on stomatal conductance in upland rice genotypes

MWUR 4 and NERICA 4 upland rice varieties had high leaf chlorophyll content and also high stomatal conductance during the two seasons (Table 4.1). In season one and two there was variation in varietal stomatal conductance due to N application, however effect due to nitrogen levels of 0kgN, 26kgN and 52kgN per hectare were not significantly different in all the tested rice varieties. (Table 4.1).

This was consistent with the findings of Maruyama and Tajima (1990) who reported that varietal differences in stomatal pore and guard cell length are limited but that stomatal density and stomatal aperture show clear varietal difference with the rice genotype. Stomatal conductance is an important trait responsible for the genotypic difference in gas diffusion for photosynthesis and transpiration in rice (*Oryza sativa* L.).The interaction between rice varieties and N-level was not significant different during the two seasons. Stomatal functioning play important roles for the two plant processes of transpiration and photosynthesis. Increase in stomatal conductance which regulates gas exchange (Carbon dioxide and water) can allow plants to increase their Co_2 uptake and subsequently enhance photosynthesis. This confirmed Taylaran *et al.,* 2011, findings that, leaf nitrogen content affects photosynthesis which is strongly correlated with stomatal conductance.

Nitrogen is vital as it is a major component of chlorophyll which plants use sunlight energy to produce sugars from water and carbon dioxide. Healthy plants often contain 3-4% nitrogen in their above ground tissues. From the study, both NERICA 4 and MWUR 4 rice varieties were more responsive to nitrogen fertilizer application as the chlorophyll content increased with increase in nitrogen application level (Appendix 4). Rice variety IRAT 109 was not responsive to high nitrogen application level as the leaf chlorophyll content started to decrease after nitrogen application level 52kgN/ha in the first season, however during the second season the difference was not significant. There may be many factors affecting the photosynthesis besides nitrogen availability. The main factors are, light intensity, carbon dioxide concentration, level of other nutrients, water stress and temperature. The chlorophyll content could depend on seasonal and environmental changes.

4.3. Effect of N-levels on leaf area at maximum tillering (40DAS) and at panicle initiation Stage (60DAS).

Leaf area in the two seasons was highly significant ($P \leq 0.05$) due to nitrogen level application at both maximum tillering and panicle initiation stages (Table 4.2). The largest leaf area was recorded in rice variety MWUR1 and IRAT. This was followed by MWUR 4 and NERICA 4 respectively.

Table 4.2: Effect of N-rate and variety on leaf area at maximum tillering and panicle initiation stages

	Season 1		Season 2	
	LAMT	**LAPl**	**LAMT**	**LAPl**
Varieties				
MWUR1	31.19^{a}	34.61^{a}	50.10^{a}	60.09^{a}
MWUR4	29.25^{ab}	32.55^{a}	45.23^{ab}	53.52^{ab}
NERICA4	29.80^{ab}	33.02^{a}	41.46^{ab}	45.44^{ab}
NERICA 10	26.26^{b}	29.40^{a}	33.88^{b}	40.69^{b}
IRAT	34.52^{a}	37.39^{a}	51.98^{a}	57.36^{a}
LSD	5.14	6.85	8.06	10.73
N rates kgha^{-1}				
Control	26.14^{b}	28.34^{b}	34.06^{b}	46.22^{b}
26	28.21^{b}	30.17^{b}	44.38^{a}	51.22^{ab}
52	31.49^{ab}	34.20^{ab}	46.72^{a}	61.66^{a}
78	34.99^{a}	40.87^{a}	52.97^{a}	46.57^{b}
LSD	4.28	5.21	6.95	9.94
VxNR	NS	NS	NS	NS

Values with the same letter(s) within the column are not significantly different

(**LAMT**=leaf area at maximum tillering, **LAPI**= leaf area at panicle initiation)

The lowest leaf area was observed in rice variety NERICA 10 at maximum tillering stage in the two seasons. There was no significant difference in leaf area at panicle initiation stage among all the tested rice varieties in season one (Table 4.2).In season two, significant variation on leaf area was observed with MWUR 1 recording the highest value followed by MWUR 4 and NERICA 4. The lowest value was observed in rice variety NERICA 10 at panicle initiation stage. Significant variation was observed due to N-levels. The biggest leaf area (34.9cm and 52.97cm respectively) was recorded on 78kgN ha^{-1} at maximum tillering stage during both seasonsThe control treatment (0kgN ha^{-1}) recorded the lowest leaf area at maximum tillering and panicle initiation stages during both seasons (Table 4.2). Leaf area is important in determining plant productivity since it is the efficiency of photosynthetic process and photosynthetic surface (Lockhart and Wiseman, 1988). The productivity of a crop depends on the ability of a plant cover to intercept the incident radiation, which is a function of leaf area available.

4.4. Effect of N-level (KgN/ha) on plant height of rice varieties

Plant height was significantly affected by N-level application (Fig. 4.3). Plant height increased with increasing N-level in all tested rice varieties, however there was no varietal difference. The height ranged from 66 cm to 79 cm in the first season and 80 cm to 97cm in the second season. The highest plant height was produced by 78kgN ha^{-1} and decreased gradually with decreasing levels of N-fertilizer application during the two seasons (Table 4.3). Plants receiving 0 kgN ha^{-1} were significantly shorter than other in the different treatments. This trend in plant height confirms the findings of Talukder (1973).

Upland rice variety MWUR 1 recorded the highest plant height (90.4 cm and 97.9 cm in season one and two respectively), indicating its responsiveness to high nitrogen application. It also recorded high plant height (68.9 cm and 85.0 cm) at low N-rate of 26 kgNha^{-1} compared with other tested rice varieties in season 1 and season 2 respectively (Appendix 6). According to Jan *et al*. (2002) plant height reveals the overall vegetative growth of rice crop in response to nitrogen. In the study, nitrogen treatments had significant effect on plant growth. Plant height was significantly affected by different nitrogen levels ($P \leq 0.05$). The increase in height with increasing nitrogen levels is also in agreement with the findings of Ethan *et al.* (2011) who observed that there were significant increases in plant height with increasing N- levels in rice. The superiority in height due to increase in nitrogen rate was possibly due to enhanced rate of translocation of nitrogen from culm to leaves leading to production of photosynthates which enhance translocation of nutrients for developing panicles (Adam *et al*. 2001). These results are consistent with the findings of Manzoor *et al*. (2006) on effect of N-levels on paddy yield and plant height. Plant height is an important morphological character that acts as an indicator of availability of growth resources and it depends on nutrient especially nitrogen (Ferdous, 2001). Thus, a responsible nitrogen application becomes increasingly important for sustainable agronomic production (Zhang, 2015). Nitrogen is a major contributor to crop growth, size and total dry matter production. There was no significant difference in N-level **x** variety interaction in this study. This confirms the results reported by Memory *et al.*, 2013.

4.5. Effect of N-rate (KgN/ha) on rice tillering

The number of tillers per hill significantly increased with nitrogen application, (Table.4.3). However the effects of nitrogen levels, 26kgN/ha, 52kgN/ha and 78kgN/ha were not significantly different. Rice variety MWUR 1 was more responsive to high nitrogen application than other upland rice varieties since it recorded increased tiller numbers at 78kgN (Fig 4.1). IRAT 109 was more adaptive to low nitrogen as it had more tillers at low nitrogen. Upland rice varieties MWUR 4 and NERICA 4 were also responsive to high nitrogen application levels though lower than MWUR 1 on the number of tillers per hill (Appendix 3).

Table 4.3: Effect of upland rice variety and N-rates on plant height (cm) and tiller numbers per plant.

	Season 1		Season 2	
	HT	**TL**	**HT**	**TL**
Varieties				
MWUR1	79.11^a	7.98^a	97.12^a	11.64^a
MWUR4	74.14^a	7.69^a	93.03^a	11.61^a
NERICA4	67.03^a	8.08^a	90.50^a	11.42^a
NERICA 10	71.14^a	7.28^a	82.56^a	12.56^a
IRAT	66.53^a	7.72^a	80.61^a	12.64^a
LSD	9.63	1.86	11.75	2.70
N rates kgha^{-1}				
Control	55.76^c	5.75^c	74.18^c	8.58^c
26	70.73^b	7.61^b	84.59^b	11.78^b
52	78.53^a	8.15^{ab}	92.42^b	13.24^{ab}
78	81.33^a	9.49^a	103.87^a	14.29^a
LSD	5.21	1.30	7.84	1.77
VxNR	NS	NS	NS	NS

Values with the same letter(s) within the column are not significantly different

(**HT**= plant height (cm), **TL**= tillers).

Tillering is an important trait for grain production and is therefore an important aspect in rice yield. According to Singh *et al*. 1972, increase in tiller numbers is due to influence of different fertilizer combinations and more tillers per hill might be due to more availability of nitrogen which plays a vital role in cell division. The results from the study are in agreement with the findings of Chaturvedi, (2005) who reported that fertile tillers per hill significantly increased with increase in N-level. According to Imolehin (1991), growth of tiller buds was determined by genetic factors and the growing conditions hence growth and development of tillers in upland rice depend partly on environmental factors especially radiation, temperature and nutritional conditions. Varietal characteristic is of major significance in the tillering ability of the rice crop.

Tillers produced are a good indicator as it is a major determinant of yield since the number of tillers has been reported to have a positive association with plant biomass and economic yield in rice (Deng *et al*. 2015). Application of N-fertilizer may increase the tiller numbers (Budhar and Palaniappan, 1996), however, not every tiller contributes to high productivity (Sahu *et al*. 2004).Tiller numbers were low in all the varieties in the control during the early growth stage. With little nitrogen application, IRAT had high responsiveness however the tillers declined after some time (Fig 4.1). In other varieties, tiller numbers were not significant different. The results from this study confirm the findings of Budhar and Palaniappan (1996), who reported that nitrogen fertilizer application, may increase the number of tillers in rice.

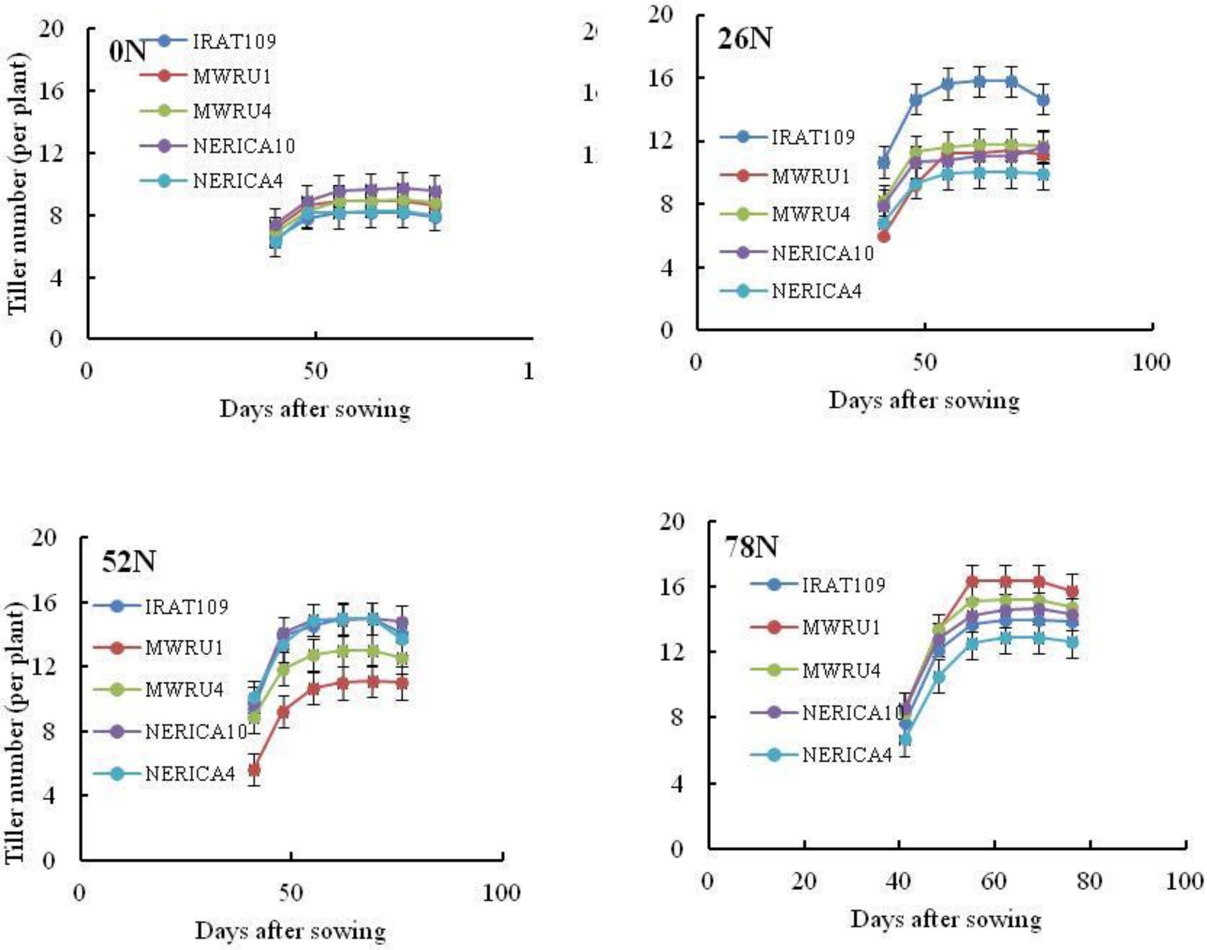


Fig.4.1: Influence of nitrogen levels on upland rice tiller numbers at vegetative growth.

Tillering is an important agronomic trait for rice population quality and grain production (Ling, 2000). They are a form of branching which comprises of protective mechanism for higher plants by assisting with reducing the impact of injury and adaptation to environment (Hervath *et al.*,2003). According to Deng *et al.* (2015), the number of tillers has a positive association with plant biomass and economic yields**.** Rice tillering ability was significantly influenced by different nitrogen levels ($P \leq 0.05$). Rice varieties IRAT 109 and MWUR 4 recorded statistically significant higher tiller numbers at low nitrogen level in season one as compared with others (Fig. 4.1). This could possibly be due to varietal performance difference. Enhanced tillering with increase in nitrogen levels could be attributed to more nitrogen supply to the plant at vegetative stage enhancing cell division. According to Power and Alessi (1978), grain yield of cereals is highly dependent upon the number of tillers per plant which is controlled by genotype and environmental interaction. There was positive correlation between yield components and tiller numbers, hence increasing N-level led to increase of tiller numbers which impacted on yield. The results of this study confirm the research findings of Singh *et al.* 1972.

There was no varietal difference in plant height and tiller numbers in the two seasons. In addition there was no interaction effect between N rate and variety (Table 4.3). The highest plant height was produced by 78kgN ha^{-1} in both seasons. Furthermore plants receiving 0kgNha^{-1} were significantly shorter than those in other nitrogen treatments. This confirms the findings of Talukder *et al*. (1973) who reported on the effects of nitrogen on yield and other characteristics of three rice varieties.

4.6 Rice yield components

4.6.1 Culm length

Culm length and culm stiffness in rice is an important factor determining the nitrogen responsiveness. This is because tall, weak-strawed rice varieties lodge early and severely at high nitrogen levels and lodging decreases the rice yield. In upland rice, culm length is an important morphological character affecting lodging resistance.

4.6.1.1 Effect of N-levels (KgN/ha) on culm length in upland rice

Excess nitrogen causes lodging which is a major constraint limiting rice yield and quality due to the unexpected bending or breaking stems. There was significant effect on culm length ($P \leq 0.05$) due to nitrogen application, with lengths increasing with increase in nitrogen level. Varieties MWUR1 and MWUR4 recorded long culm length (Table.4.4). All upland rice varieties tested showed responsiveness to high nitrogen fertilizer application since culm length increased with increase in nitrogen level in both seasons. Failure to add nitrogen to any of the varieties led to low culm length in the two seasons. Thick culm has more vascular bundles and has fewer tendencies to lodge thus giving better support of panicles and probably a large area for carbohydrate accumulation. According to Tripath *et al.* (2003), culm diameter, wall thickness and dry weight per unit length of basal internodes are positively correlated with lodging resistance. From the study, culm length was significantly different in the tested upland rice varieties due to both variety and nitrogen rates applied. Grain yield eventually indicated significant correlation to the culm length (Fig 4.6).

4.6.2 Panicles

4.6.2.1 Effect of N-rate (KgN/ha) on number of panicles per hill in five upland rice genotypes

The number of panicles per hill was significantly ($P \leq 0.05$) influenced by different nitrogen fertilizer application levels with the control recording the lowest number in both seasons. Among the rice varieties, MWUR1 gave higher values of the number of panicles per plant in season one while MWUR 4 registered the highest panicle numbers per hill in season two and rice variety IRAT recorded the lowest values in both seasons. However, all rice varieties had the same statistical rank (Table 4.4). Rice variety IRAT recorded the lowest value during the two seasons.

Table 4.4: Effect of N-rate and variety on yield components in upland rice varieties

	Season 1			**Season 2**		
Varieties	**Culm length**	**Panicle length**	**Panicle number**	**Culm length**	**Panicle length**	**Panicle number**
MWUR1	70.86^{a}	20.08^{a}	13.81^{a}	75.97^{a}	19.72^{ab}	9.67^{a}
MWUR4	59.81^{ab}	20.61^{a}	11.08^{a}	72.39^{ab}	19.81^{ab}	10.08^{a}
NERICA4	64.22^{ab}	21.11^{a}	13.28^{a}	63.79^{b}	20.01^{ab}	8.39^{a}
NERICA 10	55.81^{bc}	19.31^{a}	11.89^{a}	62.94^{b}	21.47^{a}	9.53^{a}
IRAT	47.11^{c}	17.42^{b}	11.08^{a}	63.78^{b}	18.43^{b}	9.15^{a}
LSD$_{0.05}$	8.41	1.35	3.40	10.93	1.83	2.53
Nitrogen rates kgha^{-1}						
Control	49.58^{c}	18.84^{b}	8.60^{b}	54.53^{c}	16.80^{c}	5.69^{c}
26	57.09^{bc}	19.40^{ab}	11.60^{ab}	70.38^{b}	19.98^{b}	9.03^{b}
52	68.13^{a}	21.04^{a}	14.91^{a}	78.49^{ab}	20.88^{ab}	10.60^{ab}
78	63.44^{ab}	19.53^{ab}	13.80^{a}	83.69^{a}	21.89^{a}	12.13^{a}
LSD$_{0.05}$	8.03	1.42	2.54	7.59	1.01	1.35
VxNR	*	*	NS	NS	NS	NS

Values with the same letter(s) within the column are not significantly different.

There was significant variation in panicle numbers per plant due to N-levels in the two seasons. In season one, high panicle numbers was observed at N-level 52kgN ha^{-1}, which had the same statistical rank as 78kgN ha^{-1}. In season two, the highest panicle number value was observed at 78kgN ha-1 followed by 52kgN ha^{-1}. Control N-level (0kgN) recorded the smallest value during the two seasons. This might be due to the difference in the genetic background among the rice varieties. Nitrogen significantly improved rice yield by improving yield components like panicle numbers. The results of this study confirm the findings of Metwally *et al.* (2017) who reported that high N application improved the number of panicles per plant. Similar results were reported by Abd El Hamed (2002).

4.6.2.2 Panicle length

Panicle numbers per plant and panicle length are important yield parameters as grain yield in rice is a function of panicles area. There were significant difference ($P \leq 0.05$) in panicle length between the varieties in season two, unlike in season one. In addition significant variation in panicle length was observed due to nitrogen levels during both seasons. NERICA 4 recorded higher panicle lengths followed by MWUR 1 and MWUR 4 (Table 4.4). Rice variety NERICA 4 was more adaptive to low nitrogen level application since it had the highest panicle length at 0kgN/ha among all the tested rice varieties in both seasons (Appendix 8). It was also more responsive to high nitrogen application level as the panicle length increased with increase in nitrogen levels (Table 4.4).The highest panicle length was obtained at 52 kgN/ha in the first season and at 78 kgN/ha during the second season. The results confirm findings of Fageria (2007), that nitrogen is one of the most important nutrient in increasing yield components of rice including panicle numbers and a thousand grain weight. The study also confirms the findings of Fageria and Baligar (2001), who reported a significant increase of panicle numbers with increasing nitrogen rates in lowland rice.

These results are also consistent with the findings of Metwally *et al.*, 2011, Metwally (2017) and Yoseftabar (2013) who reported that there was maximum panicle length at high N-level application in rice. The interaction between rice variety and N-level for panicle length trait was significantly different only in season one. The control N-level (0kgN) registered the lowest panicle length in the two seasons (Table 4.4).

4.6.3 Grains per panicle (GPP)

Significant variations ($P \leq 0.05$) on grains per panicle was observed due to rice varieties during the two seasons. The rice varieties differed significantly (Table 4.5). Rice variety MWUR 4 recorded the highest value of grains per panicle followed by rice variety NERICA 4. The lowest value was observed in variety IRAT during the two seasons. Increased N-rate enhanced the number of grains per panicle in all the rice varieties(Table 4.5).The rice varieties differed in their response to N-levels which might be due to source sink interaction meaning maximum proportion of N source was used to produce more spikelets per panicle and grain filling (Noor, 2017).The results are consistent with the findings of Weerakoon *et al.* 2005, who reported that panicle number per square meter and grain number per panicle increased with increasing nitrogen rates.

Table 4.5: Effects of N-rates and rice varieties on other yield components for the two seasons.

	Season 1			**Season 2**		
Varieties	**Filled grain weight(g)**	**Grains per panicle**	**Filled Grain ratio**	**Filled grain weight(g)**	**Grains per panicle**	**Filled Grain ratio**
MWUR1	24.96^{ab}	91.36^{abc}	78.89^{a}	22.36^{a}	91.06^{ab}	91.62^{a}
MWUR4	27.39^{ab}	111.06^{a}	80.43^{a}	24.42^{a}	99.31^{a}	91.99^{a}
NERICA 4	39.69^{a}	96.08^{ab}	83.24^{a}	20.36^{a}	92.43^{ab}	91.53^{a}
NERICA10	22.47^{ab}	71.22^{bc}	78.51^{a}	24.36^{a}	81.29^{ab}	90.54^{a}
IRAT	17.26^{b}	67.75^{c}	62.60^{b}	25.31^{a}	71.13^{b}	80.46^{b}
LSD $_{0.05}$	9.43	19.84	6.61	10.43	18.94	3.12
Nitrogen rates kgha^{-1}						
Control	14.43^{b}	75.93^{a}	72.51^{a}	8.22^{c}	57.29^{c}	86.93^{a}
26	24.15^{ab}	89.31^{a}	74.94^{a}	21.43^{b}	90.16^{b}	89.57^{a}
52	32.29^{a}	93.04^{a}	79.78^{a}	26.36^{b}	91.07^{b}	90.07^{a}
78	27.34^{a}	91.69^{a}	79.69^{a}	37.43^{a}	109.66^{a}	90.34^{a}
LSD $_{0.05}$	7.53	20.88	7.74	4.65	11.28	2.75
VxNR	NS	NS	NS	NS	NS	NS

Values with the same letter(s) within the column are not significantly different

MWUR 4 and NERICA 4 were more responsive to nitrogen fertilizer application levels as they recorded higher grains per panicle in the two seasons which increased with increase in nitrogen level. The results disagree with the findings of Heluf and Mulugeta (2006) who reported that N- fertilizer application reduced the number of grains in a panicle. The highest value of grains per panicle was observed at N-rate of 52kgN ha^{-1} in season one, however all the N-treatments had the same statistical rank indicating no significant difference. In season two, the highest value in grains per panicle was observed in N-treatment 78kgN ha^{-1}. The lowest value was recorded in N-control treatment (0kgN ha^{-1}) in the two seasons. This trait is highly dependent on the genetic makeup of the variety hence the nitrogen influence was possibly limited. Similar results were reported by Abd El Hamed (2002) and Sovour *et al*., 2016. The interaction effect on grains per panicle was not significant for rice varieties and N-levels (Table 4.5).

4.6.4 Filled grain ratio (%)

There was no varietal difference in filled grain ratio during the two seasons (Table 4.5).Rice varieties NERICA 4, MWUR 1 and MWUR 4 recorded higher filled grain ratio in season one. The same trend was repeated in season two, however, all the rice varieties had the same statistical rank indicating no significant difference in filled grain ratio. Rice variety IRAT recorded the lowest value in filled grains ratio during the two seasons. Rice varieties MWUR 1, MWUR 4 and NERICA 4 were adaptive to low nitrogen application level, hence had higher filled grains ratio at low nitrogen levels in both seasons (Appendix 7).

4.6.5: Thousand grains weight

Thousand grain weight is a parameter used to measure grain quality and determine grain yield in rice. High nitrogen rates reduce the mass of thousand grains weight since the amount of carbohydrates is not sufficient to fill the greater number of spikelets produced (IRRI, 1974). There was significant variation on thousand grain weight on rice varieties with no difference due to nitrogen levels. MWUR 1 rice variety recorded higher weight at no added nitrogen application in the two seasons compared with the other tested varieties. In addition, this variety was not responsive to high nitrogen application since the weight did not change with change in N rates (Fig 4.2 ii). This implies that MWUR 1 is more adaptive to low nitrogen level. On the other hand, IRAT 109 responded to high nitrogen application, although insignificantly. A thousand grain weight is an important parameter among the yield controlling characters and it was not significantly affected by N-rates in this study. The results confirm previous studies on upland rice in Nigeria, which indicated no significant influence of nitrogen in grain size (Oikeh *et al*, 2008). However, this is in contrast with the results of Fageria and Baligar (2001) who reported that weight of thousand grains increased significantly and quadratically with increasing nitrogen rates.

Although there was varietal difference in thousand grains weight, there was no significant change on this parameter due to nitrogen fertilizer application in both seasons (Fig 4.2i (a)& (b) respectively).

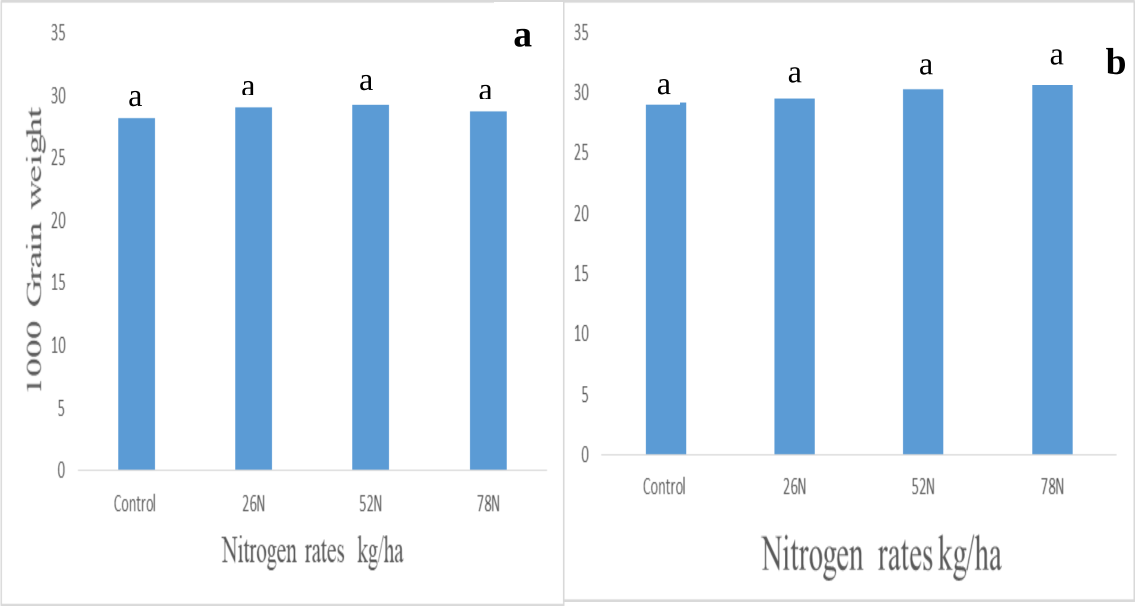


Figure 4.2(i): Mean thousand grain weight as affected by nitrogen rates, season1-a, season 2-b.

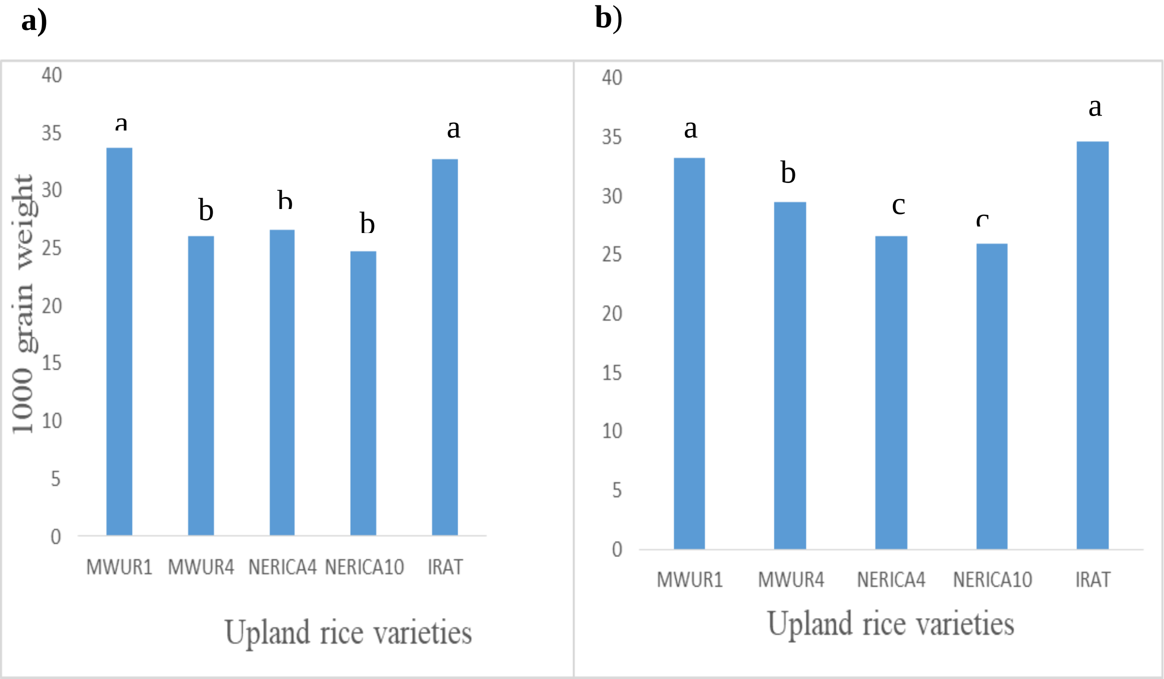


Figure 4.2(ii): Mean thousand grain weight as affected by upland rice varieties, season1-a, season 2-b.

These results further disagree with the findings of Weerakoon *et al*. 2005, who reported that a thousand grains weight increased with increasing nitrogen rate. Rice variety MWUR 1 exhibited higher thousand grain weight in both seasons while variety NERICA 10 gave the lowest values of one thousand grain weight in both seasons

4.8. Harvest Index

Harvest index is the physiological efficiency and ability of a crop to convert the total dry matter into economic yield. It shows the physiological capacity of a plant to change the product of photosynthesis to final yield. Rice varieties NERICA 4, followed by MWUR 4 recorded higher harvest index in season one. Variety NERICA 10 recorded higher value of harvest index in season two. All the other rice varieties had similar harvest indices in both seasons. There was no significant effect ($P \leq 0.05$) of adding Nitrogen fertilizer during the two seasons. Harvest index is not always constant but varies between sites and seasons due to differences in environment and nitrogen supply among the rice genotypes. According to Fageria, (2007) nitrogen improves grain harvest index and plant height which are positively associated with grain yield. Higher harvest index indicates a larger percentage of total dry matter transformed into grain yield (fig 4.3).

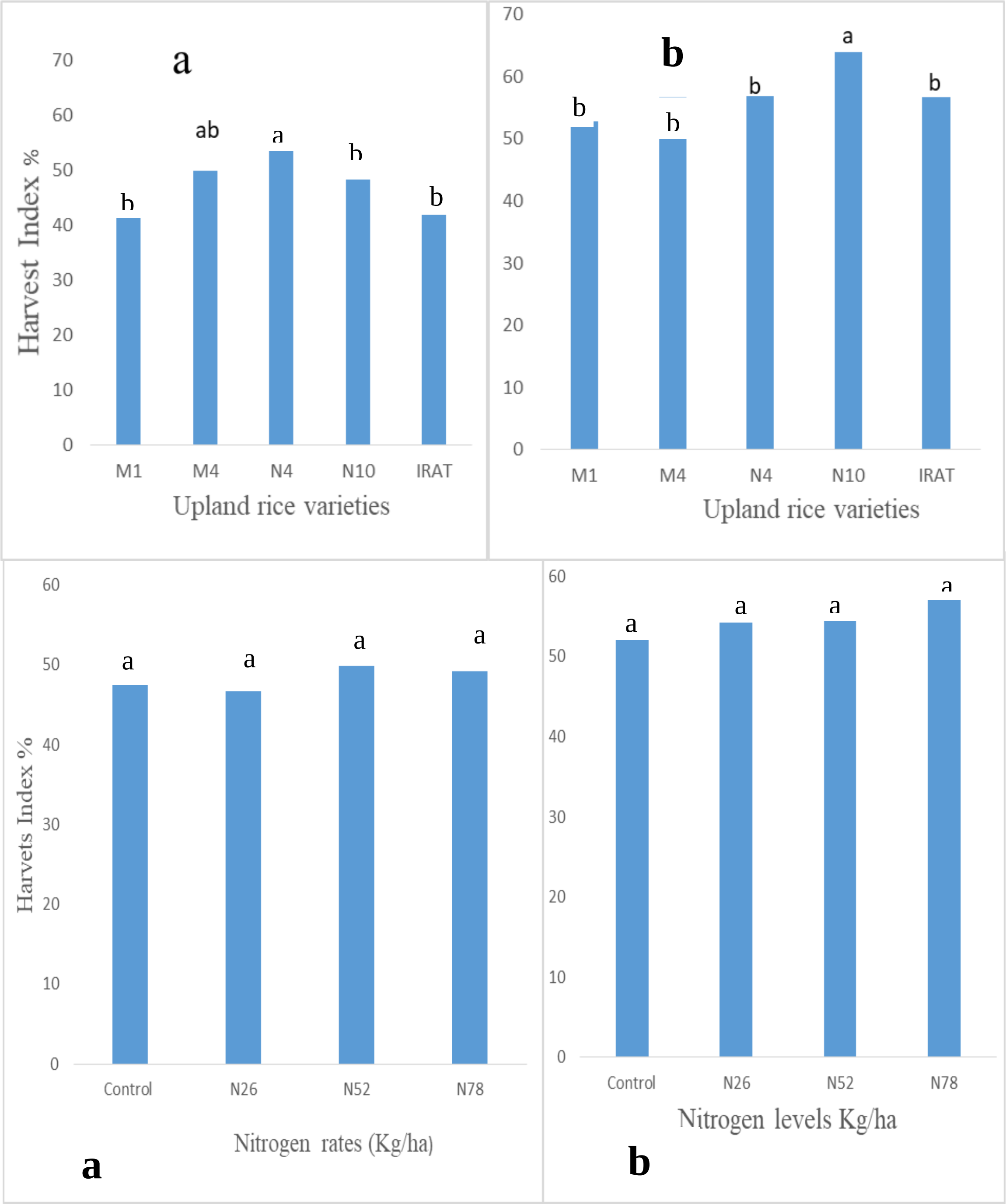


Fig 4.3 **Effects of rice varieties and N-rates on harvest index for two seasons, (a) S1 (b) S2 respectively.**

From the study, varietal difference was observed in harvest index which is a sign of efficient translocation of assimilates for grain formation. The harvest index did not increase with increase in N-level (Fig 4.3). The results from this study disagree with the findings of Fageria, (2007) who reported that nitrogen levels improve the harvest index.

4.6.6 Grain yield (gm/plant)

There were significant differences ($P \leq 0.05$) among the various rice varieties in grain yield produced. In addition yield increased with increasing nitrogen levels (Fig. 4.4).

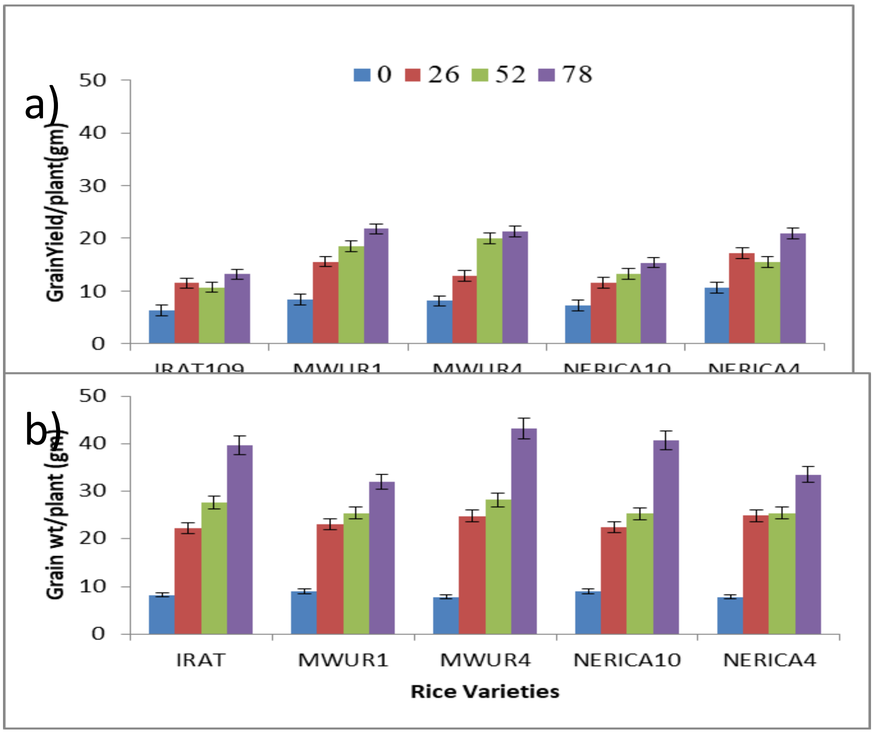


Figure 4.4: Interaction influence between upland rice varieties and N-levels on grain yield during, (a) season 1 and (b) season 2.

Varieties MWUR 1 and NERICA 4 recorded higher grain yield at low nitrogen levels in season one however; they were more responsive to high nitrogen application levels. There was no significant interaction between variety and nitrogen rates. These results on yield disagree with those of Fageria and Barbosa Filho (2001) that depicted an interaction effect of nitrogen and rice genotypes on their yield and yield components. However they confirm the findings of Li *et al*. (2012) and Manzoor *et al.* (2006) who reported that rice grain yield significantly increased with increasing nitrogen fertilizer application. The increase in grain yield might be due to nitrogen application enhancing the dry matter production, improving rice growth rate promoting elongation of internodes and activity of growth hormones like gibberellins. These results are supported by the findings of Singh *et al. (*2000).

There was high significant effect ($P \leq 0.05$) on grain yield with nitrogen application with MWUR1, MWUR4 and NERICA4 rice varieties producing higher grain yields at low nitrogen levels (Fig.4.4). These same upland rice varieties had higher plant height, more tiller numbers and higher shoot dry weight compared with the other tested varieties. The results of this study agree with the findings of Garba *et al*. (2013) who reported that nitrogen application enhances dry matter production and improves rice growth rate. Generally, grain yield increased with increasing N-level (Shaobing *et al*., 2010).

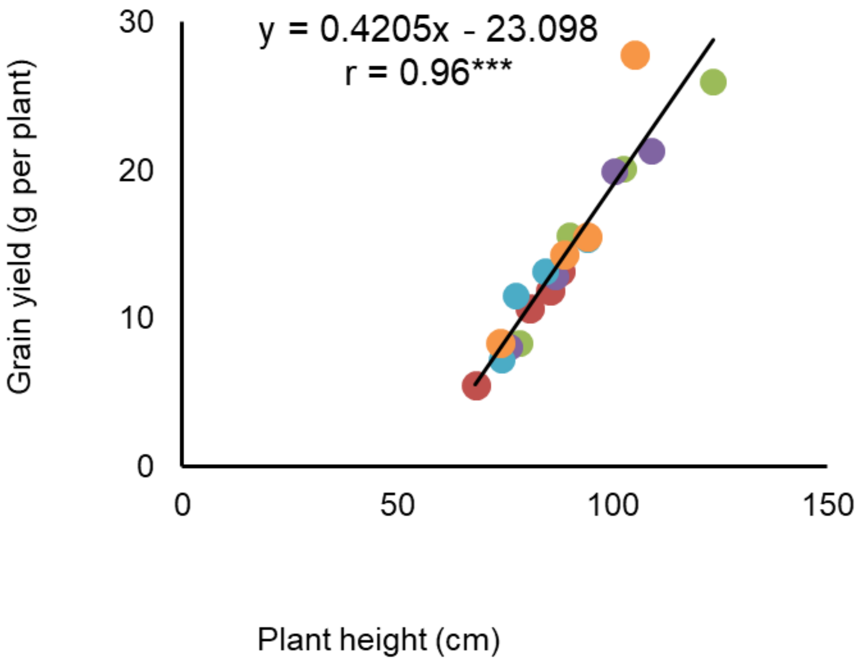


Fig. 4.5: Relationship between mean grain yield and plant height in upland rice varieties

The interaction between N-level and variety was not significant in both seasons. This confirms the findings of Memory *et al.* (2013) who reported that there were no significant differences in N rates and variety interaction. From the study, grain yield was significantly and positively correlated with panicle length in the tested upland rice varieties (fig 4.6). Rice grain yield increased with increase in panicle length in the tested rice varieties

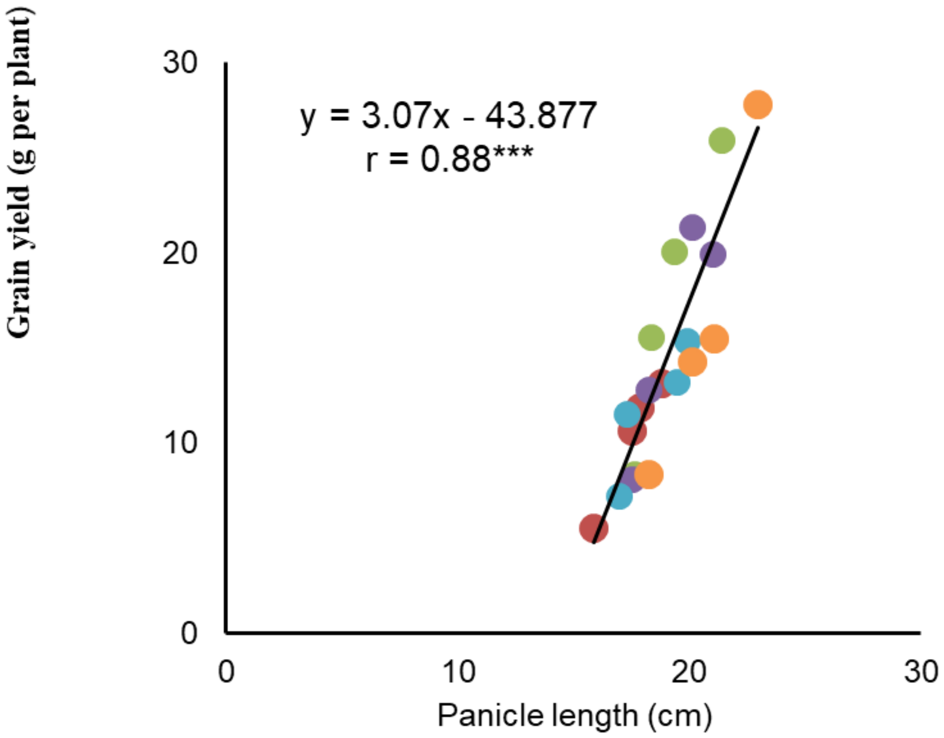


Fig. 4.6: Relationship between mean grain yield and panicle length in upland rice varieties

Low nitrogen stress slow down carbohydrate synthesis and/or weaken the sink strength at reproductive stages and abortion of fertilized ovaries (Rahman *et al*. 2002). Leaves are sink for nitrogen during the vegetative stage, and afterwards this nitrogen is remobilized for use in the developing seeds. There was significant effect ($P \leq 0.05$) on yield components and grain yield due to both variety and N-fertilizer rate applied in all the upland rice varieties. From the study, grain yield had significantly positive correlation with plant height (Fig. 4.5). Yield components like culm length had positive correlation with grain yield (Fig 4.7).

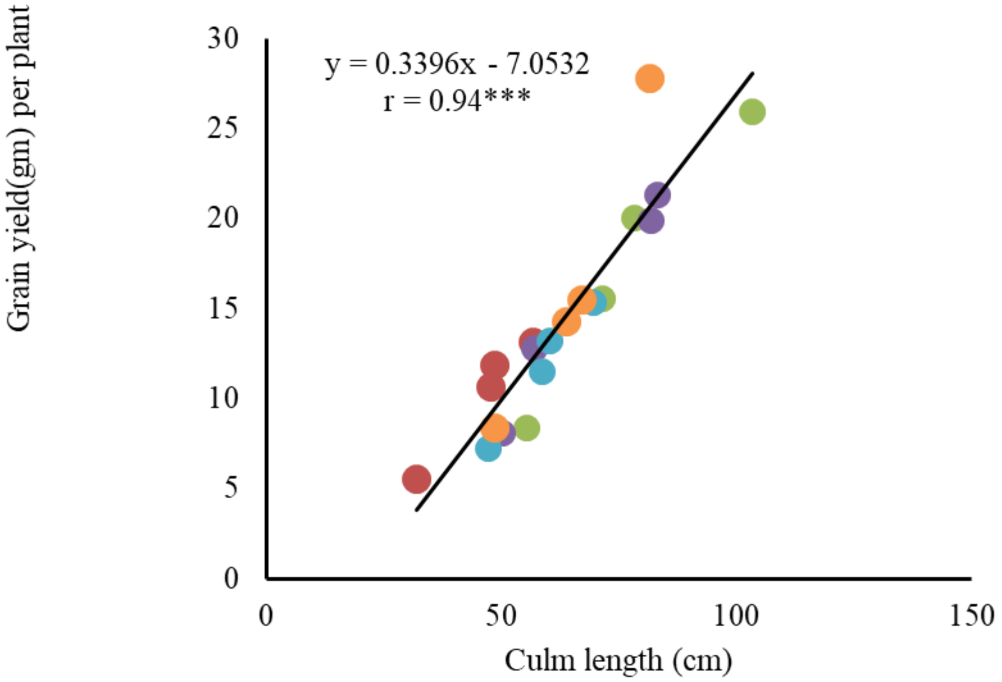


Fig. 4.7: Relationship between mean grain yield and culm length in upland rice varieties

4.7 Association between grain yield and nitrogen fertilizer levels in rice varieties.

In season one (July-September, 2015), the tested rice varieties showed positive yield response to nitrogen rate however, this started to decrease after a certain level of nitrogen application, while in season two (December 2015-February, 2016) the same varieties had positive yield responsiveness to increase in nitrogen levels. The regression was polynomial between grain yield and N-levels for all tested rice varieties except NERICA10 which was linear, in season one (Fig.4.8i). This could possibly be due weather conditions which exhibited low temperatures and less rainfall during the first season (Appendix 2).

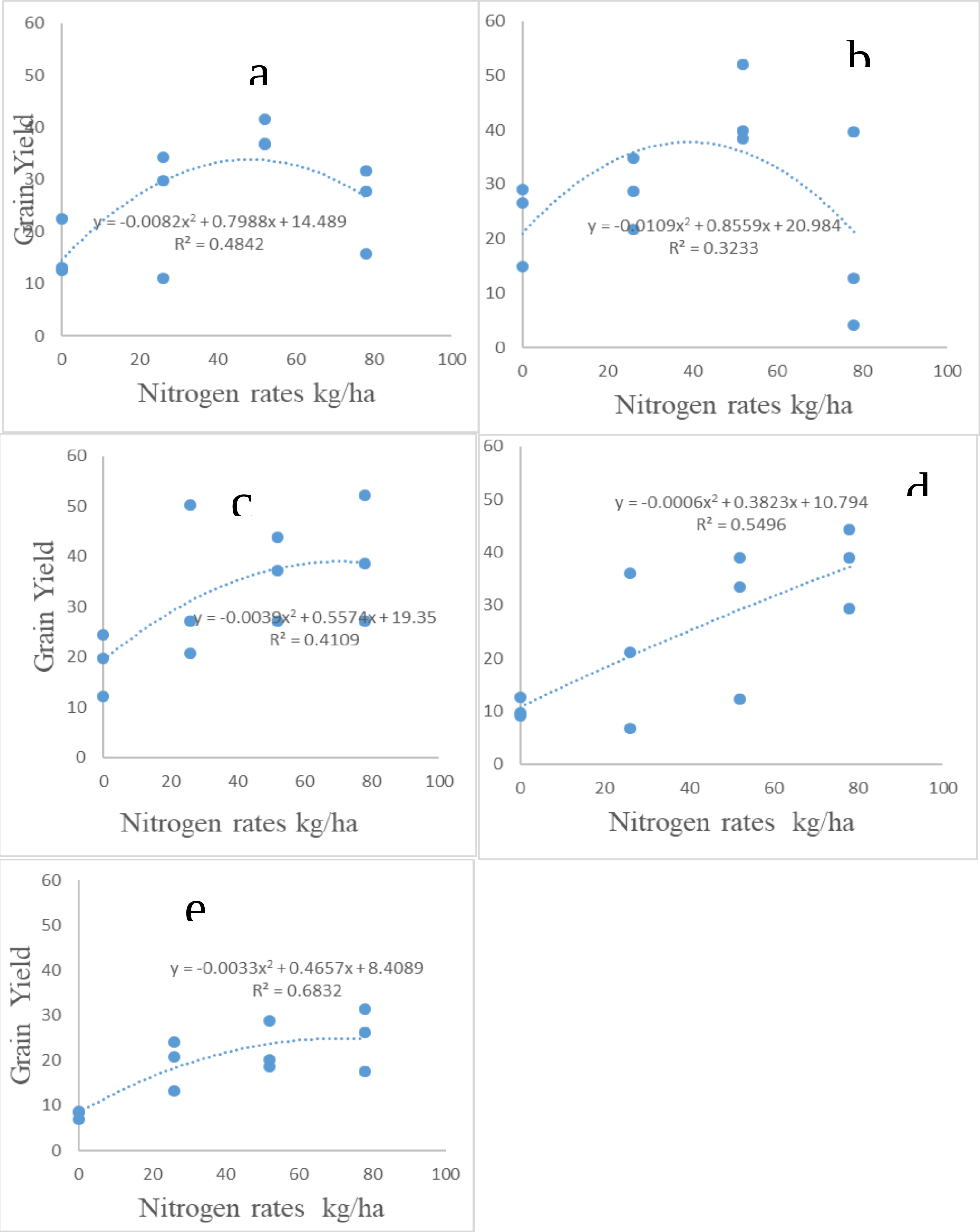


Fig. 4.8(i) Regression analysis as a polynomial function on grain yield (g/plant) and nitrogen rates for five rice varieties in season 1. (Mwur1 (a), mwur4 (b), Nerica 4 (c), Nerica10 (d), IRAT (e).

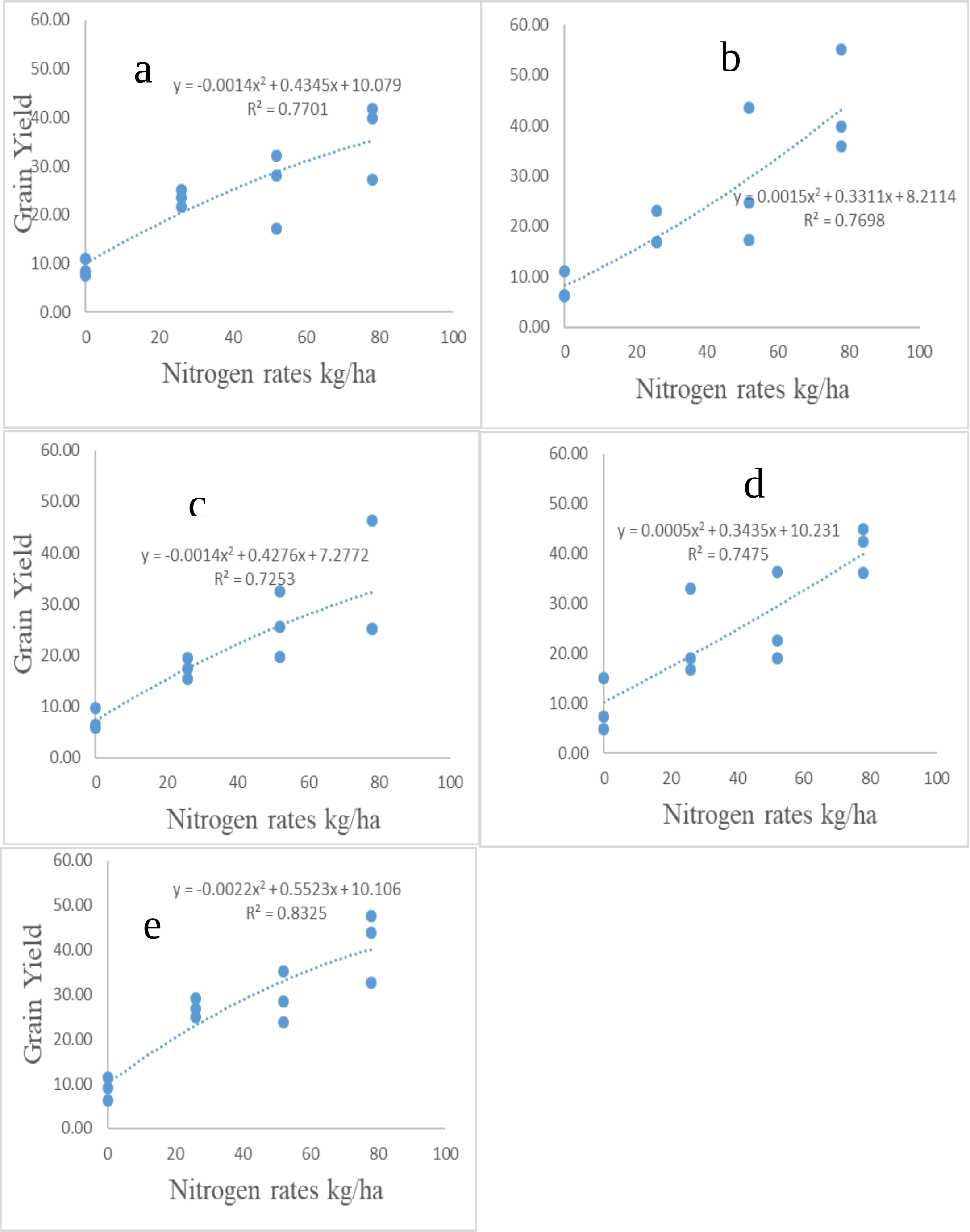


Figure 4.8(ii): Regression analysis as a polynomial function on grain yield (g/plant) and nitrogen rates for five rice varieties in season 2. (Mwur1 (a), Mwur4 (b), Nerica 4 (c), Nerica10 (d), IRAT (e))

Maximum crop production requires complete capture of incident radiation and can only be achieved with supporting level of water and nutrients (Loomis and Connor, 2002). Plants adjust their own traits to adapt to different environments. Precipitation might affect the photochemical activity of chloroplasts consequently; chlorophyll synthesis and water are closely related.

From the study there were clear and positive correlations between nitrogen levels and grain yield. The regression was polynomial between N rates and grain yield for most of the tested rice varieties in season one, implying any amount of N applied beyond 78kgN/ ha may not elicit further yield increment, except NERICA 10 which was linear implying further yield increment could still be achieved by applying N beyond 78kgN/ha. The same was repeated in season two. While the associations may imply that any amount of N applied beyond the higher rate of 78 kg/ha may not trigger further yield increment in MWUR1, MWUR 4 NERICA 4 and IRAT 109, further yield increment could still be achieved for NERICA 10 by applying N beyond 78 kg/ha which was the highest amount in this experiment.

4.8 Rice Biomass as affected by nitrogen fertilizer levels application.

4.8.1 Shoot Dry weight

There was significant variation ($P \leq 0.05$) in shoot dry weight among the varieties in season one. MWUR 1 recorded higher shoot dry weight followed by MWUR 4 and NERICA 4. The lowest shoot dry weight value was observed in IRAT 109 during the two seasons (Table 4.6). There was significant effect on shoot dry weight in the tested upland rice due to variety and N-fertilizer rate applied in both season one and season two. There was significant effect due to interaction between variety and N-rates evaluated (Table 4.6).

Table 4.6: Effects of N-rates and rice variety on shoot dry weight , root dry weight and above ground biomass for the two seasons

	Season 1			**Season 2**		
Varieties	**SDW**	**RDW**	**AGBM**	**SDW**	**RDW**	**AGBM**
MWUR1	30.30^{a}	2.82^{a}	56.17^{a}	19.73^{a}	1.25^{a}	42.92^{a}
MWUR4	24.75^{ab}	2.87^{a}	54.31^{a}	18.20^{a}	1.26^{a}	42.42^{a}
NERICA 4	24.87^{ab}	2.97^{a}	33.74^{a}	16.27^{a}	1.41^{a}	34.90^{a}
NERICA10	19.70^{b}	2.82^{a}	41.83^{a}	14.18^{a}	1.21^{a}	37.24^{a}
IRAT	18.38^{b}	2.89^{a}	37.88^{a}	12.46^{a}	1.29^{a}	42.82^{a}
$LSD_{0.05}$	6.85	0.39	15.86	6.62	0.38	16.82
N rates kgha^{-1}						
Control	16.51^{c}	2.56^{b}	32.40^{c}	6.52^{c}	0.85^{a}	24.95^{c}
26	21.55^{bc}	2.88^{a}	44.55^{bc}	14.69^{b}	1.25^{a}	36.60^{b}
52	29.85^{a}	2.91^{a}	63.15^{a}	18.53^{b}	1.42^{a}	42.57^{b}
78	26.49^{ab}	3.14^{a}	56.27^{ab}	24.95^{a}	1.60^{a}	63.13^{a}
$LSD_{0.05}$	5.69	0.31	11.98	3.46	0.27	7.10
V*NR	*	NS	NS	*	NS	NS

Values with the same letter(s) within the column are not significantly different

(SDW= shoot dry weight, RDW= root dry weight, AGBM= above ground biomass)

In the second season, the same trend was repeated with MWUR 1 recording higher shoot dry weight compared with other tested varieties. However, there was no significant difference among the rice varieties in season two. High nutrient use efficiency is generally important under conditions of low nitrogen availability, as it entails high biomass production per unit nitrogen uptake. It is a useful index that combines both plant physiological and morphological responses along the nutrient availability gradient. Fageria *et al.* (1997) and Fageria (2007) reported that rice yield was highly correlated with shoot dry weight. Leaves are sinks for nitrogen (N) during the vegetative stage, and afterwards this nitrogen is remobilized for use in the developing seeds. Nitrogen is an essential component of plant cells at the structural, genetic and metabolic levels getting involved in many processes of plant growth and development which finally lead to yield as well as the quality of the harvested organs which include seeds and above ground biomass (Dardanelli *et al.,* 2004).

4.8.2 Root dry weight (gm)

Roots are crucial for water uptake and nutrient supply both under water limiting and non-limiting conditions, thus influencing crop water use- efficiency, (WUE) and grain yield. Under soil water deficit, crop water extraction depends on root distribution and depth (Dardanelli *et al.*, 2004). Rice variety NERICA 4 recorded higher root dry weight in both seasons among the tested varieties however; the difference in root dry weight was not significantly different. The lowest weight was observed at 0kgN ha^{-1} (Table 4.6). According to Paul *et al.* (1997) and Malamt, (2005), low nitrogen supply generally leads to decreased root growth, suppression of lateral root initiation, reduction in photosynthesis and early leaf senescence. In rice plants, roots play a significant role in absorption of nitrogen with root density and distribution in the soil being the major determinant (Youngdahl *et al.,* 1982). Root characteristics such as root length density and root weight have been identified as important factors since nitrogen uptake is determined by root mass and nitrogen uptake per root volume (Shimono and Bruce, 2009). Nutrient uptake in root is primarily by mass flow and diffusion. These mechanisms decrease as the moisture content of the soil decrease (Parish, 1971).

The rooting system in plants is the main organ of nutrient absorption and transportation. It is also the direct user of soil nutrients and an important contributor of the yield. According to Wu *et al.* (1995), good morphological development of the root system increases the surface of its contact with nitrogen, hence promoting the efficiency of nitrogen absorption. Roots are an important factor that influences the efficiency of nitrogen absorption from the fertilizer. Greater root length density can improve nutrient acquisition by increasing the root surface area (Marschner, 1995). The results from our study showed that root condition was important for upland rice growth. There was a positive correlation with root dry weight and grain yield under low nitrogen condition (Fig 4.9).

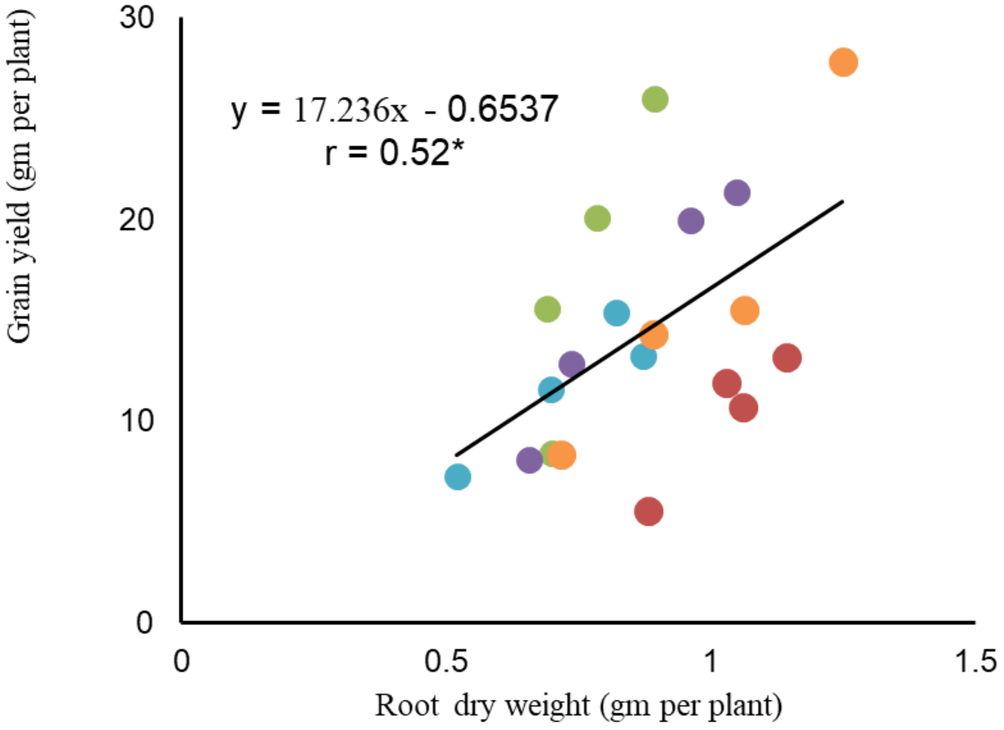


Fig. 4.9: Relationship between grain yield and root dry weight under low nitrogen

4.8.3.1 Effect of N-level on rice biomass

There was no varietal difference in above ground biomass among the tested rice genotypes (Table 4.6). Although MWUR 1 recorded higher above ground biomass followed by MWUR 4 in both season one and two. There was no interaction between nitrogen rates and rice varieties during the two seasons.

4.9: Root: Shoot ratio

Varietal difference in root-shoot ratio was not significantly different in the two seasons. Root: Shoot ratio is symbiotic. The root surface makes up the interface with below ground environment and determines the efficiency of obtaining external resources such as water and nutrients. From the study, NERICA 4 registered higher root: shoot ratio in both seasons (Fig 4.10).When soil nitrogen is readily available, plants tend to develop a smaller root to shoot ratio (Marschner, 1995). Root to shoot ratio also changes with plant developmental history.

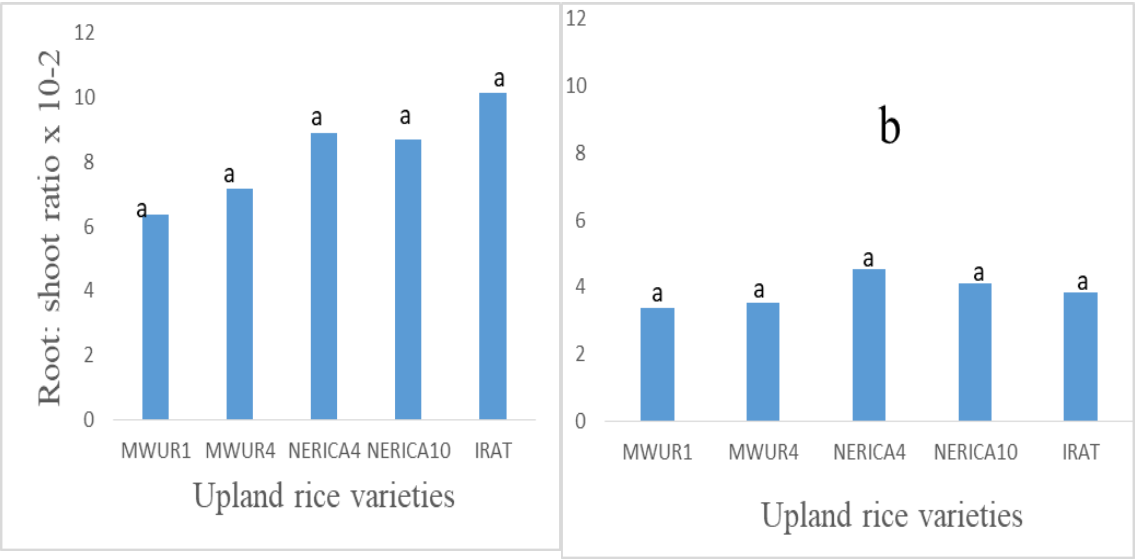


Fig 4.10: Varietal effect on root-shoot ratio for two seasons

Excess root system reduces growth of shoot due to competition of carbohydrates between root and shoot. The uptake rate depends on the root surface area as photosynthesis rate depends on leaf area. With high nutrition levels, root to shoot ratio are generally lower (Glass, 2003). Roots are an important factor that influences the efficiency of nitrogen absorption from the fertilizer. For a plant to access more nutrients, bigger root system is paramount. This strategy comes at a cost, since bigger roots take more carbon away from the shoots limiting the plant's capacity to fix and store carbon in the harvested yield.

Table 4.7: Correlation between grain yield and yield components of upland rice varieties

Plant Parameter	**Grain yield**
Plant height	0.96**
Tillers	0.315*
Culm length	0.503**
Panicle length	0.88**
Number panicles	0.447**
Grains per panicle	0.042 ns
Filled grain wt.	0.477**
Harvest index	0.886**
Shoot dry wt.	0.656**
Root dry wt.	0.52*
Stomatal conductance	0.418**

** Correlation is significant at 0.01 level

* Correlation is significant at 0.05 level

ns not significant

In the study, nitrogen treatments led to significant difference in plant growth and plant height which increased with nitrogen levels. Nitrogen increased plant vigor, hence the increase in plant height due to enhanced cell division and cell elongation, leading to vigorous vegetative growth. When nitrogen is absorbed during vegetative phase, it enhances synthesis of chlorophyll which is necessary for photosynthesis and this promotes rapid leaf, stem and root growth. These results are consistent with the findings of Manzoor *et al.* (2006) and Meena *et al.* (2003). Nitrogen improved plant height, which is positively associated with grain yield (Fageria, 2007).

Generally rice varieties MWUR1, MWUR4 and NERICA4 exhibited higher plant heights at low nitrogen level of 26kgN/ha. The vigorous growth or slow growth of different upland rice genotypes could be due to varietal difference and their inherited characteristics.

Tillers are an important component of yield. Branching comprises of a protective mechanism for higher plants, by assisting in reducing the impact of injury and adaptation to environment (Harvath *et al*., 2003). According to Ling, (2000) tillering is an important agronomic trait for rice population quality and grain production. Though number of tillers per hill increased with increase in nitrogen levels, they were not significantly different at 26kgN/ha and 52kgN/ha. Similar results were reported in hybrid rice by Chaturvedi, (2005). The enhanced tillering by increased nitrogen level might be attributed to more nitrogen supply to the plant at vegetative stage enhancing cell division.

There was positive correlation between yield components and tiller numbers, hence increasing nitrogen level led to increase of tiller numbers which impacted on yield. Upland rice varieties MWUR1 and MWUR4 recorded higher tiller numbers at low nitrogen level. This could possibly be due to difference of varietal performance. The rate of leaf expansion significantly increased with increase in nitrogen level leading to increased interception of daily solar radiation hence higher photosynthetic activities. Rice N- requirements are closely related to yield levels, which in turn are sensitive to climate particularly solar radiation and supply of other nutrients (Peng *et al.*, 1995a).

There was very high significant effect on root weight due to nitrogen level which increased with increase in nitrogen level. Wu and Tao (1995) held that the good morphological development of the root system increased the surface of its contact with nitrogen thus promoting its efficient nitrogen absorption and the ratio of root and stem. Roots play a significant role in nitrogen absorption in rice during vegetative stage with root density and distribution in the soil being the major determinants (Youngdahl *et al.*, 1982).Under low nitrogen conditions, rice plants attempts to acquire more nitrogen by increasing the root surface area, which increases the root to shoot ratio (Marschner *et al.*, 1986). According to Song *et al.*, (2004) the response of rice to nitrogen is a complicated dynamic process influencing photosynthesis and root system response to nitrogen absorption. The beneficial effects of nitrogen occur by influencing yield components, number of panicles per unit area, one thousand grain weight and panicle length (Fageria *et al.*, 2001).

There was significant influence on grain yield, thousand grain weight and above ground biomass due to nitrogen levels. Yield components including Culm length and thousand grain weight increased with increase in nitrogen level. The improved growth attributes such as plant height and more number of tillers at higher nitrogen levels might be responsible for improved yield attributes. Filled grain ratio increased with increase in nitrogen level. These results confirm the findings of Rafey *et al.* (1989). At low nitrogen MWUR1, MWUR4, NERICA 10 and IRAT 109 recorded high nitrogen percent in rice grain respectively. Human nutritional value of rice was not really changed with the improvement of N- use efficiency. At 0kgN/ha, more nitrogen nutrient was partitioned to the rice grain, which confirms that plants develop a mechanism to sense nitrogen status in the root system and the surrounding soil environment. Rice varieties MWUR1, NERICA 10 and IRAT 109 recorded high nitrogen percent in the straws at low nitrogen level of 0kg /ha. This study has shown that all yield components strongly correlate with grain yield. This confirms that changes in these components will affect rice grain yield.

4.10 Grain and plant tissue Nitrogen analysis

In plant life cycle, there are two general stages of nitrogen use. During biomass formation there is the amount of nitrogen uptake, storage and assimilation into amino acids and other important nitrogenous compounds. The second stage is the proportion of nitrogen that is partitioned to the seed, resulting in final yield. Plants have developed efficient methods and mechanisms that release tied-up nitrogen entities from source tissues via protease activities during leaf senescence. Approximately 80% of the total leaf nitrogen is located in the chloroplast mainly in the form of proteins and this is an important nitrogen pool for remobilization (Adam *et al*., 2001). After protein degradation during senescence, the amino acids released from roots and leaves are loaded into the phloem. The amino acids are the major form for nitrogen transport required for grain development. The absorbed nitrogen used for rice straw and leaf growth at tillering stage was transported to the panicles at advanced developmental stages (Guindo *et al*., 1994, Liu *et al*., 2007). Large differences occur among cultivars of the same species in absorption, translocation and utilization of mineral nutrients. Nitrogen harvest index is a measure of N partitioning in rice, which provides an indication of how efficient the plant utilizes the acquired N for grain production. Up to 80% of grain nitrogen contents are derived from leaves in rice and wheat (Kichey *et al.,* 2007). The N content (%) in the rice grains was higher than that in the above ground biomass in all rice varieties tested (Table 4.8). This confirms the findings of Liu *et al*. 2007.

Table 4.8: Mean % N in rice seed and biomass as affected by different nitrogen rates in upland rice varieties.

	Season 1&2	
Varieties	**%N B/mass**	**% Seed**
MWUR1	2.98^{a}	7.81^{a}
MWUR4	3.13^{a}	7.65^{a}
NERICA 4	2.90^{a}	6.50^{a}
NERICA10	3.05^{a}	7.77^{a}
IRAT	2.62^{a}	7.25^{a}
LSD	0.79	1.10
N rates kgha^{-1}		
Control	3.41^{a}	7.95^{a}
26	2.82^{a}	7.32^{ab}
52	2.70^{a}	6.44^{b}
78	2.82^{a}	7.87^{a}
LSD	0.68	0.94
V*NR	*	NS

Means followed by the same letter(s) within the same column are not significantly different. V-varieties of rice, NR- nitrogen rates

There was no significant effect on percent nitrogen in both rice seed and biomass due to variety; however there was significant effect due to nitrogen rates in the seed. The interaction between variety and N-rate was significant in the biomass. The nitrogen partitioned to the grain was higher compared to the biomass in all tested varieties. This is an important characteristic in terms of efficiency and health but has implication in nitrogen transport since the grain is always sold away unlike the straw that is at times left in the field to decompose and enhance nitrogen cycle.

4.10.1 Effect of N-levels on nutrient nitrogen partitioning to the seed and above ground biomass in upland rice varieties.

There was also significant effect ($P \leq 0.05$) on percent nitrogen in rice grain due to variety. Upland rice varieties MWUR 1, MWUR 4 and NERICA 10 recorded high nitrogen percentage in grains at low N-levels of 0kgN/ha and 26kgN/ha (Fig. 4.11).

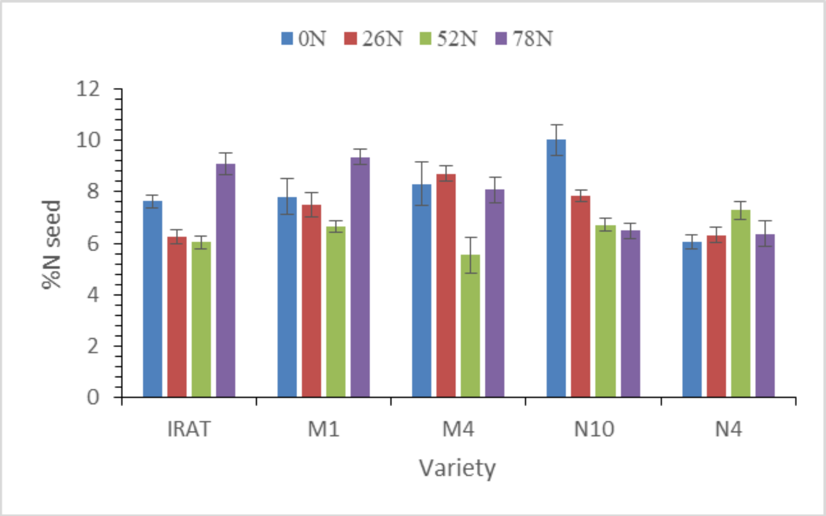


Figure 4.11: N-level influence on nitrogen nutrient content (%) in upland rice grains

The seed N-concentration pattern during the seed filling is the result of seed dry matter accumulation and seed nitrogen accumulation. The rate of seed dry matter accumulation is not significantly responsive to changes in plant nitrogen availability because it is determined before the beginning of the seed filling by the seed cell number (Jenner, 1991). In contrast, the rate of seed nitrogen accumulation can vary during seed filling upon nitrogen availability in plant.

From the study, more nitrogen was partitioned to the seeds at low nitrogen level, which confirmed results by Gombet *et al*. (2006), that under low mineral nitrogen availability, seed nitrogen accumulation comes from nitrogen accumulated daily by the plant and from nitrogen remobilizes from vegetative parts, which is partitioned among plant organs.

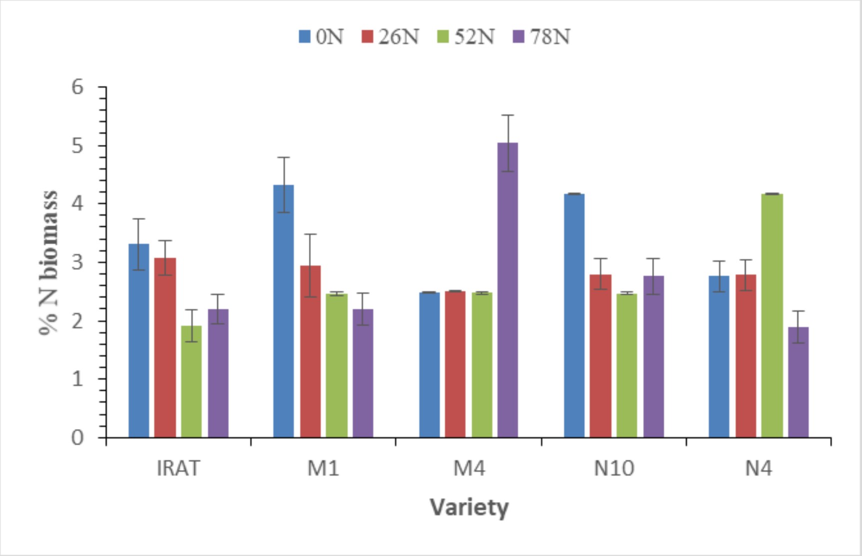


Figure 4.12: N-level influence on nitrogen nutrient content (%) in upland rice varieties biomass

There was significant effect ($P \leq 0.05$) on percent nitrogen in rice straws due to nitrogen levels. Upland rice varieties MWUR 1, NERICA 10 and IRAT 109 rice varieties recorded high nitrogen percentage in above ground biomass under low nitrogen levels of 0kgN/ha and 26kgN/ha (Fig.4.12). Leaves are a sink for nitrogen during the vegetative stage, and afterwards this nitrogen is remobilized for use in the developing seeds. According to Kichey *et al.* (2007) up to 80% of grain nitrogen contents are derived from leaves in rice and wheat. Plants have developed efficient methods and mechanisms that release tied-up nitrogen entities from source tissues though protease activities during leaf senescence.

CHAPTER FIVE: CONCLUSIONS AND RECOMMENDATIONS

5.1 Conclusions

Food production has to increase to meet the demand of a growing population. In light of the high energy costs and increasingly scarce resources, agricultural systems in future should be more productive and more efficient in terms of production inputs such as fertilizers. Despite the apparent yield barrier, the quest for higher yield potential continues. To increase rice production in Kenya, we need to improve productivity and yield stability. Upland rice is expected to be incorporated into the Kenyan cropping system. However, various biotic and abiotic stresses constrain rice production.

The current study has shown that increase in nitrogen level lead to significant increase in plant height, tiller numbers, leaf chlorophyll content and leaf area. However, this increase in nitrogen level had no significant effect on panicle length and harvest index. Upland rice variety MWUR1 registered higher plant height, filled grain ratio and thousand grain weights at low nitrogen level while rice varieties MWUR4 and NERICA4 registered higher harvest index, higher grains per panicle and higher root dry weight at low nitrogen. In upland conditions, appropriate crop, soil and water management practices can result in high rice grain yield. Such improved technologies can help to significantly improve yields and contribute to enhancing food availability and security in our country. Rain fed rice varieties for the future should be more responsive to mineral fertilizers, but they should retain the stress tolerance and grain quality. It may therefore be concluded that the nitrogen efficiencies are inherently affected by the rice varieties and levels of nitrogen. Upland varieties MWUR1, MWUR4 and NERICA4 were more adaptive to low nitrogen consequently, varieties MWUR 1 and NERICA4 may be good for both low input and also potentially good for high input N- levels.

5.2 Recommendations

Global food security is at stake since the demand for rice is exceeding production. To achieve the goal of doubling rice production, it is necessary to generate rice varieties that can overcome the stresses and establish cultivation methods that extract the full potential of the varieties. Low soil fertility is a major limit to Kenyan rice productivity (Gicheru, 2012). Currently, of importance is to minimize nitrogen use in crop culture under climate change condition. Kenya has potential of about 540,000 ha of irrigable land and about 1.0 million ha rain fed for rice production (MOA, 2010).

An increased knowledge on the physiological mechanisms controlling plant nitrogen economy under different nitrogen management practices is critical for improving NUE as well as reducing excess input of fertilizers while maintaining an acceptable yield and environmental quality (Ma and Biswas, 2015). Some upland rice varieties may do reasonably well under low soil nitrogen conditions.

These varieties need to be identified and the mechanism conferring their low soil nitrogen traits studied and identified. MWUR1, MWUR4 and NERICA4 proved to be plastic to external nutrient availability hence can survive and yield at low nitrogen stress. Nitrogen rate of 26kgN ha^{-}1 would be recommended for upland rice production in Kenya. This would save the resource poor farmer some costs, alleviate poverty and ensure clean environment for sustainable agriculture. However, there is need for further research on low soil nitrogen rice cultivation so as to develop a low cost upland rice cultivation technology for our Kenyan farmers.

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Appendices

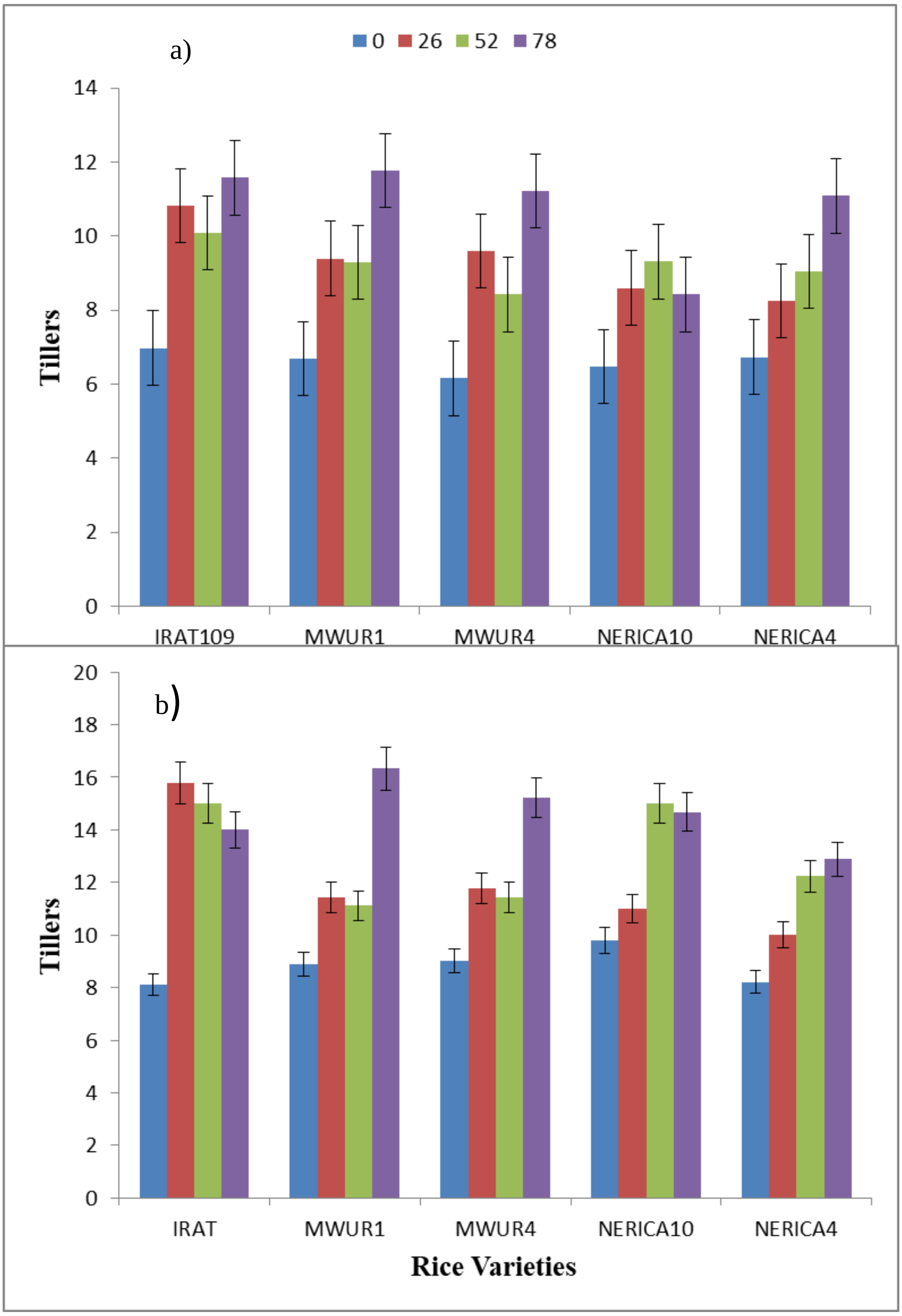
Appendix 1: Soil chemical properties at KALRO-Mwea experimental field season 1 and season 2.

Soil chemical characteristics			
	Season one		**Season two**
Soil PH	5.84	Soil PH	5.56
Total Nitrogen%	0.11	Total Nitrogen%	0.16
Total Organic Carbon%	1.09	Total Organic Carbon%	1.53
Phosphorus ppm	310	Phosphorus ppm	325
Potassium me%	0.98	Potassium me%	0.96
Calcium me%	2.7	Calcium me%	2.0
Magnesium me%	8.06	Magnesium me%	8.57
Manganese me%	0.11	Manganese me%	0.13
Copper ppm	2.65	Copper ppm	1.86
Iron ppm	18.3	Iron ppm	42.0
Zinc ppm	2.27	Zinc ppm	1.74
Sodium me%	0.3	Sodium me%	0.36

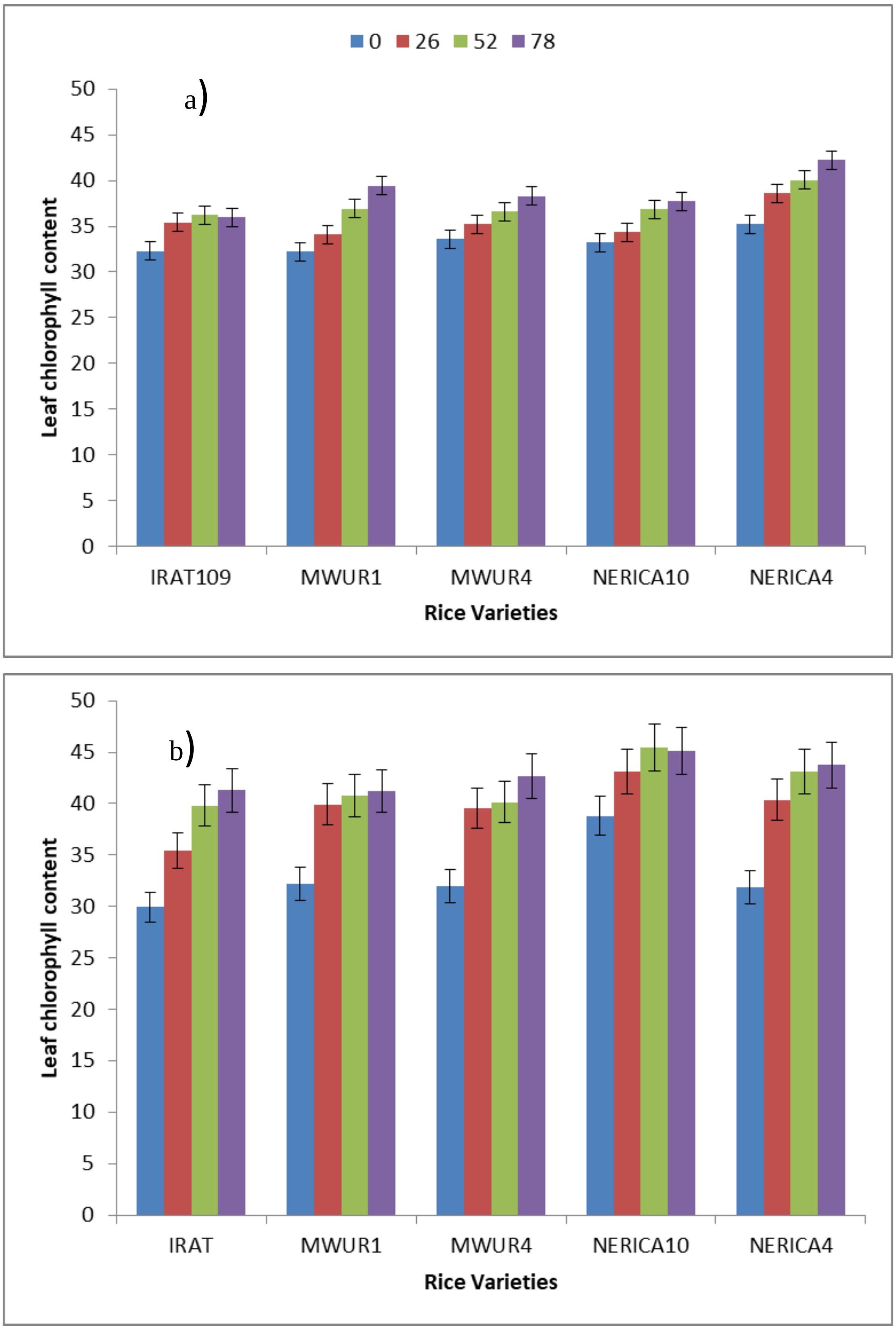
Appendix 2: Weather data for KALRO-Mwea experimental field

Rainfall (mm)			**Temperature (˚C)**	
Year 2015/16			**2015/16**	
Month	**mm**	**Rain days**	**Max**	**Min**
May 2015	84.4	8	27.6	18.0
June	17.4	5	26.3	16.7
July	10.2	1	25.9	15.5
Aug	9.8	2	26.7	16.0
Sept	0.0	0	28.5	16.5
Oct	70.2	13	29.5	17.6
Nov	375.3	22	27.3	17.1
Dec	86.5	8	28.3	15.7
Jan2 016	84.8	7	29.0	16.3
Feb	23.8	1	31.1	15.4
March	47.3	5	31.2	17.4
April	331.4	16	29.1	16.9
Total	**1141.1**	**88**	**340.5**	**199.1**
Mean	**95.1**	**7.3**	**28.4**	**16.6**

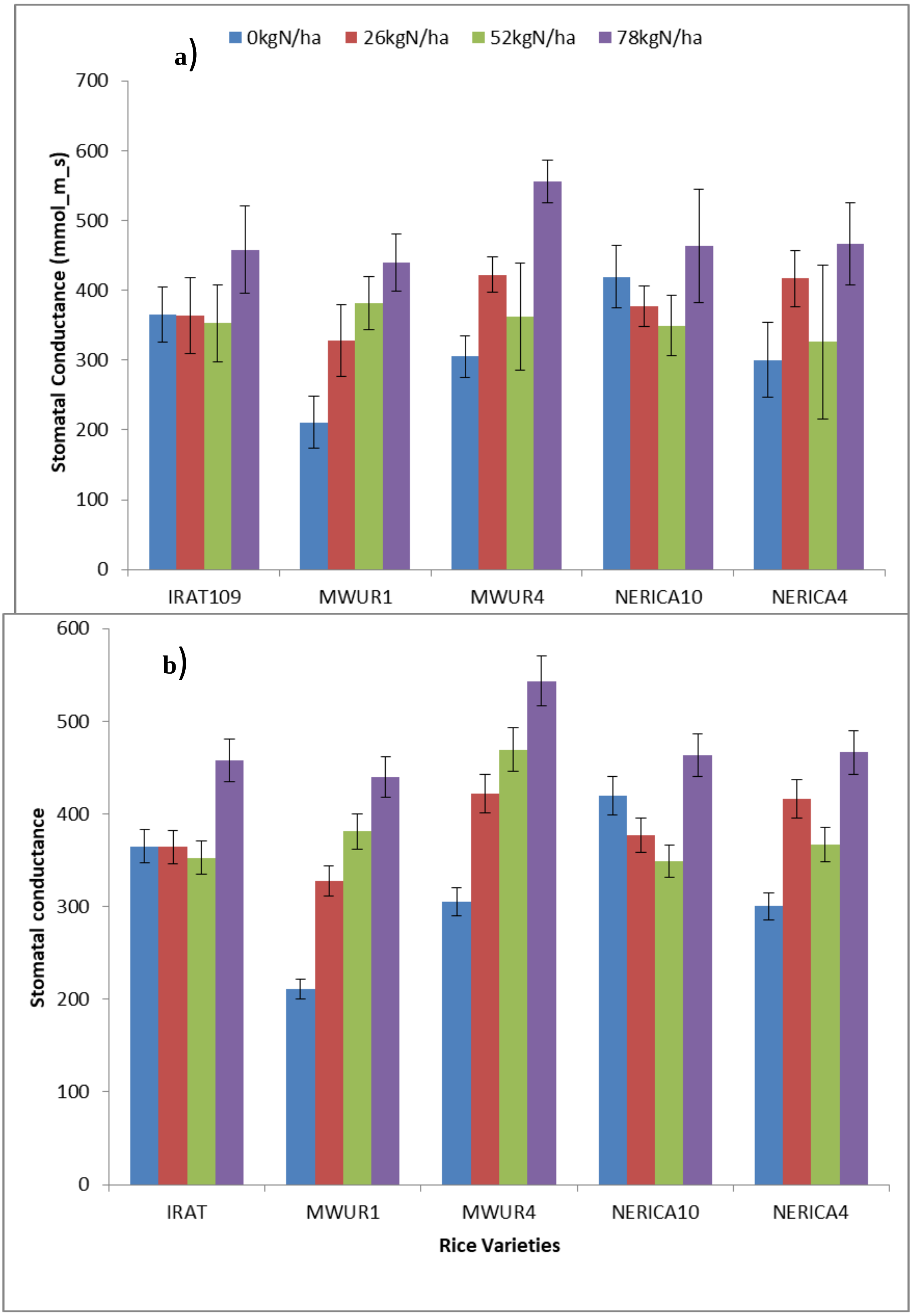
Appendix 3: Effect of N-levels on tiller numbers per hill in upland rice varieties.



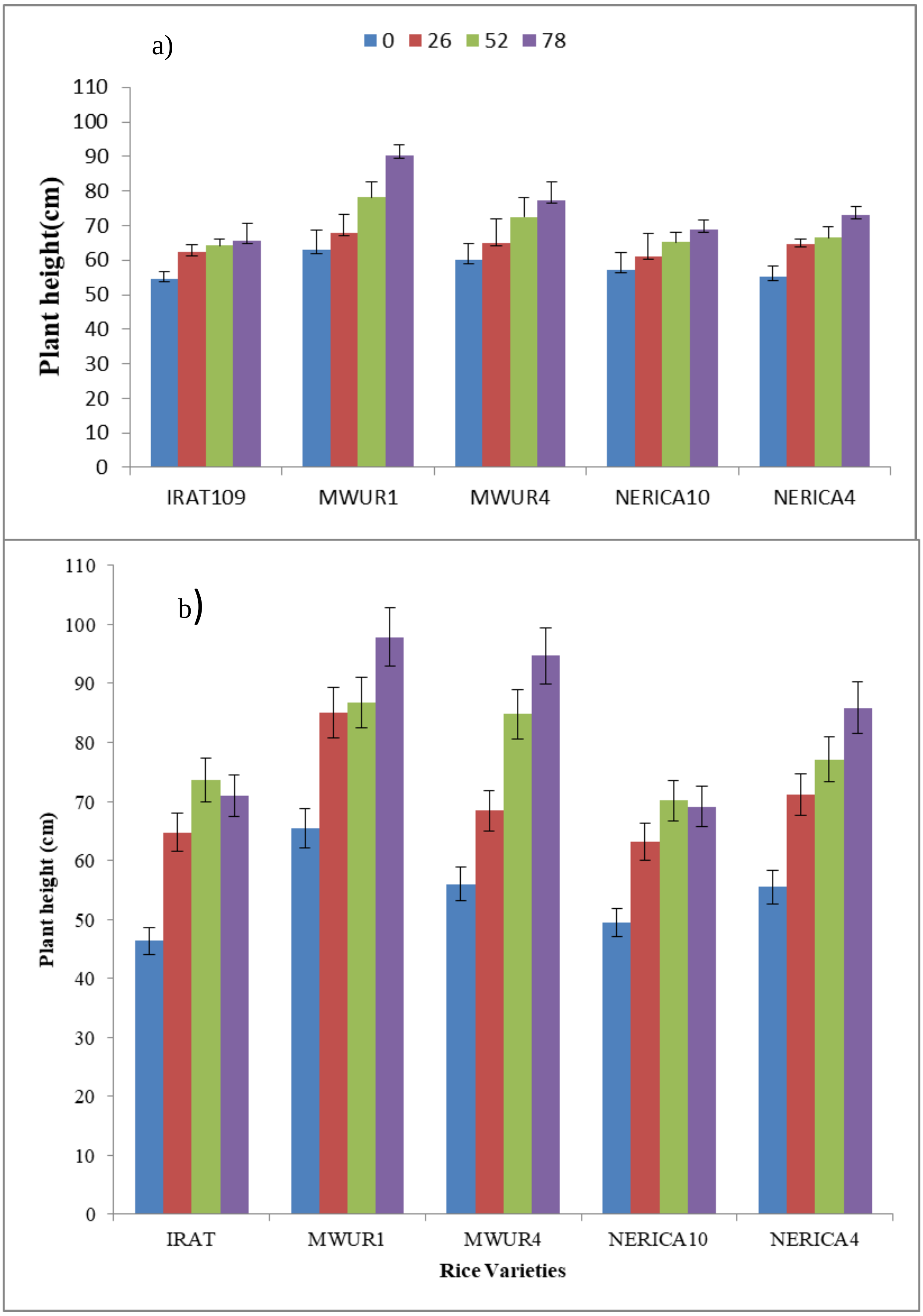
Appendix4: Effect of N-levels on leaf chlorophyll content in upland rice varieties, season one and two



Appendix 5: Effect of N-levels on stomatal conductance in upland rice, season one and two

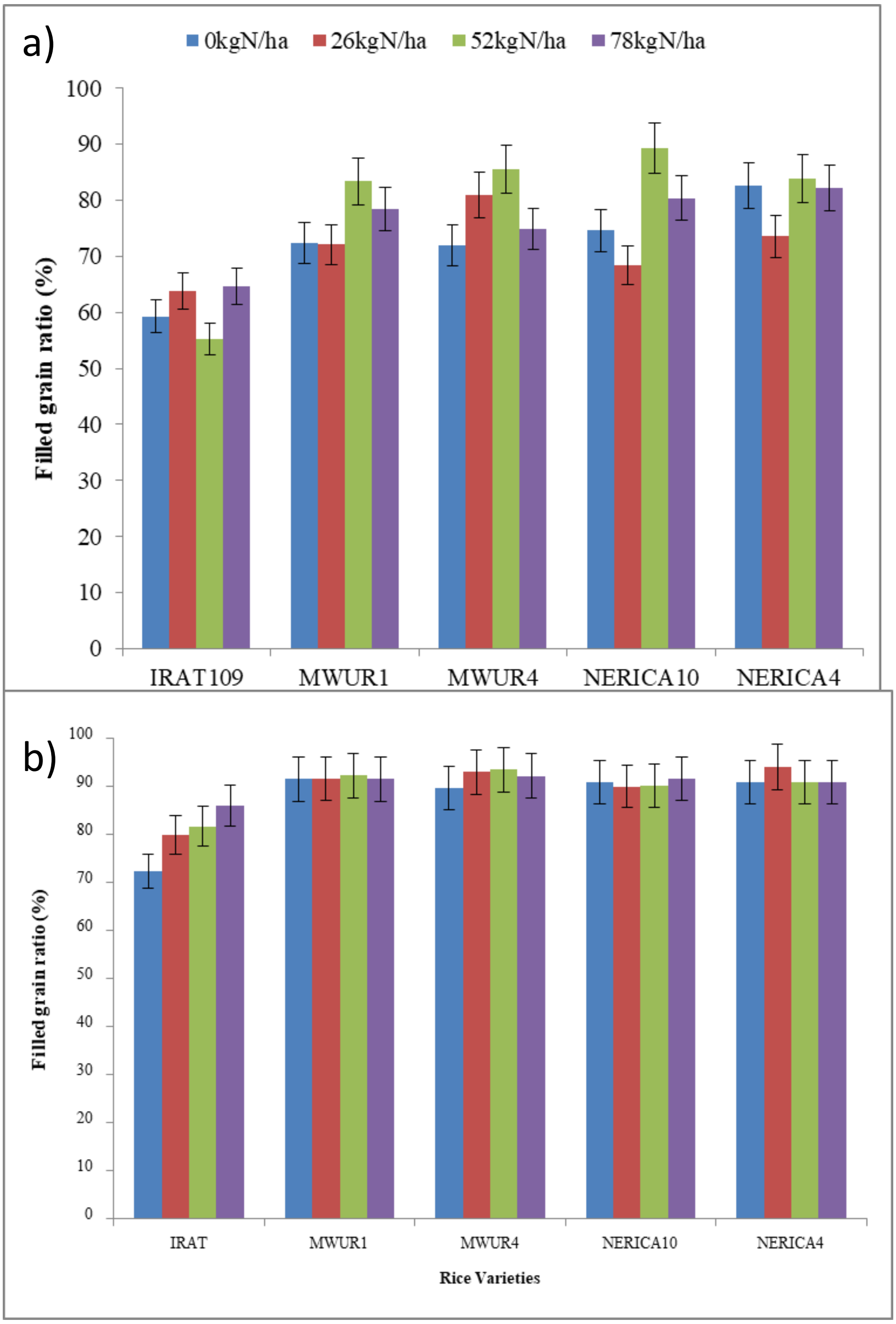


Appendix 6: Effect of N-level on plant height in upland rice varieties, season one and two.

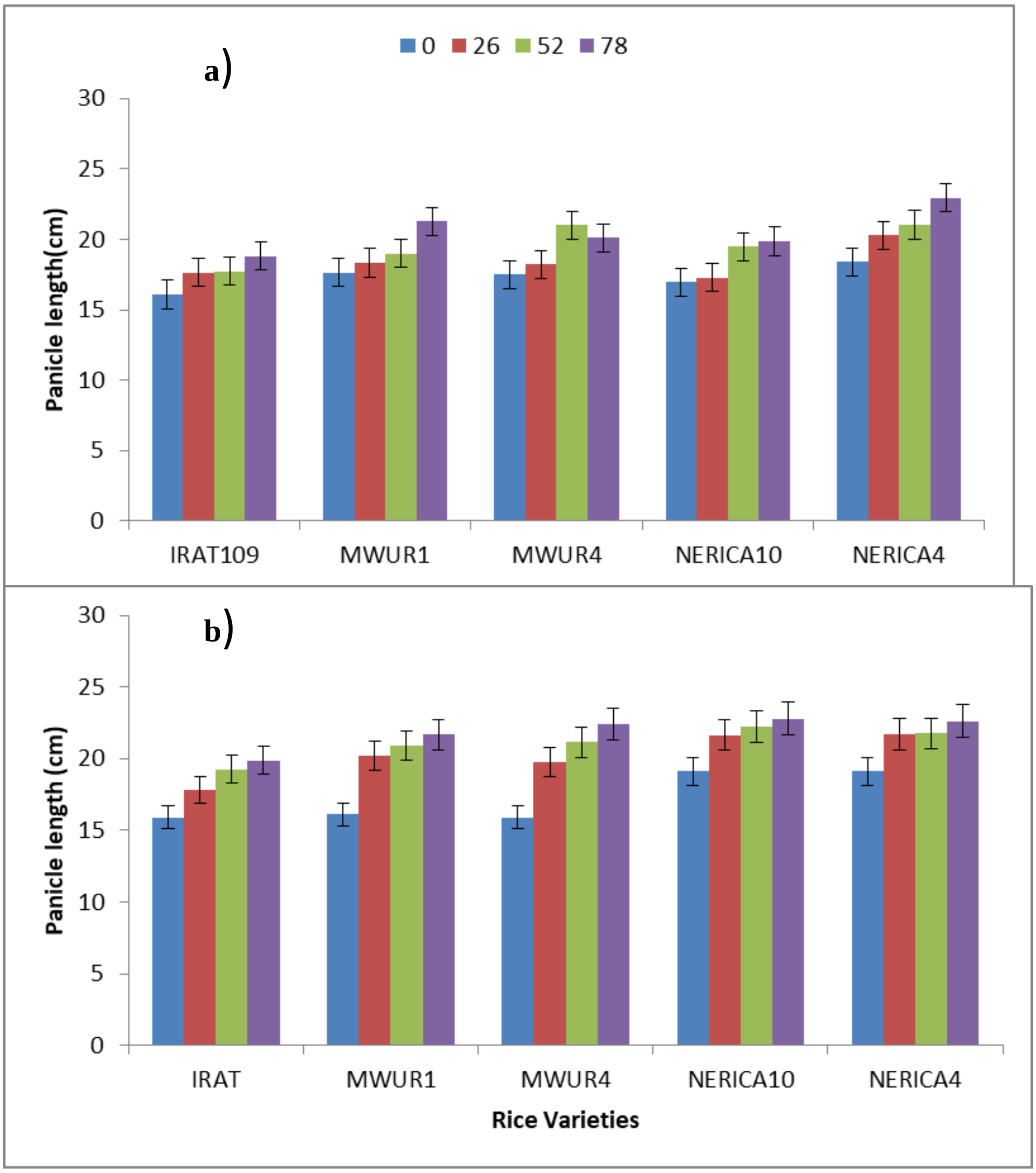


Appendix 7: Effects of N-level application on upland rice filled grain ratio season one and

two



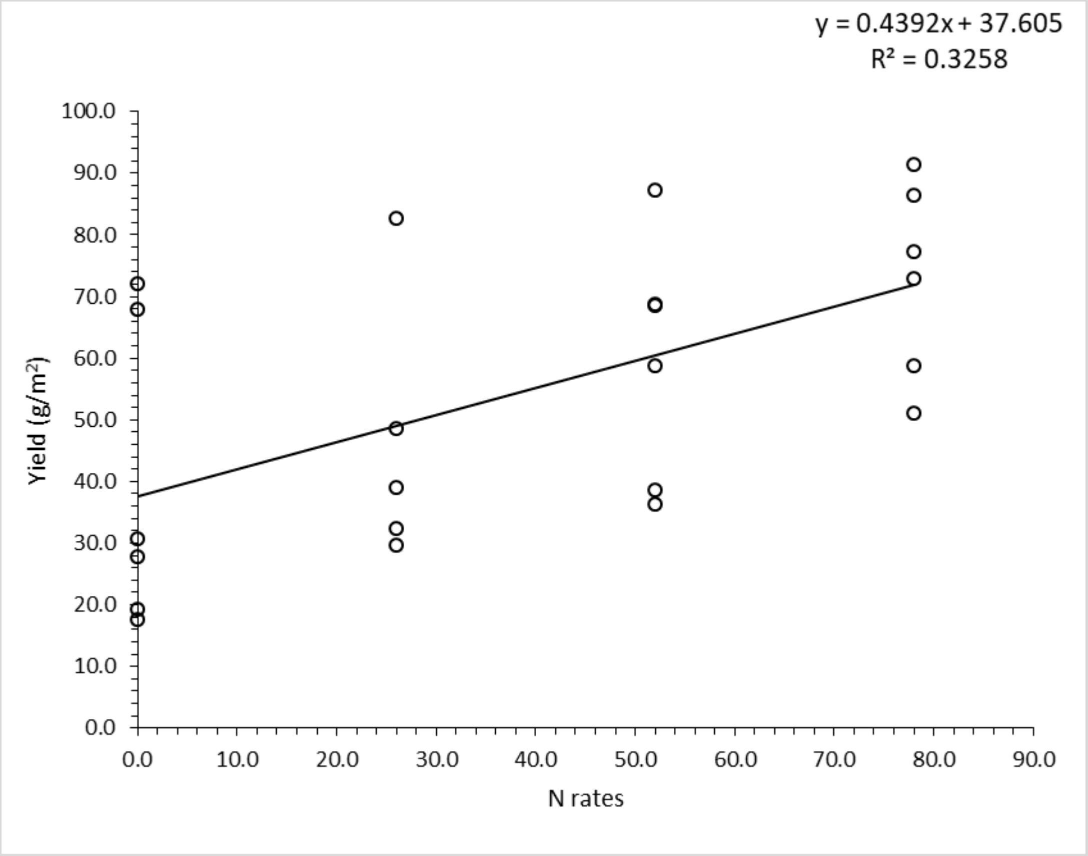
Appendix 8: Effect of N-level on rice panicle length, season one and two



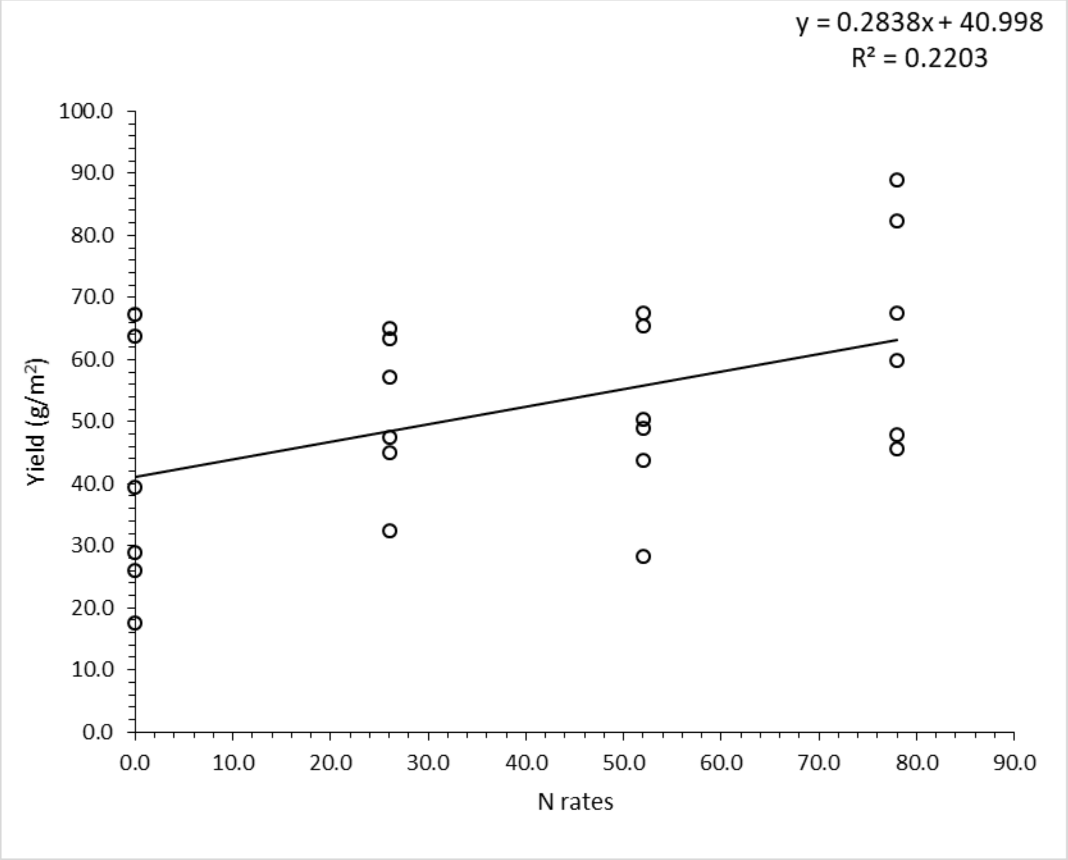
APPENDIX9: Results of Analysis of Variance of studied variables.

		CL	**PL**	**PN**	**FGW**	**SDW**	**RDW**	**TGN**	**1000GW**	**GPP**	**FGR**	**HI**	**AGW**	**R/S ratio**
Season 1	Nitrogen (N)	***	***	***	***	***	**	***	ns	*	**	*	***	***
	Variety (V)	***	***	*	***	***	ns	***	***	***	***	***	**	**
	N x V	Ns	*	ns	*	ns	ns	*	ns	*	ns	ns	ns	*
Season 2	Nitrogen (N)	***	***	***	***	***	***	***	**	***	*	**	***	***
	Variety (V)	***	***	*	ns	***	ns	**	***	***	***	***	**	***
	N x V	**	Ns	Ns	ns	***	**	ns	ns	ns	**	*	ns	***

APPENDIX 10: Regression analysis of mean grain yield vs. nitrogen rates (MWUR 1)



APPENDIX 11: Regression analysis of mean grain yield vs. nitrogen rates (NERICA 4)



APPENDIX 12: Effects of nitrogen rates on grain yield and yield components of five upland rice varieties

Variety	Trt	P.Ht (cm)	Number Panicles	Panicle Length	Culm Length	Grain wt/plant	1000 grain wt	Harvest Index (%)	Shoot Dry wt	Root dry wt
MWUR1		98.6a	8.9a	19.1b	77.0a	18.2a	33.8a	50.7b	18.1a	0.77bc
MWUR4		93.0ab	8.0ab	19.2b	67.9b	15.6ab	31.3b	54.2ab	13.0b	0.85ab
NERICA4		90.5b	7.6b	20.6a	66.0bc	17.1a	27.5c	56.1a	13.3b	0.98ab
NERICA10		84.0c	6.9b	18.4a	58.9c	11.8bc	25.4d	55.5a	9.1c	0.73c
IRAT 109		80.6c	7.4b	17.5c	45.9d	10.4c	33.3a	41.5c	13.9ab	1.03a
	0N	74.2d	5.6c	17.3b	47.3d	8.0c	29.7b	50.0a	7.7d	0.70c
	26N	85.9c	7.9b	18.3b	59.7c	13.2b	29.7b	52.6a	11.7c	0.82bc
	52N	93.4b	8.2ab	19.7a	66.7b	16.0b	30.7a	51.2a	15.3b	0.94ab
	78N	103.9a	9.3a	20.6a	78.7a	21.3a	30.8a	52.6a	19.3a	1.04a
Var.		***	*	***	***	**	***	*	***	*
Trt.		***	***	***	***	***	*	ns	***	**
Var× Trt		ns	ns	ns	ns	ns	ns	ns	ns	ns
CV (%)		7.7	19.6	6.9	13.4	34.3	4.1	8.0	30.	23.3

Values with the same letter(s) within the column are not significantly different