

**DIALLEL ANALYSIS OF EXTRA EARLY MAIZE (*Zea mays* L.)
INBRED LINES FOR DROUGHT AND LOW NITROGEN
TOLERANCE**

BY

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OCTOBER, 2016.

KWAME NKRUMAH UNIVERSITY OF SCIENCE AND TECHNOLOGY

KUMASI, GHANA

COLLEGE OF AGRICULTURE AND NATURAL RESOURCES

DEPARTMENT OF CROP AND SOIL SCIENCES

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**A THESIS SUBMITTED TO THE DEPARTMENT OF CROP AND SOIL
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OF

MASTERS OF PHILOSOPHY IN PLANT BREEDING

BY

ADEJUMOBI IDRIS ISHOLA (B.Agric. Plant Science)

OCTOBER, 2016.

DECLARATION

This research was carried out at the breeding field (Finatrade Farm of Animal Science) under the close supervision of Prof. Richard Akromah and Dr. Daniel Nyadanu of the Department of Crop and Soil Sciences, Kwame Nkrumah University of Science and Technology (KNUST), Kumasi, Ghana.

The research presented in this thesis is the original work carried out by the author and has never been submitted for any degree or diploma in the University or any other University in the past.

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ABSTRACT

The importance of maize has extended beyond human consumption into livestock feeds and industrial uses in sub-Saharan Africa. However, increasing drought and infertile soils (Low-N) have become important abiotic stresses affecting maize production. Varieties with improved water use efficiency and/or nitrogen would be beneficial for subsistence farmers via reduced production cost. Unfortunately, few breeding programs focus attention on breeding for low N and drought stress conditions. Hence, little is known about the genetic control and heritability to drought and low-N tolerance in maize. The study sought to improve maize production and productivity in the low rainfall zones of Ghana. Five parental inbred lines TZEEI-6, TZEEI-21, TZEEI-29, TZEEI-38, and TZEEI-76 were crossed in a complete diallel mating design and the resulting F1 hybrids were evaluated in four environments (optimal-N and low-N, well-watered and induced drought stress). The performance of hybrids varied significantly in different environments with respects to yield and contributing characters. Non-additive gene action controlled grain yield under low-N stress and optimal-N conditions and biomass yield under drought stress and well-watered conditions. Additive gene action controlled anthesis-silking interval, leaf senescence, leaf chlorophyll content, and ears per plant under low-N stress. Under managed drought stress, additive gene action was only important for days to silking, anthesis-silking interval, and tassel blasting. The inbred lines TZEEI-76 and TZEEI-21 were the best combiners for grain and biomass yield under low-N and drought stress. The best combiner under low-N stress for anthesis-silking interval was TZEEI-38, for plant height and leaf senescence was TZEEI-76, and for leaf chlorophyll content was TZEEI-29. The best combiner under drought stress for anthesis-silking interval, leaf senescence, leaf chlorophyll content, and tassel blasting was TZEEI-21. Desirable SCA effects were found for the crosses TZEEI-21×TZEEI-38, TZEEI-21×TZEEI-76, TZEEI-

29×TZEEI-76, and TZEEI-38×TZEEI-76 under both stressed and non-stress conditions. Hybrid crosses TZEEI-6×TZEEI-29, TZEEI-6×TZEEI-76, and TZEEI-21×TZEEI-29 had desirable SCA estimates under drought stress. Hybrid crosses TZEEI-29×TZEEI-38 and TZEEI-29×TZEEI-6 had desirable SCA estimates for low-N. The cross TZEEI-29×TZEEI-38 had the highest heterosis under low-N stress while TZEEI-29×TZEEI-21 had the highest heterosis under drought stress. The narrow sense heritability of secondary traits; anthesis-silking interval, leaf senescence, and leaf chlorophyll content were higher under stressed conditions than non-stressed conditions. Significant correlation ($p<0.05$) was observed between yield and secondary traits; anthesis-silking interval, leaf senescence, and leaf chlorophyll content under low-N and drought stress environments. Across stress environments, leaf chlorophyll content and leaf senescence were significant ($p<0.001$). Hybrids selection combining tolerance to both stress conditions in order to stabilize yield on farmers' field should concentrate on combining mean yield performance with positive leaf chlorophyll content and negative leaf senescence.

DEDICATION

This work is dedicated to Almighty Allah for making it a successful beginning to higher grounds, my lovely parents (Mr. and Mrs. Adejumobi), fiancée (Habibah) and siblings (Ilyas, Fatimah, Muyinat, and Sekinah).

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LIST OF ABBREVIATIONS AND ACRONYMS

ANOVA	Analysis of Variance
TZEEI	Tropical <i>Zea</i> Extra Early Inbred
IITA	International Institute of Tropical Agriculture
CRI	Crops Research Institute
OPV	Open Pollinated Variety
N	Nitrogen
F ₁	First filial progeny
Low-N	Low nitrogen stress environment
Opt-N	Optimal nitrogen environment
Dro-str	Drought stress environment
WW	Well-watered environment
DA	Days to 50 % Anthesis
DS	Days to 50 % Silking
ASI	Anthesis-silking interval
PLHT	Plant height
EHT	Ear height
LFS	Leaf senescence
CHL	Leaf chlorophyll content index
HUSKC	Husk cover
MSVD	Maize streak virus disease
PASP	Plant aspect
EASP	Ear aspect
EPP	Ear per plant

EL	Ear length
EDIAM	Ear diameter
EWT	Ear weight
EPO	Ear position
GY	Grain yield
1000KW	One thousand kernel weight
LFR	Leaf rolling
TB	Tassel blasting
BYLD	Biomass yield
YLD	Yield
ARL	Average root length
RBR	Root branching
DRW	Dry root weight
MOI	Grain moisture content
SCA	Specific Combining Ability
GCA	General Combining Ability
P	Parental inbred lines
H	Hybrid combination/crosses

CHAPTER ONE

1.0 INTRODUCTION

Maize (*Zea mays* L.) belongs to the crop family called *Poaceae*. Maize is a vital cereal crop for global food security, as its importance is seen in the wide economic uses ranging from direct human consumption to processing into other food materials, animal feeds, and industrial products. It is ranked first among the cereal with an annual global production of one billion metric tons (FAOSTAT, 2014). On the African continent, maize is grown primarily for its carbohydrate-rich kernel and provides staple food for more than 300 million people (Banziger and Diallo, 2001) that reside both in urban and rural locations. In sub-Saharan Africa, maize is essentially regarded as an important food and feed crop, as it provides energy and protein for both human and livestock (Diallo *et al.*, 2001; Ngaboyisonga, 2008). In West and Central Africa (WCA), maize provides 15 % of the total caloric intake (Badu-Apraku *et al.*, 2011a). In addition to the highlighted importance, maize also serves as a means of generating revenue through agricultural export of the crop within the Africa regions (Asea, 2005). An even higher demand is projected in these regions as population growth rate and livestock feed requirement increase.

In Ghana, maize is a central cereal crop widely cultivated with a national mean yield of 1.7 tons/hectare (MoFA, 2011), and consumed as a staple food as the population grows since 1965 (FAO, 2006). Therefore, increasing maize productivity will be an adequate means to ensure food security in Ghana. Maize cultivation in Ghana is largely by smallholder resource-poor farmers whose cultivation is mainly dependent on rainfall (SARI, 1996).

Successful maize production is, however, dependent on the influence of both biotic and abiotic factors which constitute an extensive range of production constraints. Abiotic factors

play a key role in determining the success of maize production. Of these limiting factors, the most important are the increasing drought incidence and infertile soils mainly nitrogen deficiency (Edmeades *et al.*, 2006; Meseke *et al.*, 2008).

In the tropics, drought results from increasing climate change and reduction in the water-holding capacity of the soil leading to high evaporation rate. Drought and low nitrogen stresses jointly occur frequently in the tropical region (Nyombayire *et al.*, 2011). This results in substantial yield losses or complete crop failure under severe conditions. Maize is usually cultivated in the savannah regions of West and Central Africa which are characterized by high temperature and relative humidity ideal for maize plant growth and development. However, the soils of this region are low in organic matter/nitrogen resulting from high leaching rate, denitrification, decomposition, erosive action of rainfall, continuous cropping, removal of crop residues, and use of low inputs (Adekayode and Ogunkoya, 2010).

With the increasing unpredictability of global climate pattern usually in the amount and distribution of precipitation, the use of extra-early maturing maize cultivars has become a means by which farmers use to reduce the risks. Extra early maturing maize provides options concerning relay crops, intercropping, late planted crops, drought escape and earlier harvest (CIMMYT-Zimbabwe, 2000).

Drought stress can occur at any development stage of the crop causing a varying magnitude of implications. When drought occurs few days to flowering, it can cause grain yield reduction by 90 % (Setter and Parra, 2010). When it occurs during flowering it is capable of causing grain yield reduction by 50 % (Durães *et al.*, 2002). When drought occurs during grain filling, it causes yield reduction by 17 % (Edmeades *et al.*, 1992). Drought affects vast expanse of total tropical agricultural land (about 60 %) and causes a yield reduction of maize

over large expanses annually (Pingali, 2001). These losses estimate to 16 million tons of maize grain (Banziger and Diallo, 2001). Supplementary irrigation would offer a means to solving drought problem so as to improve maize productivity in drought predisposed areas (Boyer and Westgate, 2004; FAO, 2006; Derera *et al.*, 2008). However, the bulk of tropical farmers do not have the means to this, due to various barriers (Banziger and Diallo, 2001; FAO, 2006).

Nitrogen is a limiting nutrient in the tropical agro-ecologies coupled with sub-optimal nitrogen conditions that characterize a considerable percentage of maize production in these areas by small-scale farmers (Banziger *et al.*, 2004). Nitrogen deficit is a common phenomenon in tropical soils with the exception of newly cleared land (Bellon, 2001), and rated to have a significant contribution to yield reduction (Banziger and Lafitte, 1997; Banziger and Diallo, 2001; Diallo *et al.*, 2001). This means that adequate nitrogen to provide an optimum growing condition for maize plant should be provided through fertilizer application. However, chemical fertilizers are expensive and, hence, unaffordable to the vast majority of small-holder farmers in many African countries (Banziger *et al.*, 2004).

There exists only a handful of maize varieties that can tolerate drought and low-Nitrogen stress in tropical Africa (Nyombayire *et al.*, 2011) and tropical maize continues to be exposed to variable forms of drought and nitrogen stresses. These periodic stresses in the production environs has led to far-reaching efforts by research institutes such as International Institute of Tropical Agriculture (IITA), and Centro Internacional De Mejoramiento De Mais Y Trigo (CIMMYT) to develop maize germplasm that exhibits tolerance to drought stress and/or nitrogen deficit stress (Banziger and Cooper, 2001; Edmeades *et al.*, 2006). The germplasm is made available as homozygote (inbred) lines from where varying hybrids and synthetic varieties with tolerance to drought and/or low-N stresses can be constituted. The genetic

control and heritability of tolerance to low nitrogen and drought stresses in this germplasm (inbred lines) need to be identified. However, little information is available on the genetic control of tolerance to drought and low-N stresses. Resource poor farmers could be aided by understanding the genetic control of tolerance to drought and low-N stresses in developing maize varieties with high nitrogen utilization efficiency and drought tolerant ability in maize producing areas of Ghana.

In view of this, the main objective of this research was to improve maize production and productivity in the low rainfall zones of Ghana.

Specifically, this study sought to

1. Evaluate growth and yield of hybrids from extra early inbred lines under low nitrogen and drought stresses.
2. Determine the general and specific combining ability (GCA and SCA) from crosses of extra early inbred lines.
3. Determine the nature of gene action for yield selected agronomic traits of extra early maize hybrids under stress and non-stress conditions.
4. Estimate heritability and heterosis for yield under stressed and non-stressed conditions in selected extra early inbred lines.
5. Determine the relationship between grain yield and selected agronomic characters.

CHAPTER TWO

2.0 LITERATURE REVIEW

Maize (*Zea mays* L.) belongs to the grass family called *Poaceae* which is widely accepted to have originated from teosinte (*Zea mexicana*) in Mexico and Guatemala. Maize is a cereal crop with wide environmental adaptation. It is cultivated in all agro-ecology where it is used for numerous purposes such as direct human consumption, livestock feed, and agro-industrial raw materials. In spite of the economic importance, the productivity of maize in sub-Saharan Africa is largely affected by abiotic stress conditions particularly drought and low-N stresses (Banziger *et al.*, 2006). There is, therefore, an urgent need to increase the productivity of maize crop through selection/breeding for stress tolerant genotypes in order to stabilize yield and meet the increasing demand for food as well as feed in developing countries.

2.1 Drought stress and maize plant

Drought is rated the most important abiotic stress that limits maize productivity in Africa (Banziger *et al.*, 1997). Badu-Apraku *et al.* (2011a) reported that recurrent drought is the cause of frequent food shortage in WCA. Drought effect on maize can be seen at different developmental stages starting from seedling emergence or establishment up to grain filling. The physiological responses of maize to drought stress are a complex phenomenon and often unpredictable (Moreno *et al.*, 2005). The productivity of a maize plant is dependent upon the presence or absence of drought stress at three critical developmental periods. The first being crop establishment, followed by flowering phase, and lastly, grain filling phase. However, the yield is most severely affected when drought stress strikes during the flowering and grain filling period (Banziger *et al.*, 2000; Setter and Parra, 2010). Under severe drought stress at flowering, the kernels number, and the anthesis-silking interval (ASI) are affected (Setter and

Parra, 2010; Boyer and Westgate, 2004). Extreme sensitivity is observed at 2 to 22 days after silking, with a peak at 7 days. At the peak, kernels number may be reduced to 45 % of their potential and ASI is extended (Setter and Parra, 2010). When the crop is stressed during the period of 12 to 16 days after silking, kernel weight may fall to 51 % of potential weight. Drought stress during the two weeks bracketing flowering may cause yield loss up to 90 %, causing almost complete barrenness (Setter and Parra, 2010).

Maize is prone to drought stress at flowering phase than any other cereal crop due to the floral biology. Unlike many other cereal crops, the monoecious nature of plant exposes both the male and female reproductive structure to the desiccative environment during pollination. When drought occurs few days to flowering, silk growth is apparently delayed owing to the reduction in photoassimilate production. The delayed silk growth results in increased ASI and ineffective pollination (Banziger *et al.*, 2000) Even though appreciable quantities of photo-assimilates are accumulated and stored in the secondary storage organ (stem) prior to flowering, the developing ear could not fully access these reserves. The inability of the developing kernel to access the photoassimilate reserve in the stem is principally due to the diminished functioning of the enzyme called acid invertase (Boyer and Westgate, 2004; Setter and Parra, 2010). This proves why successful pollination in drought-stressed maize is often followed by kernel abortion few days after pollination.

2.1.1 Physiological effects of drought stress on maize plant

Drought affects varying physiological activities which are essential for proper growth and development of maize plant. Abscisic acid (ABA) production level increases as drought stress intensify. ABA is produced from the below ground part of the plant and transported to the above ground part where it controls varying physiological responses of maize to water stress such as rolling of leaves, leaf senescence acceleration and stomata closure (Banziger

et al., 2000). This occurs prior to reduction of leaf turgor by hydraulic mechanisms. ABA as a hormone only aid the plant survival during drought stress but does not have any impact on production. During drought stress, cell expansion is inhibited and. this is evident as reduced leaf area, delayed silk emergence, and reduced plant size. When the severity of drought becomes high, affected organs may lack the cell for expansion even when the stress is alleviated (Banziger *et al.*, 2000).

At the cellular level, plants generally produce osmotically active substance in response to drought. This allows plants to take up more water from the soil in order to maintain their turgor and also allow cell functioning for a longer period of time (Bolanos and Edmeades 1991). Drought stress also affects the photo-assimilates production of a maize plant. This is evident in Photosystem II than Photosystem I. During this period, the linkage between the two photosystems is broken and hence, there is a free flow of high energy electrons in the leaf. These free flowing electrons result in chlorophyll oxidation expressed phenotypically as leaf bleaching (Banziger *et al.*, 2000). This differs from leaf senescence in that photosynthetically active leaves (above the ear) are affected. Hence, there is a loss of photoassimilate production capacity. Enzyme activity is generally reduced under drought.

2.1.2 Effects of drought on maize at the whole plant level

Drought stress following seeding results in poor plant stands establishment as the soil dries out. When drought strikes after plant stand establishment, the vegetative phase suffers in terms of morphological characters such as reduced leaf area, plant height, and acceleration of leaf senescence from the leaves below the ear. Senescence brought about by photo-oxidation can occur leading to the reduced interception of solar energy for photoassimilate production (Banziger *et al.*, 2000). The closure of leaf stomata by the plant in an attempt to reduce water loss through evapotranspiration consequently reduce photosynthetic capacity.

Therefore, assimilate partitioning to growing sinks is affected. When drought occurs at flowering phase, silk growth is delayed resulting in an extended ASI. Abortion of the ear and grains increases and may cause total barrenness of the plant which translates to total grain yield loss. The silk is more seriously affected as compared to the male counterpart, though tassel blasting becomes more pronounced when temperatures exceed 38°C (Banziger *et al.*, 2000). The below-ground effects include the increase in root/shoot ratio but as the stress intensifies and become severe, the growth of the root also decreases consequently affecting nutrient uptake. When drought stress coincides with linear grain growth, remobilization of stem reserves can occur leading to early lodging.

2.2 Low soil nitrogen (N) and maize plant

Nitrogen is an essential element necessary for plant growth and development. Maize growth is highly sensitive to soil nitrogen variation and tropical soils are characterized by low soil nitrogen (Banziger and Diallo, 2004). Nitrogen stress reduces photo-assimilates production in the leaf via a reduction in leaf chlorophyll, leaf area and increased rate of senescence. Nitrogen plays a significant role in leaf chlorophyll formation and hence, determine the photosynthetic efficiency of the plant. This is an indication of nitrogen being a determinant of yield (Banziger *et al.*, 2000). In spite of the importance of nitrogen to maize, the availability in the soil is controlled by many factors and hence, not always available for the plant uptake, for example, nitrogen can be locked in soil organic matter making it unavailable for the plant use (Banziger *et al.*, 2000). Soil N is available to maize and other non-leguminous plants as nitrate (NO_3^-) and ammonium (NH_4^+), known as mineral N. These are made available through mineralization of organic matter in the soil and fertilizer application. However, N predominant form is as nitrate, formed by the conversion of NH_4^+ to NO_3^- by nitrifying bacteria (Vance, 2001). Maize respond to N stress in varying ways such as reduced

plant size and radiation use efficiency, remobilization of stem N to the growing sinks, etc. (Vance, 2001). Presterl *et al.* (2002) reported nitrogen deficit as a cause of delayed leaf area expansion and shoot growth but increases the growth of the root. This is likened to drought stress effect. Severe N stress typically delays silking than do for anthesis. This extends the ASI. Similar to drought stress, the delay in silk emergence also causes ineffective pollination and, kernel and ear abortions under N stress. When N stress occurs during grain filling, it increases the rate of leaf senescence through remobilization and reduces the rate of photoassimilate production and kernel weight. According to Banziger, *et al.* (2000), the yield of maize is affected by nitrogen supply more strongly before anthesis than after anthesis.

Additionally, low-N stress induces premature leaf yellowing (first stage of senescence) though reversible. Banziger *et al.* (2000) reported that mineralization of soil N is less than 1kg/ha/day which may be sufficient for the crop at seedling stage as mineral N exceeds uptake capacity of the plant but as the crop progresses to vegetative stage through growth, N need of the crop increases to about 4 to 5 kg/ha/day. Since the rate of net N mineralization does not increase with the crop growth rate, the crop is therefore exposed to N stress.

Soil N deficit can be improved via inorganic fertilizer application to enhance the crop growth and development. The only challenge is the expensive nature of inorganic fertilizer which makes it a choice for only a handful of farmers who can afford it and apply at the required rate. This financial challenge affects the majority of the resource poor farmers and consequently reduce maize productivity in many developing countries. Larsson (2005) reported a survey conducted in the sub-Saharan part of Africa in the year 2000-2002 that 53 % of small-scale farmers applied 0 kg/ha of fertilizer while the rest 47 % who applied actually applied at the average rate of 14 kg/ha which is far below the requirement of maize crop for optimal yield as compared to commercial farmers who apply an average of 300 - 400 kg/ha.

It is, therefore, essential to initiate a breeding approach to developing N stress tolerant varieties as a recommendation to increase productivity under low input conditions.

2.2.1 Nitrogen stress and maize development

Maize growth and development show a similar pattern to N stress irrespective of the location, unlike drought stress. During the early growth stage (seedling), the N requirement of the crop is far less than the available soil N from mineralization but as the growth progresses, the crop N requirement increases until it matches up the rate of mineralization of soil N and even beyond (Banziger *et al.*, 2000). This phenomenon is usually observed at the vegetative phase of the crop, during this phase there is a need to meet the N deficit gap that has been created in the soil due to the crop demand exceeding the rate of N supply in the soil, using external means such as inorganic fertilizer application. Failure to do this usually results in N stress to the plant. In general, maize plant tries to adjust to N stress to some extent by remobilizing the N reserve in older leaves (below the ear) that contribute very little to photoassimilate production. This action does not significantly influence final crop yield but may affect some yield-determining components depending on the time of occurrence (Banziger *et al.*, 2000). When N stress occurs prior to flowering, affects leaf area development, photosynthetic rate, and ear spikelets. When it occurs during flowering, enhances kernel and ear abortion. When it occurs at grain filling phase, leaf senescence is accelerated, and kernel weight becomes reduced (Banziger *et al.*, 2000).

2.3 Breeding for maize genotypes tolerant to low-N and/or drought stresses

Maize breeding strategies in tropical Africa has been influenced by maize breeding in the temperate environment (Banziger *et al.*, 2000). In the temperate environs, maize cultivation is done under stress-free condition by farmers but in the tropics, maize production is affected

by varying kinds of stresses with additive effect in some cases such as drought and low soil N combined. The latter results in farmers yield being significantly different from that obtain at the research station or demonstration plot. This implies that selection under high yielding condition may not be the best strategy to improve maize productivity in farmers' field.

2.3.1 Breeding methods for drought tolerance

Breeding for tolerance to drought can be viewed from two breeding approaches. The first approach being escape mechanism which involves selection of genotypes with the ability to complete the critical phases of development in their life cycle before drought ensued, hence the name extra early and early genotypes. However, earliness is often subject to a penalty in yield when the rainfall amount and duration are higher than average. With respect to these conditions, the yield of extra early and early maturing genotypes is dependent on the amount of radiation captured for photoassimilate production during their cycle. This yield is usually less than that of late maturing genotypes (Banziger *et al.*, 2000).

The second approach is to select for drought-tolerant maize genotypes. This is achieved by the introgression of drought tolerant genes for better adaptation of the genotype to drought prone growing environs. However, selection for both high yield and drought tolerance in maize is hampered by (i) difficulty in controlling the condition of stress (ii) large yield variation which is not ideal for selection purpose in trials conducted in uncontrolled drought predisposed environs (iii) large error variances when trials are tested in drought stress environs, and (iv) high G x E as a result of different genotype characteristics or environmental variations from one location to another as well as year differences. (Banziger and Cooper, 2001).

Using the CIMMYT drought tolerant breeding program as a case study, maize genotypes were evaluated under drought stress at flowering, grain filling and under optimal conditions. The selection under drought was based on an index that combines increased grain yield, stem elongation, leaf area development, reduced ASI, leaf senescence as well as canopy temperature. For the optimal environment, index combines constant ASI and grain yield (Bolanos and Edmeades, 1993). Gain from selection under drought was principally due to the large variability in the genotypes ability to partition photoassimilate into the growing sinks even as water uptake and biomass production rate remain constant (Monneveux *et al.*, 2006).

The understanding of maize physiological functions reveals that certain characteristics which may not be important under optimal environmental conditions play a significant role in determining final yield under drought stress condition such as the development of ear with grains when drought stress strikes at flowering phase (Diallo *et al.*, 2003). For selection for drought tolerance, trials are established during the off-season when precipitation is very low with supplemental irrigation to provide the water requirement based on the crop need. The main aim is to estimate the genotypic value of drought tolerance in such experiment (Musila *et al.*, 2010). Many of the drought tolerant germplasm of CIMMYT were assessed in the environment where they were selected, with the hypothesis that gains from selection is restricted to the drought pattern in the environment (Banziger *et al.*, 2004). But the improved stability of yield has been achieved across different testing locations for a selected drought tolerant population when compared to the conventionally selected ones. Improvements for drought tolerance was associated with gains from selection across variable nitrogen levels (Banziger *et al.*, 2000). This shows that the use of managed drought stress is a better screening method for drought tolerance selection. (Banziger *et al.*, 2004).

2.3.2 Breeding approaches for low-N tolerance

A significant quantity of maize production in the tropical regions is carried out on soils with low-N condition. Many smallholder farmers either cannot afford inorganic N sources due to high cost or access it due to unavailability or do not find the returns from N use necessary to justify its cost. In addition, production of organic N source (manure) is not efficient for these farmers. Breeding for maize genotypes that combine the high efficient use of N with high yield potential is an effective means to reduce smallholder farmers' expenditure on fertilizer (Joseph *et al.*, 2001; Meseke *et al.*, 2006). Findings have shown that maize genotypes performances vary with soil fertility levels and the plant's N use efficiency (Scharf *et al.*, 2002). Commercial maize varieties selected for increased yield in the tropics are selected under optimum N fertility conditions and may not meet the interests of resource-poor farmers who cannot afford inorganic fertilizer. Hence, the need to develop maize varieties tolerant to N stress.

The main obstacle in improving tolerance to N stress is that of successful selection. It is important for a breeder to have the appropriate source of germplasm. Following this is the ability to evaluate tolerance to N accurately under relevant conditions to the target area and the intensity of selection. To achieve this requires a better knowledge of the crop's behaviour under low-N environment, ability to impose suitable levels of stress, a set of secondary traits related to yield under N stress, the use of appropriate experimental designs, proper germplasm choice and breeding strategy (Banziger *et al.*, 2000). Improvement of genotypes for low-N tolerance requires less breeding stress when compared to improvement for drought tolerance as N stress affect the growth of plant more evenly than compared to drought stress (Banziger *et al.*, 2000). Hence, evaluating genotypes under a single level of severe N deficit is sufficient to assess the level of tolerance to low-N stress for variable levels of soil N deficit.

Badu-Apraku *et al.* (2013) reported that genotypes with drought tolerance capability also show to a significant degree, tolerance to low soil N stress. Banziger *et al.* (1999) reported that improvement for drought tolerance using recurrent selection leads to correlated improvement for low-N tolerance.

A strategy to reduce the effect of N stress on maize productivity is the selection of cultivars that use available N more effectively, due to their enhanced capacity for its uptake as well as the use for grain production (Lafitte and Edmeades, 1994).

2.4 Relationships between grain yield and key secondary traits under drought and

Low-N

Most maize in tropical regions, particularly in sub-Saharan Africa are grown under drought and low-N stress (Diallo *et al.*, 2001). This justifies the need for efficient breeding strategies for such conditions. In breeding for improved genotypes, breeders are primarily concerned with selection for increased grain yield but selection for grain yield under stress condition may introduce some level of bias as heritability that determines genetic advance under selection changes with stress pattern and severity (Banziger *et al.*, 2000). Likewise, Bolanos and Edmeades (1993) observed that the use of grain yield alone as a measure of a genotype improvement for stress tolerance is inefficient, but the use of secondary traits that are highly correlated to grain and as well possess high heritability will help to increase selection efficiency. Secondary traits are beneficial as they strengthen the ability to identify genotypes with genetic potentials to tolerate drought and/ low-N stresses. This is made possible as the heritability of some secondary traits remains high even under stressed conditions which is not possible for grain yields (Banziger and Lafitte, 1997). Banziger *et al.* (2000) highlighted some secondary traits that can guide selection for both drought and low-N tolerance in addition to grain yield. Some of these traits overlap for both drought and low-N stresses such

as anthesis-silking interval, leaf senescence and ears per plant. In general, secondary traits can be used to develop selection index for improving maize genotype for drought and/or low-N stress.

2.5 Hybrid maize genotypes

Hybrid maize cultivars, produced by crossing contrasting inbred genotypes can offer a means for stabilizing yield and ensuring food security. This is mainly attributed to the advantage of hybrid vigor. In the production of hybrid cultivars, the most important consideration is the choice of inbred lines used in various cross combination using a designed system of mating in order to evaluate their combining ability. Rojas and Sprague (1952) divided the performance of a line into two categories namely – general combining ability (GCA) and specific combining ability (SCA).

2.5.1 Combining ability

Allard (1960) defined combining ability as a measure of the genetic potential of any genotype based on the performance of their offspring produced in some definite mating system. Combining ability cannot be predicted based on the phenotypic value of the parents but assessed only by progeny testing using a designed mating system. Parental plants that produce vigorous progeny in a cross combination are said to have a good combining ability. Combining ability analysis is of great importance in cross-pollinated species such as maize as it helps to identify parents that can be used to produce hybrids as well as synthetic cultivars (Amiruzzaman *et al.*, 2011) Combining ability was formally used to classify inbred lines relative to their crosses performances but was later refined into two expressions namely; General Combining Ability (GCA) and Specific Combining Ability (SCA) according to Sprague and Tatum (1942).

The mean phenotypic performance of a genotype in series of crosses with other genotypes is termed the general combining ability (GCA) of that genotype. Specific combining ability (SCA) on the other hand is the specific cross combination whose performance is better or worse than the average performance of the parents in combination. GCA is usually a measure of the additive gene action and SCA is a measure of the non-additive gene action (Aguilar *et al.*, 2003). Rojas and Sprague (1952) reported that SCA variance contains deviations attributable to genotype by environment interaction as an addition to those from dominance and epistasis effects. GCA and SCA estimates of genotypes can be obtained using the analysis of variance method for different mating designs such as diallel. Lines with high GCA are said to be good combiners and can be used in the development of synthetic variety more effectively. However, when specifically high yielding combinations are desired such as in hybrid production SCA serves as better means for parental selection.

2.5.2 Diallel mating design

Varietal development using cross-breeding involves the use of a definite mating design and one of the most powerful of such mating design is diallel. The diallel mating design involves the selection of a group of b inbred lines (as parents) with crosses/mating made among the selected lines. The procedure results in a maximum of b^2 combinations (complete diallel mating). Data from these combinations is presented in $b \times b$ table where; x_{ii} represents the mean phenotypic performance for i^{th} genotypes, x_{ij} as the mean phenotypic value of the F_1 resulting from the cross of the i^{th} and j^{th} parental lines and x_{ji} symbolizes the reciprocal crosses (Griffing, 1956). A population can easily attain Hardy-Weinberg equilibrium with the use of the diallel mating system. The diallel mating system has proved very effective in genetic research for determining the inheritance of important traits among genotypes, investigating the GCA of the parents, identifying superior parents for hybrid cultivars development,

categorizing inbred genotypes into various heterotic groups and for identifying appropriate testers for breeding purpose (Bhatnagar *et al.*, 2004; Menkir *et al.*, 2003; Gethi and Smith, 2004; Yallou *et al.*, 2009). According to Griffing (1956), four diallel methods exist and they differ in the omission of parents or reciprocals in the crosses. The number of progeny for each method from I to IV are b^2 , $\frac{1}{2}b(b + 1)$, $b(b - 1)$ and $\frac{1}{2}b(b - 1)$ respectively. Nevertheless that the diallel mating system is known to be very powerful in genetic research, it may be challenging in cases where the inbred lines involved are many.

2.5.3 Heterosis and hybrid development in maize

Heterosis refers to the increase in performance (size and growth rate) of an offspring over the parental genotypes. Heterosis in crop plants can be seen as increased grain yield, reduced flowering period etc. (Duvick, 1997). Shull (1952) explains heterosis as increased size, vigor, productivity and developmental rate, resistance to pest and disease, and tolerance to climatic stresses manifested in the organism from crossbreeding. Heterosis can be of two types based on the hybrid performance standard namely: mid-parent heterosis and better parent heterosis. According to Falconer and Mackay (1996), mid-parent heterosis explains the performance of a hybrid relative to the mid-performance of the parental lines. Better parent heterosis otherwise called heterobeltiosis explains the performance of a hybrid relative to that of the better parent. Hybrid maize production has increased around the world due to heterosis in order to meet the increasing demands.

Badu-Apraku *et al.* (2011b) evaluated some groups of extra-early inbred and single cross genotypes under drought stress, low-N and optimal conditions in four locations. The result indicated that difference in grain yields between the single cross hybrids and their parental inbred increases as drought intensity increases indicating heterosis. Alam *et al.* (2008) reported that inbred lines are low yielding as compared to hybrids due to a high level of

heterosis for yield in hybrid genotypes. Hybrids can yield as much as three times the yield of their parental inbred lines but superior hybrids genotypes from farmers' point of view are not necessarily genotypes with high heterosis (Duvick, 1997).

2.5.4 Gene action

Gene action simply refers to how the genes (hereditary unit) operates to determine the phenotypic expression of a character. Gene action may be additive or non-additive in nature. Breeders prefer additive gene action to non-additive in that it is responsible for the transfer of character from the parents to the offspring. Gene action controlling yield with respect to tolerance to drought and low-N stresses has been reported in many genetic studies but reports of these have been inconsistent. To report a few, Betran *et al.* (2003) and Katsantonis *et al.* (1998) observed the dominance of non-additive gene action for hybrid maize grain yield under low-N and the dominance of additive gene action under high N. On the contrary, Below *et al.* (1997) and Kling *et al.* (1997) observed the dominance of additive gene action under low-N and non-additive gene action under high N conditions. Guei and Wassom (1992) also observed the dominance of non-additive gene action for hybrid maize grain yield under drought but Betran *et al.* (2003) and Gethi and Smith (2004) observed additive gene action for grain yield under drought in their studies.

Gene action is deduced from the variances of general and specific combining abilities (Singh, 2003). GCA mean square expresses additive variance while SCA mean square expresses the non-additive variance which includes dominance mainly and all forms of epistatic interaction components provided epistasis is existing. Heterosis shown by hybrids may not be mainly attributed to dominance effect of the genes but also due to epistasis. Hence, the failure of wide crosses in showing heterosis may be attributed largely to the epistatic interaction of the

genes (Goodnight, 1997). Goodnight (1997) simply defined epistasis as the condition where one gene masks the effect of another gene for expression.

CHAPTER THREE

3.0 MATERIALS AND METHODS

3.1 Experimental site

The study was conducted at the Finatrade farm (Animal Science Department) and Plant house (Mechanization) of the Kwame Nkrumah University of Science and Technology (KNUST) Kumasi- Ghana: lat. 6.40°, long. 1.37°, 300 meters above sea level (masl) and 1400 mm annual rainfall. The experimental site is in the forest zone with haplic Alisols (Jones *et al.*, 2013).

3.2 Germplasm

A selected group of extra early inbred lines from International Institute of Tropical Agriculture (IITA) consisting of eight (8) genotypes (TZEEI-4, TZEEI-6, TZEEI-15, TZEEI-18, TZEEI-21, TZEEI-29, TZEEI-38, and TZEEI-76) previously evaluated for drought stress tolerance in various environments constituted the germplasm for this study. Table 3.1 shows the inbred lines and their source and tolerance to drought.

3.2.1 Development of F₁ generation

The inbred lines were planted out on 5 m, 6 rows plots with a planting spacing of 0.75 m x 0.4 m between and within rows respectively at the Finatrade farm (Animal Science) of the KNUST in the major season (April, 2015). Prior to planting, land preparation was done by ploughing, followed by harrowing to produce soil with good tilth. At planting, three seeds were sown per hole and thinned to two after seedling establishment at two weeks after planting (WAP). Compound fertilizer (NPK 15:15:15) was applied at 10 days after planting (DAP) at 12 g/hill (60 kg/ha) and top-dressed with Urea 46:0:0 at 30 DAP at 30 kg/ha to achieve 90:60:60 of N:P:K. Pava insecticide (Lambda cyhalothrin) was applied at 2 weeks

after planting (WAP) and 4 WAP at the rate of 2.0 L/ha. Fulan 3 % G (Carbofuran) was applied at the rate of 2 kg/ha at 6 WAP into the whorls of each plant to prevent stem borer attack. At flowering, silks and tassels were well covered for ease of controlled pollination. Hand pollination using the complete diallel mating design (Griffing, 1956) method I was used to produce the F₁'s consisting of single crosses, reciprocals and parental inbred lines (selfing). Out of the whole cross combinations generated, crosses from three inbred lines; TZEEI-4, TZEEI-15, and TZEEI-18 were omitted from the genotypes evaluation due to poor germination, poor stand establishment, and poor tassel and silk development. The resultant combinations provided 25 genotypes consisting of 20 hybrid crosses and 5 inbred parents from selfing. These crosses, inbred parents and two local checks (Abontem and Dodzi) were evaluated for low-N and drought stresses tolerance from October, 2015 to January, 2016.

Table 3.1 Description of the germplasm used in the diallel study to evaluate the performance of extra early maize lines for drought and low-Nitrogen tolerance.

Parent	Pedigree	Source	Property
TZEEI-4	TZEE-WSRBC5x1368STRS7Inb.85	IITA	white, dent, drought tolerant
TZEEI-6	TZEE-WSRBC5x1368STRS7Inb.100	IITA	white, dent, drought tolerant
TZEEI-15	TZEE-WPopxLDS6(Set A)Inb.44	IITA	white, dent, drought tolerant
TZEEI-18	TZEE-WPopxLDS6(Set A)Inb.49	IITA	white, dent, drought tolerant
TZEEI-21	TZEE-WPopxLDS6(Set B)Inb.44	IITA	white, dent, drought tolerant
TZEEI-29	TZEE-WSRBC5x1368STRS7Inb.27	IITA	white, dent, drought tolerant
TZEEI-38	TZEE-WSRBC5x1368STRS7Inb.92	IITA	white, dent, drought tolerant
TZEEI-76	TZEE-YSRBC1x9450STRS6Inb.8B	IITA	white, dent, drought tolerant
Dodzi	Commercial variety	CRI	OPV, white, dent, drought tolerant
Abontem	Commercial variety	CRI	OPV, yellow, dent, drought tolerant

Source: Badu-Apraku *et al.* (2011b)

TZEEI- Tropical Zea Extra-Early Inbred; CRI- Crops Research Institute

IITA- International Institute of Tropical Agriculture

3.3 Evaluation of F₁ progeny

3.3.1 Low nitrogen (N) evaluation

The experiment was carried out in two environmental blocks, a block for the low-N evaluation whose performance was compared to the optimal N block meant as a control experiment. Low-N evaluation was achieved by the application of N-fertilizer at the rate of 45 kg/ha as against optimal N of 90 kg/ha. Prior to the establishment of trials for evaluation, soil samples from the field were analyzed to ascertain the initial N level of the field. This was done to provide an idea of how much depletion has been suffered by the soil in nitrogen (N) as a result of continuous cropping of maize on the land. The low-N block soil analysis results showed 0.11 % of N, 15.26 mg/kg of P and 0.08 cmol/kg of K in the top 30 cm soil (Table 4.1).

3.3.1.1 Field layout

Land preparation began with the application of roundup (Glyphosate) herbicide at the rate of 4.0 litre/ha in the minor season of 2015. Ploughing of the field followed using a tractor mounted heavy disc plough. Harrowing was done after ploughing to provide a good tilt for seedling growth and establishment. The field was laid out in a Randomized Complete Block Design (RCBD) with three replications for low-N as well as optimal N. Planting of the trials was done on 7th October, 2015 with each plot consisting of two rows, each 5m long, spaced at 0.75 m between rows and 0.40 m within rows. Two to three seeds were sown per hill depending on the seed quantity available per genotype.

3.3.1.2 Fertilizer application and water management

Compound fertilizer (NPK 15:15:15) was applied at 10 days after planting (DAP) at the rate of 9 g/hill to achieve 45 kg/ha targeted low-N rate. Phosphorus (P) and potassium (K) were

kept constant at the recommended rate of 60 kg/ha for both low-N and optimal N experiments. Since the application of the compound fertilizer only gave 45 kg/ha of both phosphorus and potassium, the remaining 15 kg/ha was obtained from the application of straight fertilizers; Muriate of potash for potassium (K) at the rate of 0.75 g/hill and Triple superphosphate for phosphorus (P) at the same rate. The optimum-N condition received compound fertilizer at the recommended rate of 12 g/hill to meet an application rate of 60 kg/ha of N as well as P and K. In order to meet the targeted recommended rate of 90 kg/ha of N, the optimal N plants were top dressed with urea (46: 0: 0) at the rate of 30 kg/ha. The top dress application was done at 32 DAP due to insufficient soil moisture content during the period (Appendix 1). Supplemental irrigation using overhead sprinkler system was later used to support the plant development to prevent further drought stress.

3.3.1.3 Weed management

During the experiment, both the low and optimal N conditions were kept weed free via herbicide application supplemented with hoe weeding. A mixture of Atrazine (Atrazine WP) and Gramoxone (Paraquat) was applied at the rate of 4.5 and 1.0 L/ha respectively as pre-emergence to control the weed. Supplemental hoe weeding was used to keep the two experimental blocks weed free after flowering till harvest.

3.3.1.4 Pest management

The prevalence of stem borer (*Buseola fusca*) attack during the minor season maize cultivation necessitated the use of Pawa insecticide (Lambda cyhalothrin) at 2 WAP and 4 WAP at the rate of 2.0 L/ha. Fulan 3 % G (Carbofuran) was applied at the rate of 2 kg/ha at 6 WAP into the whorls of each plant. Due to late planting of the trial which extended the

experiment through the harmattan period, losses from rodent attack (Grand Squirrel) was minimized using numerous control measures such as trap setting, baiting etc.

3.3.1.5 Harvesting

Harvesting of ears was done at physiological maturity (86 DAP). Twenty plants were harvested per plot or genotype. Harvested ears were bagged in labeled large brown envelopes and transported to the plant house for data collection on yield and yield parameters. Grain moisture was adjusted to 15 %. Grain yield was estimated from ear weight by using an assumed shelling percentage of 80 %. Grain yield per plot, as well as yield per hectare basis, were extrapolated from the harvest data using the formula below;

$$\text{Grain yield (t/ha)} = EWT * \left[\frac{(100 - MOIST)}{85} \right] * (10000 * SHELL) \dots\dots\dots eqn. 1$$

Where; EWT = ear weight per genotype

MOIST = grain moisture content per genotype

SHELL = shelling percentage per genotype

3.3.2 Drought stress evaluation

Evaluation for drought was done at the plant house (Mechanization) following the assumption of unpredictability in the rainfall pattern of the agroecology (rain forest). The hybrid crosses, parental inbred lines and local checks (20+5+2 = 27 genotypes) were evaluated under controlled drought stress condition and adequate moisture growing condition. The evaluation was done in a bucket experiment to make a simulation of natural drought possible through artificial means at the preferred period (5 WAP).

3.3.2.1 Plant house management

Two hundred and sixteen (216) 20-litre buckets each filled with 24 kg of well-drained black loamy soils were used for the study. The soil was purchased and sterilized prior to filling the buckets to prevent possible infection of soil-borne pathogens. The buckets were perforated below to allow drainage of water and were arranged into two moisture variation blocks (drought stress block and well-watered block) with each block consisting of 108 buckets after filling with soils. Each of the 27 treatments was replicated in fours arranged in a Completely Randomized Design (CRD). Fulan 3 % G (Cabofuran) was immediately applied to the soil to eliminate all possible surviving insects and nematode after sterilization.

3.3.2.2 Water stress management

Adequate initial watering was done to the soils by the application of 4 litre of water per bucket and left for 24 hours to allow drainage. This was done to simulate field capacity moisture level. Seeds were treated with dress force chemical (Imidacloprid + Metalaxyl-M + Tebuconazole) to prevent soil-borne infections. Sowing of the seeds was done after 24 hours of initial watering at four seeds per bucket and thinned to two seedlings at 10 days after planting (DAP) when seedlings have fully established. Watering was done at the rate of 0.6 litre/bucket/3days (modified from Akinwale *et al.*, 2016) until 3 WAP when it was increased to 1.5 liters/bucket/3days. The increase in water application came about as a result of the increasing temperature averaging 38.29 °C observed in the plant house during first 8 weeks into the experiment (Appendix 1). The temperature increase could have been enhanced by the nature of the structure of the plant house. Drought stress was imposed at 35 DAP on the drought block experiment by the withdrawal of water and the plants were left to depend on the available soil water throughout the flowering period (tassel emergence and silk initiation).

During the evaluation, tassel blasting was observed in both experimental blocks. Leaf bleaching (photo-oxidation) were also observed in many of the genotypes (both drought and well-watered). At 70 DAP, watering was resumed to support the growth of the developing grains that were formed during drought stress. The total drought period observed for the experiment was five weeks.

3.3.2.3 Fertilizer application, weed and pest management

Application of compound fertilizer NPK 15:15:15 was done at 400 kg/ha rate at 10 DAP and later top dressed with urea at 26 DAP to meet the recommendation of 90:60:60 kg/ha of nitrogen, phosphorous and potassium respectively. Weeding was done by hand picking alone when needed. In the drought-stress evaluation, Fulan 3 % G (Cabofuran) was solely applied into the whorls of the plants at 2 WAP and 4 WAP to prevent stem borer (*Buseola fusca*) attack.

3.3.2.4 Harvesting

Harvesting was done at 89 DAP. About 60 % of the well-watered conditioned trial had ears without kernels or kernel being immature. About 90 % of the drought stress trial had the same problem. Due to this challenge, biomass yield was used in place of grain yield at harvest. The plants were harvested with care to prevent root damage, root systems were detached from the above ground mass. The above ground mass and root system were weighed freshly to obtain their fresh weights and packaged in separate envelopes for oven drying at 75 °C. Drying was done for 72 hours after which the oven dried above ground mass and root system samples were weighed to obtain their dry masses (biomass/ dry matter content).

3.4 Collection of agronomic data

In the low-N study, data were collected in an alternate manner on 10 plants per plot on phenological attributes; days to 50 % anthesis (DA) and days to 50 % silking (DS) from where anthesis-silking interval (ASI) was estimated. Morphological attributes; plant height (PLHT), ear height (EHT), leaf senescence I and II (LFS) at 56 and 70 DAP, leaf chlorophyll content index (CHL), husk cover (HUSKC), maize streak virus disease score (MSVD) and plant aspect (PASP). Yield parameters; ear aspect (EASP), ears per plant (EPP), ear length (EL), ear diameter (ED), ear weight (EWT), grain yield per hectare (GY), thousand kernel weight (1000KW) and grain moisture content (MOI) of all the genotypes used for the study.

In the drought stress study, data were collected on phenological and morphological attributes. Leaf rolling (LFR) and tassel blasting (TB) were included and leaf senescence was taken once. Biomass yield (BYLD) was used in place of grain yield due to lack of ear with fully developed grains in many genotypes. Data were also taken on root system consisting of root length (ARL), root branching (RBR), and root biomass (DRW). Table 3.2 shows the data that were taken and how they were taken.

Table 3.2 List of data collected and procedure for collection for evaluation of extra-early maize lines for drought and low-N tolerance

Traits	Abbreviation	Units	Procedure
50% anthesis	DA	days	number of days from planting to 50% of plants having begun pollen shed
50% silking	DS	days	number of days from planting to 50% of plants showing silk emergence
Anthesis-silking interval	ASI	days	the difference between 50% silking and 50% anthesis dates (ASI = DS – DA)
Plant height	PLHT	cm	distance from the base of the plant to where tassel branching begins
Ear height	EHT	cm	distance from the base of the plant to the node bearing the upper ear
Leaf chlorophyll content	CHL	cci	Chlorophyll content meter (CCM 200-plus)
Maize streak virus disease score	MSVD	1-5	1= no symptoms on leaves, 2 = light disease symptoms on 20 to 40 % leaf area, 3 = moderate symptoms on 40 to 60 % leaf area, 4 = severe symptoms on 60 % of leaf area, 5 = severe symptoms on 75 % or more of the leaf area.
Plant aspect	PASP	1-5	Overall plant phenotype where; 1= excellent phenotype; 2=good phenotype; 3=average phenotype; 4= Bad phenotype; and 5= worst phenotype.
Tassel blasting	TB	0-1	Only for drought as percentage of plants with blasted tassel per genotype

Table 3.2. Cont'd

Traits	Abbreviation	Units	Procedure
Leaf senescence	LFS	1-10	1 = 10% dead leaf area; 2 = 20% dead leaf area; 3 = 30% dead leaf area; 4 = 40% dead leaf area; 5 = 50% dead leaf area; 6 = 60% dead leaf area; 7 = 70% dead leaf area; 8 = 80% dead leaf area; 9 = 90% dead leaf area; 10 = 100% dead leaf area
Ears per plant	EPP	0-1	The number of ears with at least one full grain harvested as a ratio of total maize crop harvested
Ear length	EL	cm	Length of the cob in centimeter (cm)
1000 grain weight	1000KW	g	Weight of a thousand grain
Grain yield	GY	t/ha	Calculated from ear weight using assumed shelling percentage of 80% per plot adjusted to 15% moisture content
Leaf rolling	LFR	1-5	1 = unrolled, turgid; 2 = leaf rim starts to roll; 3 = leaf has a the shape of a V; 4 = rolled leaf rim covers part of leaf blade; 5 = leaf is rolled like an onion
Biomass yield	BYLD	t/ha	The weight of oven dried above ground mass
Root biomass	DRW	g	The weight of oven dried below ground mass
Root length	ARL	cm	Length in centimeter (cm) of the primary root

3.5 Statistical analysis of data

Data collected under the field and plant house growing conditions were tested for normality and homogeneity of variance using PBTools version 1.4 (IRRI, 2014). Data were analyzed using PROC GLM in SAS (SAS institute, 2001), according to Griffing's (1956) method 1, Model 1. Estimate of the combining ability of the genotypes was done using the same software. The relative importance of additive and non-additive genetic effects (GCA and SCA, respectively) was estimated according to Baker (1978). Least square mean was used to estimate mid-parent and better parent heterosis according to Rai (1979). The variance component method was used to estimate genetic component and heritability according to Teklewold and Becker (2005).

3.5.1 Analysis of variance (ANOVA)

3.5.1.1 Individual analysis of variance

The variance estimate of each variable was done independently for field evaluation as well as screen house evaluation based on ANOVA format presented in Table 3.3. The GCA and SCA were computed from the mean square of the treatments based on Griffing method I model I where genotype was considered as fixed effects and replication was considered as random effect as shown below;

$$Y_{ijk} = \mu + g_i + g_j + s_{ij} + r_{ij} + b_k + e_{ijk} \dots\dots\dots eqn. 2$$

Where; μ is the overall mean, g_i is the general combining ability effect of the i^{th} parent, g_j is the general combining ability of the j^{th} parent. s_{ij} is the specific combining ability effect (interaction effect) of the i^{th} and j^{th} parents, r_{ij} is the reciprocal effect, replication, b_k is the effect of k^{th} replications and e_{ijk} is the error.

Table 3.3 Format for individual analysis of variance using Griffing's method I

Sources of variation	DF	MS	EMS
Genotypes (G)	$g-1$	MS_G	$\sigma_e^2 + r\sigma_g^2$
General combining ability (GCA)	$b-1$	MS_g	$\delta^2_e + 2b \left(\frac{1}{b-1}\right) \sum g_i^2$
Specific combining ability (SCA)	$b(b-1)/2$	MS_s	$\delta^2_e + \left[\frac{2}{b(b-1)}\right] \sum_i \sum_j s_{ij}^2$
Reciprocal effect (R)	$b(b-1)/2$	MS_r	$\delta^2_e + 2\left[\frac{2}{b(b-1)}\right] \sum_{i<j} \sum r_{ij}^2$
Error	$(g-1)(r-1)$	MS_E	δ^2_e
Total	$gr-1$		
CV (%)			
GCA/SCA			
Mean			

Source: Adapted from Griffing's (1956)

Where, r = number of replication; g = number of genotypes; b = the number of parents; MS_G = mean square due to genotypes; MS_g = mean square of GCA; MS_s = mean square of SCA; MS_r = mean square of reciprocal; MS_E = mean square of error; σ_e^2 , σ_g^2 and σ_r^2 are variances due to error, genotypes and replications respectively.

3.5.1.2 Combined analysis of variance

A two-factor analysis of variance was undertaken using Diallel-SAS procedure developed by Zhang *et al.* (2005), according to Griffing's (1956) method 1, Model 1. This Diallel-SAS program evaluated main genotype effects containing GCA, SCA, reciprocal, and their respective interaction with environment using the model below where genotype remained fixed with replication and environment being random.

$$Y_{ijk} = \mu + g_i + g_j + s_{ij} + r_{ij} + l_k + (gl)_{ik} + (gl)_{jk} + (sl)_{ijk} + (e)_{ijk} \dots\dots\dots 3$$

Where; Y_{ijk} is the observation of parents i and j in environment k ; μ is the general mean; g_i and g_j are the general combining ability effect of parents i or j ; s_{ij} is the specific combining ability of effect of the cross between parents i and j ; r_{ij} is the effect of the reciprocal crosses between parents j and i ; l_k is the effect due to location k ; $(gl)_{ik}$ and $(gl)_{jk}$ interaction effect

between general combining ability of parents i or j with location k ; $(sl)_{ijk}$ interaction effect between specific combining ability of cross ij and location k ; and $(e)_{ijk}$ = error attributed to observations ijk .

Table 3.4 Format for combined ANOVA using Griffing's method I

Source of variation	DF	MS	EMS
Environment (E)	$e-1$	MS_E	$\delta^2_e + r\delta^2_{GE} + rg\delta^2_E$
Genotype (G)	$g-1$	MS_G	$\delta^2_e + r\delta^2_{GE} + rE\delta^2_G$
General combining ability (GCA)	$b-1$	MS_g	$\delta^2_e + 2b \left(\frac{1}{b-1}\right) \sum g_i^2$
Specific combining ability (SCA)	$b(b-1)/2$	MS_s	$\delta^2_e + \left[\frac{2}{b(b-1)}\right] \sum_i \sum_j s_{ij}^2$
Reciprocal effect (R)	$b(b-1)/2$	MS_r	$\delta^2_e + 2\left[\frac{2}{b(b-1)}\right] \sum_{i<j} \sum r_{ij}^2$
G x E	$(g-1)(e-1)$	MS_{GE}	$\delta^2_E + r \delta^2_{GE}$
GCA x E	$(b-1)(e-1)$	MS_{gE}	$\delta^2_E + r \delta^2_{gE}$
SCA x E	$b(b-1)(e-1)/2$	MS_{sE}	$\delta^2_E + r \delta^2_{sE}$
R x E	$b(b-1)(e-1)/2$	MS_{rE}	$\delta^2_E + r \delta^2_{rE}$
Error	$(ge-1)(r-1)$	MS_e	δ^2_e
Total	$ger-1$		
CV (%)			

Source: Modified from Griffing's (1956)

MS_E = mean square due to environment; MS_G = mean square due to genotype; MS_{GE} = mean square due to genotype x environment; MS_e = error mean square; δ^2_E = environment variance; $\delta^2_{G|}$ = genotype by environment variance; $\delta^2_{g|}$ = general combining ability by environment variance; $\delta^2_{s|}$ = specific combining ability by environment variance; $\delta^2_{r|}$ = reciprocal by environment variance; δ^2_e = error variance; r = number of replications; e = number of environments and g = number of genotypes.

3.5.1.3 Estimation of combining ability, heritability and heterosis

Mean squares due to general combining ability, specific combining ability and the interaction of general combining ability by environment and specific combining ability by environment were used to estimate the additive and dominance variances across stressed environments,

optimal environments, contrasting N environments and moisture variation environments. Narrow and broad sense heritability were determined according to Teklewold and Becker (2005) and genetic ratio was estimated according to Baker (1978).

Table 3.5 Estimation of genetic variance and heritability

Genetic parameters	Symbols	Determination
Additive variance	δ^2_A	$2 \delta^2_{gca} = 2 \left[\frac{MS_{gca} + MS_{scaenv}}{MS_{sca} + MS_{gcaenv}} \right]$
Dominance variance	δ^2_D	$\delta^2_{sca} = \frac{MS_{sca}}{MS_{scaenv}}$
Narrow sense heritability	h^2	$\frac{\delta 2A}{\delta 2A + \delta 2D + \left(\frac{\delta 2A}{Env} \right) + \left(\frac{\delta 2D}{Env} \right) + \left(\frac{\delta 2e}{REnv} \right)}$
Broad sense heritability	H	$\frac{\delta 2A + \delta 2D}{\delta 2A + \delta 2D + \left(\frac{\delta 2A}{Env} \right) + \left(\frac{\delta 2D}{Env} \right) + \left(\frac{\delta 2e}{REnv} \right)}$
Genetic ratio		$\frac{2MS_{gca}}{2MS_{gca} + MS_{sca}}$

Source; Teklewold and Becker (2005)

δ^2_{gca} and δ^2_{sca} = variances of general and specific combining abilities estimates; δ^2_e = error variance; MS_{gca} and MS_{sca} = mean squares of general and specific combining ability; MS_{gcaenv} and MS_{scaenv} = mean squares of the interaction between general and specific combining ability and environment respectively; Env = number of environment; R = number of replicate per environment

Heterosis was estimated using the least square means. Mid and high parent heterosis were estimated using the formula below;

$$MPH = \frac{H - MP}{MP} \times 100\% \dots \dots \dots \text{eqn. 4}$$

Where, MPH = mid-parent heterosis; H= F1 hybrid phenotypic performance and MP = mid or average parents phenotypic performance and calculated as;

$$MP = \frac{P1 + P2}{2} \dots \dots \dots \text{eqn. 5}$$

$$\text{HPH} = \frac{H-HP}{HP} \times 100\% \dots\dots\dots \text{eqn. 6}$$

Where, HPH = high parent heterosis; H= F1 hybrid phenotypic performance; HP = high parent phenotypic performance.

The student *t*-statistics was used to determine the statistical difference of F1 hybrid means and the mid-parent and better parent means according to Wynne *et al.* (1970) adapted from Iqbal *et al.* (2009).

$$\text{Mid-parent heterosis (t}_1\text{)} = \frac{F1 - MP}{\sqrt{(3/2r)(EMS)}} \dots\dots\dots \text{eqn. 7}$$

$$\text{Better parent heterosis (t}_2\text{)} = \frac{F1 - BP}{\sqrt{(2/r)(EMS)}} \dots\dots\dots \text{eqn. 8}$$

Where; t₁ and t₂ = calculated t-statistics values compared to t-statistics tabulated for significant differences

MP = mid-parent mean performance for the specific F1 hybrid cross ($\frac{P1 + P2}{2}$)

BP = better parent mean performance for the specific F1 hybrid cross (P1 or P2)

r = number of replication

EMS = error mean square from ANOVA component for specific character in question

3.5.1.4 Correlation coefficient estimation

Genotype means were used to compute Pearson correlation coefficients between the agronomic parameters measured during the study and the yields observed (grain and biomass) within individual experiments and across related environments combined. Yield data across drought and low-N environments were then pooled together to perform centered

scattered plot for the hybrid crosses and local checks to identify the best hybrid genotype that combines drought and/or low-N tolerance ability with high yield.

CHAPTER FOUR

4.0 RESULTS

4.1 Nutrient status of experimental site prior to planting

Result of initial soil nutrient characteristics of the experimental blocks at Finatrade Farm in the minor season of 2015 was presented in Table 4.1. Nutrients status, in accordance with Landon (2014) interpretation of soil nutrients analysis, were generally low in the experimental blocks except for phosphorus level. Nitrogen level was considered low in both blocks since the amount less than 0.5 % were recorded, hence it is expected that results obtained in the study would reflect the true response of genotypes to nitrogen applied externally.

4.2 Crop performance under optimal nitrogen environment (opt-N)

In the Opt-N environment, significant ($p < 0.001$) differences were observed in the performances of the hybrids and the parental inbred lines for all the traits except for LFS where significant ($p < 0.01$) differences was found and EPP where there was no significant difference among the genotypes (Table 4.2). The mean entry grain yield observed was 3.68 t/ha and 4.00 t/ha for the local checks respectively (Table 4.2). When the entry was decomposed into hybrids and parental inbred lines, the mean grain yield observed for the hybrids was 4.17 t/ha while the inbred lines recorded an average of 1.78 t/ha (Table 4.3). The best hybrid crosses under Opt-N condition were TZEEI-29×TZEEI-38, TZEEI-76×TZEEI-21, TZEEI-38×TZEEI-76, and TZEEI-21×TZEEI-76 with average grain yield greater than 5 t/ha and the best inbred line was found to be TZEEI-38 with average grain yield of 2.03 t/ha (Table 4.3). Combining ability analysis revealed the presence of significant additive and non-additive gene actions influencing the selected traits (Table 4.2).

Table 4.1 Soil characteristics of nitrogen trial site at Finatrade Farm of Animal Science Department of KNUST Kumasi

Block	Sample	pH	Total N	Avail P	K	Ca	Mg	Na	Org. C	Org. matter
			%	mg/kg	-----cmol/kg-----			-----%-----		
Low-N	T1	5.79	0.113	13.72	0.14	1.50	0.24	0.88	2.47	4.26
	T2	5.76	0.104	16.77	0.07	1.42	0.36	0.43	2.41	4.16
	T3	5.74	0.125	12.98	0.07	1.38	0.24	0.26	2.41	4.16
	T4	5.79	0.101	17.56	0.04	1.24	0.42	0.30	2.17	3.75
	Mean	5.77	0.11	15.25	0.08	1.38	0.31	0.46	2.36	4.08
Optimum-N	M1	5.83	0.106	8.72	0.12	2.56	0.48	0.88	2.29	3.96
	M2	5.83	0.100	16.77	0.03	1.44	0.56	0.19	1.98	3.40
	M3	5.84	0.117	4.12	0.05	1.42	0.36	0.43	2.57	4.44
	M4	5.90	0.115	9.41	0.03	1.56	0.46	0.08	2.49	4.30
	Mean	5.85	0.11	9.76	0.06	1.75	0.47	0.39	2.33	4.03
Landon ratings	High	> 6.5	> 0.5	> 50.0	> 0.6	> 10.0	> 4.0	> 1.0	> 10.0	-
	Low	< 5.8	< 0.2	< 15.0	< 0.2	< 4.0	< 0.5	< 1.0	< 4.0	-

T1-T4= Top slope soil samples; M1-M4= Middle slope soil samples; pH= Soil hydrogen ion concentration; Total N= Total soil Nitrogen; Avail P= Available soil Phosphorus; K= Soil Potassium; Ca= Soil Calcium; Mg= Soil Magnesium; Na=Soil Sodium; Org. C= Soil organic Carbon; and Org. matter= Soil organic matter.

Table 4.2 Mean squares for grain yield and other agronomic traits evaluated under optimal nitrogen environment

Source	DF	Mean squares									
		DS	ASI	PLHT	LFS	CHL	EPP	EDIAM	EL	1000KW	GY
		Days	days	cm	0-1	cci		mm	cm	g	t/ha
Entry	24	15.06***	0.49***	1177.52***	0.17**	192.65***	0.15	0.60***	9.58***	1135.72***	4.95**
GCA	4	1.75	1.33***	1159.82**	0.55***	396.79***	0.17	0.20	7.13*	831.14*	4.01
SCA	10	28.28***	0.40*	1777.73***	0.10	202.82***	0.12	1.19***	18.82***	1788.41***	9.14***
REC	10	7.15	0.26	584.38	0.08	100.82***	0.16	0.17	1.32	604.87*	1.12
Error	48	4.97	0.17	289.76	0.07	18.48	0.11	0.16	2.13	247.95	1.91
CV (%)		4.07	19.59	15.87	13.76	11.24	18.45	12.57	13.05	10.65	35.73
GCA/SCA		0.06	3.33	0.65	5.50	1.96	1.42	0.17	0.38	0.47	0.44
Mean		54.80	4.25	107.30	3.33	38.25	1.74	3.16	11.17	147.81	3.68
Min		49.00	0.00	56.00	1.67	16.50	1.00	1.64	5.62	92.00	0.92
Max		62.00	9.00	161.30	6.67	56.50	2.90	4.10	15.28	205.00	7.45
CHM		54.83	5.00	114.90	3.70	41.73	1.52	3.54	12.10	165.83	4.00

*, **, *** indicates significance at 0.05, 0.01, and 0.001 probability levels respectively; CHM indicates checks mean.

DS= days to 50% silking; ASI= anthesis-silking interval; PLHT= plant height; LFS= leaf senescence; CHL= chlorophyll content of leaf; EPP= ears per plant; EDIAM= ear diameter; EL= ear length; 1000KW= weight of a thousand grain; and GY= grain yield

Table 4.3 Yield loss of all genotypes as influenced by drought stress, low nitrogen stress and combination of drought and low nitrogen stress

Entry	BYLD			GY			ANC	ASC	% Loss
	WW	MD	% Loss	ON	LN	% Loss			
TZEEI-6	3.07	0.89	71.01	1.87	0.83	55.61	2.47	0.86	65.18
TZEEI-21	2.93	1.00	65.87	1.75	1.53	12.57	2.34	1.27	45.94
TZEEI-29	2.45	1.00	59.18	1.62	0.91	43.83	2.04	0.96	53.07
TZEEI-38	2.65	1.00	62.26	2.03	0.50	75.37	2.34	0.75	67.95
TZEEI-76	2.33	1.65	29.18	1.61	1.52	5.59	1.97	1.59	19.54
TZEEI-6×TZEEI-21	4.06	1.68	58.62	3.11	3.04	2.25	3.59	2.36	34.17
TZEEI-6×TZEEI-29	4.28	1.89	55.84	4.24	3.27	22.88	4.26	2.58	39.44
TZEEI-6×TZEEI-38	4.10	1.84	55.12	4.22	3.00	28.91	4.16	2.42	41.83
TZEEI-6×TZEEI-76	3.89	2.44	37.28	3.65	3.33	8.77	3.77	2.89	23.47
TZEEI-21×TZEEI-29	4.37	2.53	42.11	3.40	2.55	25.00	3.89	2.54	34.62
TZEEI-21×TZEEI-38	4.44	2.48	44.14	4.93	3.22	34.69	4.69	2.85	39.17
TZEEI-21×TZEEI-76	4.73	2.48	47.57	5.04	4.12	18.25	4.89	3.30	32.45
TZEEI-29×TZEEI-38	3.80	1.49	60.79	5.89	3.30	43.97	4.85	2.40	50.57
TZEEI-29×TZEEI-76	4.13	2.38	42.37	4.27	3.73	12.65	4.20	3.06	27.26
TZEEI-38×TZEEI-76	4.25	2.67	37.18	5.10	3.30	35.29	4.68	2.99	36.15
TZEEI-21×TZEEI-6	3.78	1.99	47.35	2.82	2.11	25.18	3.30	2.05	37.88
TZEEI-29×TZEEI-6	4.10	1.95	52.44	2.45	1.45	40.82	3.28	1.70	48.09
TZEEI-29×TZEEI-21	4.01	2.90	27.68	3.38	2.87	15.09	3.70	2.89	21.92
TZEEI-38×TZEEI-6	3.95	1.77	55.19	4.37	2.45	43.94	4.16	2.11	49.28
TZEEI-38×TZEEI-21	4.14	2.58	37.68	4.37	3.75	14.19	4.26	3.17	25.62
TZEEI-38×TZEEI-29	4.00	1.35	66.25	4.53	3.02	33.33	4.27	2.19	48.77
TZEEI-76×TZEEI-6	3.95	2.17	45.06	3.19	2.62	17.87	3.57	2.40	32.91
TZEEI-76×TZEEI-21	4.56	2.17	52.41	5.42	4.82	11.07	4.99	3.50	29.96
TZEEI-76×TZEEI-29	4.36	2.10	51.83	4.64	4.32	6.90	4.50	3.21	28.67
TZEEI-76×TZEEI-38	4.06	2.39	41.13	4.45	3.06	31.24	4.26	2.73	35.96
CHECK-1	2.26	1.60	29.20	4.23	2.56	39.48	3.25	2.08	35.90
CHECK-2	2.80	1.46	47.86	3.78	2.50	33.86	3.29	1.98	39.82
Mean	3.76	1.92	48.99	3.72	2.73	27.45	3.74	2.32	38.73
Min	2.26	0.89	27.68	1.61	0.50	2.25	1.97	0.75	19.54
Max	4.73	4.73	71.01	5.89	4.82	75.37	4.99	3.50	67.95
PM	2.69	1.11	57.50	1.78	1.06	38.59	2.23	1.08	50.34
HM	4.15	2.16	47.90	4.17	3.17	23.61	4.16	2.66	35.91

BYLD= biomass yield; GY=grain yield; WW= well-watered condition; MD= managed drought stress; LN=low-N stress; OP=optimal-N condition; ANC= across all normal condition; ASC= across all stress conditions; PM=parental inbred line mean; HM= F1 Hybrids mean; and %Loss=percentage loss.

GCA was significant ($P < 0.001$) for ASI, LFS, and CHL, significant ($p < 0.01$) for PLHT and significant ($p < 0.05$) for EL and 1000KW but no significant GCA effects were found for DS, EPP, EDIAM, and GY (Table 4.2). Significant ($p < 0.001$) SCA effects were found for DS, PLHT, CHL, and EDIAM, EL, 1000KW, and GY, significant ($p < 0.05$) effect was observed for ASI but no significant SCA effect was found for EPP and LFS. Reciprocal crosses were significant ($p < 0.001$) for CHL and significant ($p < 0.05$) for 1000KW (Table 4.2). The ratio of GCA/SCA mean squares showed that additive gene action was more important than non-additive gene action for ASI, LFS, CHL, and EPP while non-additive gene action was more important for DS, PLHT, EDIAM, EL, 1000KW and GY (Table 4.2).

4.2.1 Crop performance under low nitrogen environment (low-N)

In the low-N environment, significant ($p < 0.001$) differences were observed in the performances of the hybrids and the parental inbred lines used for the study (Table 4.4). Entry showed similar effects in terms of significant ($p < 0.001$) differences for all the traits observed for the study except for EPP (Table 4.4). Under low-N condition, the mean grain yield observed for the entry was 2.78 t/ha and 2.53 t/ha for the local checks respectively (Table 4.4). When the entry was partitioned into F1 hybrids and parental inbred lines, the mean grain yield observed for the hybrids was 3.17 t/ha while the inbred lines recorded an average of 1.06 t/ha (Table 4.3). Hybrids and inbred lines grain yield under low-N was about 75 % and 60 % of the grain yield under Opt-N condition respectively, indicating that the inbred lines were more sensitive to N deficit in the soil than the hybrids (Table 4.3). The best hybrid crosses under low-N condition were TZEEI-76×TZEEI-21, TZEEI-76×TZEEI-29 and TZEEI-21×TZEEI-76 with average grain yield of 4.82, 4.32 and 4.12 t/ha respectively and the best inbred lines were TZEEI-21 and TZEEI-76 with average grain yield of 1.53 and 1.52 t/ha respectively (Table 4.3).

Table 4.4 Mean squares for grain yield and other agronomic traits evaluated under low nitrogen environment

Source	DF	Mean Squares									
		DS	ASI	PLHT	LFS	CHL	EPP	EDIAM	EL	1000KW	GY
		Days	days	cm	0-1	cci		mm	cm	g	t/ha
Entry	24	19.08***	0.21***	864.57***	0.25***	170.38***	0.09	0.54***	10.28***	1333.49***	3.83***
GCA	4	8.85**	0.85***	666.39	1.13***	386.11***	0.23**	0.38**	4.02***	1998.11***	4.30***
SCA	10	38.59***	0.07	1523.52***	0.18*	182.56***	0.10	1.05***	21.32***	2006.02***	6.37***
REC	10	3.67*	0.09	284.89	0.03	71.92**	0.03	0.08	1.76**	2.18*	1.11
Error	48	1.64	0.05	273.11	0.05	20.03	0.05	0.08	0.64	181.58	0.70
CV (%)		2.26	9.05	15.99	11.60	13.33	19.33	8.65	6.81	9.30	29.97
GCA/SCA		0.23	12.14	0.44	6.28	2.12	2.30	0.36	0.19	1.00	0.68
Mean		56.67	5.53	103.31	3.48	33.58	1.21	3.27	11.74	144.89	2.78
Min		52.00	3.00	58.00	1.67	10.90	0.60	1.05	6.02	89.00	0.40
Max		64.00	10.00	144.20	7.22	51.50	2.00	4.02	14.72	211.00	6.53
CHM		57.83	5.17	104.05	4.17	33.23	1.18	3.25	12.29	144.67	2.53

*, **, *** indicates significance at 0.05, 0.01, and 0.001 probability levels respectively; CHM indicates checks mean.

DS= days to 50% silking; ASI= anthesis-silking interval; PLHT= plant height; LFS= leaf senescence; CHL= chlorophyll content of leaf; EPP=ears per plant; EDIAM= ear diameter; EL= ear length; 1000KW= weight of a thousand grain; and GY= grain yield

Analysis of variance for combining ability revealed both GCA and SCA contributed significantly to the variations in the performance of the entry for the selected traits indicating the presence of additive and non-additive gene actions. GCA had significant contributions ($p < 0.01, 0.001$) for all the traits with the exception of PLHT. Similar effects were found for SCA except for ASI and EPP (Table 4.4). Reciprocal crosses were significant ($p < 0.01$) for CHL and EL, and significant ($p < 0.05$) for DS and 1000KW. The ratio of GCA to SCA mean squares indicated that additive gene was more important for ASI, LFS, CHL, and EPP while non-additive gene was more important for DS, PLHT, EDIAM, EL, and GY (Table 4.4). Low-N influenced delay in silk emergence (57 days) when compared to Opt-N condition (55 days). An obvious difference was also observed in entry performance for secondary traits ASI, LFS, CHL and EPP as well as yield parameter 1000KW between low and opt-N environments.

4.2.2 Crop performance under well-watered environment (WW)

Entry showed significant ($p < 0.001$) differences for ASI, PLHT, CHL, DRW, and BYLD, significant ($p < 0.01$) differences for ARL and RSR, and significant ($p < 0.05$) differences for DS and LFS. This indicates significant variation between hybrids as well as the inbred lines for the traits under consideration (Table 4.5). The entry mean biomass yield under well-watered condition was 3.86 t/ha which is about 35 % greater than the biomass yield from local checks (Table 4.5). When the entry was partitioned into hybrid-inbred line components, the mean biomass yield observed was 4.15 t/ha and 2.69 t/ha for hybrids and inbred lines respectively (Table 4.3). The best hybrid cross under well-watered condition was TZEI-21×TZEI-76 with an average biomass yield of 4.73 t/ha.

Table 4.5 Mean squares for biomass yield and other agronomic traits evaluated under well-watered environment

Source	DF	Mean squares								
		DS	ASI	PLHT	LFS	CHL	ARL	RSR	DRW	BYLD
		days	days	Cm	0-1	cci	cm	0-1	g	t/ha
Entry (G)	24	22.06*	0.73***	1498***	0.21*	47.30***	160.99**	0.03**	19.91***	1.69***
GCA	4	54.34**	0.89*	687***	0.33*	71.54***	71.818	0.03*	9.71***	0.27**
SCA	10	21.27	1.04***	3226***	0.14	63.41***	316.11***	0.05***	42.76***	3.84***
REC	10	9.95	0.36	94.39	0.22*	21.49*	41.55	0.01	1.14	0.10
Error	96	11.86	0.26	126.22	0.10	10.71	70.90	0.01	0.89	0.06
CV (%)		5.74	23.64	10.97	14.65	17.48	16.54	19.82	10.14	6.42
GCA/SCA		2.56	0.86	0.21	2.36	1.13	0.28	0.60	0.23	0.08
Mean		60.05	4.58	102.42	4.36	18.73	50.92	0.52	9.30	3.86
Min		50.00	0.00	46.00	2.00	6.40	30.00	0.30	4.15	2.05
Max		67.00	9.00	140.00	8.00	32.40	85.00	0.98	14.60	5.10
CHM		59.13	4.63	93.88	6.25	16.03	41.50	0.45	5.16	2.54

*, **, *** indicates significance at 0.05, 0.01, and 0.001 probability levels respectively; CHM indicates checks mean.

DS= days to 50% silking; ASI= anthesis-silking interval; PLHT= plant height; LFS= leaf senescence; CHL= chlorophyll content of leaf; ARL= average root length; RSR= root to shoot ration; DRW= dry root weight; and BYLD= biomass yield.

Analysis of variance for combining ability showed the presence of significant additive and non-additive gene actions for the traits considered for the study (Table 4.5). GCA effects were significant ($p < 0.001$) for PLHT, CHL, and DRW, significant ($p < 0.01$) for BYLD and DS, significant ($p < 0.05$) for ASI, LFS, and RSR but no significant effect for ARL. SCA effects were significant ($p < 0.001$) for all the traits except for DS and LFS that showed no significant SCA effects while reciprocal crosses were significant ($p < 0.05$) for LFS and CHL. The ratio of GCA/SCA mean square revealed that additive gene was more important than non-additive gene for DS, LFS, and CHL while non-additive gene action was more important for the rest of the traits (Table 4.5).

4.2.3 Crop performance under drought stress environment (Dro-str)

In the drought stress environment, significant ($p < 0.001$) differences were observed in the performance of the entry for all the traits except for LFR and CHL where significant ($p < 0.01$) differences were observed (Table 4.6). Under drought stress condition, the mean biomass yield observed for the entries was 1.95 t/ha which is about 22 % higher than that observed for local check materials (Table 4.6). Upon partitioning the entry into hybrids and parental inbred lines, the mean biomass yield observed for the hybrids was 2.16 t/ha while the inbred lines recorded an average of 1.11 t/ha (Table 4.3). Both hybrids and inbred lines suffered a great loss in biomass yield under drought stress condition. Hybrids performance under drought was approximately 52 % of that under well-watered condition while inbred lines biomass yield was 41 % of the biomass yield under well-watered condition (Table 4.3).

The hybrid crosses with top performance under drought stress condition were TZEEI-29×TZEEI-21, TZEEI-38×TZEEI76, TZEEI-38×TZEEI-21, and TZEEI-21×TZEEI29 with average biomass yield of 2.90, 2.67, 2.58, 2.53 t/ha respectively.

Table 4.6 Mean squares for biomass yield and other agronomic traits evaluated under drought stress environment

Source	DF	Mean squares										
		DS days	ASI days	PLHT cm	LFR 1-5	LFS 0-1	CHL cci	TB 0-1	ARL cm	RSR 0-1	DRW g	BYLD t/ha
Entry	24	26.89***	1.04***	508.47***	0.06**	0.44***	24.09**	0.12***	779.40***	0.09***	79.10***	1.34***
GCA	4	53.56***	1.78***	218.89	0.04	0.23**	14.00	0.21***	324.95	0.06	34.71***	1.40***
SCA	10	28.84***	0.99***	898.76***	0.07*	0.64***	47.85***	0.15***	1632.52***	0.18***	170.45***	2.53***
REC	10	14.29	0.77**	234.01*	0.07*	0.32***	4.37	0.05*	108.05	0.02	5.50*	0.12*
Error	96	7.37	0.17	99.24	0.03	0.06	10.63	0.02	167.02	0.03	1.99	0.05
CV (%)		4.40	18.60	10.89	9.43	9.45	28.41	14.79	20.15	23.99	10.53	11.79
GCA/SCA		1.86	1.80	0.24	0.57	0.36	0.29	1.40	0.20	0.31	0.21	0.55
Mean		61.62	4.63	91.43	2.74	5.96	11.47	0.45	64.15	0.72	13.42	1.95
Min		55.00	0.00	60.00	1.00	1.00	2.40	0.00	21.00	0.25	6.03	0.72
Max		74.00	16.00	126.00	4.00	5.00	23.30	1.00	106.00	1.32	24.05	3.20
CHM		63.25	7.63	92.25	2.75	4.63	4.74	0.50	48.50	0.54	6.65	1.53

*, **, *** indicates significance at 0.05, 0.01, and 0.001 probability levels respectively; CHM indicates checks mean.

DS= days to 50% silking; ASI= anthesis-silking interval; PLHT= plant height; LFS= leaf senescence; CHL= chlorophyll content of leaf; ARL= average root length; RSR= root to shoot ration; DRW= dry root weight; and BYLD= biomass yield.

The best inbred line was TZEEI-76 with average biomass yield of 1.65 t/ha (Table 4.3). Out of the four crosses with the best performance, TZEEI-21 appeared in three of these crosses revealing possible good combining ability in hybrid combination (Table 4.3).

Analysis of variance for combining ability indicated the presence of significant additive and non-additive gene action influence on the traits measured. GCA and SCA contribute significantly to the variations in the performance of the hybrids as well as inbred parents. GCA effects were significant ($p < 0.001$) for DS, ASI, TB, DRW and BYLD, significant ($p < 0.01$) for LFS while non-significant GCA effects for PLHT, LFR, CHL, ARL, and RSR were observed (Table 4.6). Reciprocal crosses were significant ($p < 0.001$) for LFS, significant ($p < 0.01$) for ASI, significant ($p < 0.05$) for PLHT, LFR, TB, DRW, and BYLD but non-significant for CHL, ARL, and RSR.

The ratio of GCA to SCA mean squares indicated that non-additive gene was more important for most of the traits except for DS, ASI, and TB where additive gene predominated (Table 4.6). Drought stress like low-N also caused a slight delay in silk emergence (62 days) compared to well-watered condition (60 days). An obvious difference was also observed in ARL where drought stress increased root length by 21 % of what was observed under well-watered condition. Similar effect was found for RSR where drought stress resulted in increased RSR (0.72) compared to well-watered condition (0.51). Secondary traits ASI, LFS, and CHL also showed obvious differences in entry performance between drought stress and well-watered conditions. Slight increase in ASI was observed from 4.58 days under well-watered to 4.63 days under drought, LFS was increased by 27 % compared to well-watered environment and CHL was decreased by 39 % compared to well-watered environment.

4.2.4 Combined analysis across nitrogen (N) environments

Combined N analysis showed significant ($p < 0.001$) differences in environment (E) and entry (G) performance for grain yield but no significant G×E interaction was found indicating that entries had similar behaviour irrespective of the variability in N levels (Table 4.7). The mean entry grain yield observed across N environments was 3.22 t/ha which was slightly lesser than that observed for the local checks (3.26 t/ha). Later partitioning of the entry into hybrid-inbred patterns showed that the hybrids had grain yields ranging from 1.95 t/ha to 5.12 t/ha with a mean of 3.67 t/ha while the inbred lines range from 1.27 t/ha to 1.64 with a mean of 1.42 t/ha.

The result also showed that environment was significant ($p < 0.001$) for DS, ASI, CHL, and EPP, significant ($p < 0.01$) for EL but not significant for PLHT, LFS, EDIAM, and 1000KW. Entry was significant ($p < 0.001$) for almost all the traits except for EPP where significant ($p < 0.05$) difference was found (Table 4.7).

When the observed entry variability was partitioned into their combining ability, GCA had significant ($p < 0.001$) contributions to the entry mean squares for ASI, PLHT, LFS, CHL, 1000KW, significant ($p < 0.01$) contributions for EPP, EL, and GY, significant ($p < 0.05$) contribution for EDIAM but no significant contribution for DS. SCA effects were significant ($p < 0.001$) for DS, PLHT, CHL, EDIAM, EL, 1000KW, and GY, significant ($p < 0.01$) for ASI, and LFS, but not significant for EPP (Table 4.7). Reciprocal crosses was significant ($p < 0.001$) for CHL, significant ($p < 0.01$) for ASI and 1000KW and significant ($p < 0.05$) for DS, PLHT, and EL. Both GCA and SCA were found to highly contribute to the variations observed for PLHT, CHL, and 1000KW. The interaction of GCA×E was significant ($p < 0.05$) for EL, and 1000KW indicating that variation in the magnitude of GCA across environment exist for this traits.

Table 4.7 Mean squares for grain yield and other agronomic traits evaluated across low and optimal nitrogen environments

Source	DF	Mean squares									
		DS days	ASI days	PLHT cm	LFS 0-1	CHL cci	EPP	EDIAM mm	EL cm	1000KW g	GY t/ha
Env (E)	1	130.67***	3.87***	596.96	0.03	817.60***	10.57***	0.44	12.19**	319.74	28.77***
Entry (G)	24	31.24***	0.59***	1864***	0.36***	343.59***	0.14*	1.02***	18.53***	2206***	7.97***
GCA	4	4.40	2.11***	1692***	1.56***	773.91***	0.32**	0.30*	6.75**	2299***	6.03**
SCA	10	65.35***	0.34**	3152***	0.16**	375***	0.09	2.12***	38.86***	3551***	15.08***
REC	10	7.86*	0.24**	645*	0.08	13986***	0.12	0.21	2.92*	823**	1.63
G × E	24	2.90	0.11	178	0.06	19.44	0.10	0.11	1.33	264	0.81
GCA × E	4	6.20	0.08	134	0.12	8.99	0.08	0.28	4.40*	531*	2.28
SCA × E	10	1.53	0.12	149	0.06	10.19	0.13	0.12	1.28	242	0.43
REC × E	10	2.96	0.11	225	0.04	32.87	0.07	0.04	0.16	177	0.60
Error	96	3.31	0.11	281.43	0.06	19.25	0.08	0.12	1.38	214.77	1.30
CV (%)		3.26	14.59	15.93	12.71	12.22	19.04	10.72	10.26	10.01	35.41
GCA/SCA		0.07	6.21	0.54	9.75	2.06	3.56	0.14	0.17	0.65	0.33
Mean		55.73	4.89	105.30	5.16	35.91	1.48	3.21	11.46	146.35	3.22
Min		49.00	0.00	56.60	1.67	10.90	0.60	1.05	5.62	89.00	0.40
Max		64.00	10.00	161.30	7.22	56.50	2.90	4.10	15.28	211.00	7.45
CHM		56.33	5.08	109.47	3.93	37.48	1.35	3.39	12.20	155.25	3.26

*, **, *** indicates significance at 0.05, 0.01, and 0.001 probability levels respectively; CHM indicates checks mean. DS= days to 50% silking; ASI= anthesis-silking interval; PLHT= plant height; LFS= leaf senescence; CHL= chlorophyll content of leaf; EPP=ears per plant; EDIAM= ear diameter; EL= ear length; 1000KW= weight of a thousand grain; and GY= grain yield.

SCA \times E and REC \times E were not significant for any of the traits considered indicating the absence of variation in the magnitude of SCA and reciprocal crosses across N environments. Additive gene action was observed to be more important for ASI, LFS, CHL, and EPP while non-additive gene action dominated for the rest of the traits (Table 4.7).

4.2.5 Combined analysis across drought stress and well-watered environments

Combined analysis across moisture variation environments showed significant ($p < 0.001$) difference in environment (E), entry (G) and G \times E interaction for biomass yield indicating that entry behaviour varies with respect to variation in water conditions (Table 4.8). The mean entry biomass yield observed across drought stress and well-watered environments was 2.90 t/ha which is 24 % higher than the biomass yield observed for the local checks (2.20 t/ha). Later partitioning of the entry into hybrids and inbred parents, hybrids had biomass yield ranging from 2.64 t/ha (TZEEI-29 \times TZEEI-38) to 3.60 t/ha (TZEEI-21 \times TZEEI-76) with a mean of 3.18 t/ha while that of inbred lines ranged from 1.73 (TZEEI-29) t/ha to 1.99 (TZEEI-76) with a mean of 1.90 t/ha. The result also indicates highly significant difference of environment for all the traits except for ASI where none was found. Entry was significant ($p < 0.001$) for almost all the traits except for DS where significance ($p < 0.01$) was observed (Table 4.8).

Entry variability partitioned into combining ability showed significant ($p < 0.001$) GCA contributions to the entry mean squares for DS, PLHT, CHL, TB, DRW, and BYLD, significant ($p < 0.01$) contributions for LFS and RSR, significant ($p < 0.05$) contribution for ASI and no significant contribution for ARL. The contributions of SCA were significant ($p < 0.001$) for almost all the traits except for DS where significant ($p < 0.05$) contribution was observed (Table 4.8).

Table 4.8 Mean squares for biomass yield and agronomic traits evaluated across drought and well-watered environments

Source	DF	Mean squares									
		DS days	ASI days	PLHT cm	LFS	CHL cci	TB	ARL cm	RSR	DRW g	BYLD t/ha
Env (E)	1	121.29***	0.01	6035***	5.63***	2630***	2.27***	8752***	1.91***	847.48***	181.58***
Entry (G)	24	20.15**	0.92***	1490***	0.49***	52.20***	0.10***	684***	0.05***	70.39***	2.61***
GCA	4	51.00***	0.70*	715***	0.37**	66.20***	0.15***	238	0.08**	8.06***	1.19***
SCA	10	20.14*	1.67***	3136***	0.58***	81.79***	0.15***	1463***	0.07***	160.91***	5.73***
REC	10	7.82	0.25	154	0.45***	17.00	0.02	82.38	0.02	4.79*	0.06
G × E	24	28.81***	0.85***	517***	0.15**	19.20*	0.05***	257**	0.07***	28.62***	0.41***
GCA × E	4	56.90***	1.97***	191	0.18	19.34	0.11***	159.10	0.01	36.34***	0.48***
SCA × E	10	29.97**	0.36	989***	0.21**	29.47**	0.06***	485***	0.16***	52.30***	0.64***
REC × E	10	16.42	0.88**	175	0.08	8.86	0.03	67.23	0.01	1.84	0.16**
Error	96	9.61	0.21	113.00	0.07	10.67	0.01	118.96	0.02	1.44	0.06
CV (%)		5.09	19.25	10.96	11.99	19.63	14.09	18.95	22.93	10.57	8.23
GCA/SCA		2.53	0.42	0.23	0.64	0.81	1.00	0.17	1.14	0.05	0.21
Mean		60.84	4.60	96.92	2.58	15.09	0.25	57.54	0.62	11.36	2.90
Min		50.00	0.00	46.00	1.00	2.40	0.00	21.00	0.25	4.15	0.72
Max		74.00	16.00	140.00	5.00	32.40	1.00	106.00	1.32	24.05	5.10
CHM		61.19	6.13	93.06	3.88	10.38	0.25	45.00	0.50	5.91	2.20

*, **, *** indicates significance at 0.05, 0.01, and 0.001 probability levels respectively; CHM indicates checks mean.

DS= days to 50% silking; ASI= anthesis-silking interval; PLHT= plant height; LFS= leaf senescence; CHL= chlorophyll content of leaf; ARL= average root length; RSR= root to shoot ration; DRW= dry root weight; and BYLD= biomass yield.

It was evident from the high mean square values observed for the SCA that non-additive gene action was more important than additive gene action for many of the traits aside DS and RSR where GCA means square dominated. Reciprocal crosses were significant ($p < 0.001$) for LFS and significant ($p < 0.05$) for DRW. When $G \times E$ interaction was further partitioned into $GCA \times E$ and $SCA \times E$, the former had significant ($p < 0.001$) contributions for DS, ASI, TB, DRW, and BYLD but no significant contribution for other traits and the later had significant ($p < 0.001$) contributions for almost all the traits except for DS, LFS, and CHL where significant ($p < 0.01$) contributions were observed and ASI where no significant contribution was seen. $REC \times E$ only had significant ($p < 0.01$) contributions for ASI and BYLD (Table 4.8).

4.3 Estimates of General Combining Ability (GCA) for yield and other agronomic characters

The behaviour of the parental inbred lines varied from one another under the different environmental conditions used for the study. Only TZEEI-76 had a positive GCA estimates for yield in all the environments indicating the superiority of this line in terms of general combining ability. TZEEI-21 took after TZEEI-76 with respect to yield in all the environment considered except for optimal N condition where negative GCA estimate (-0.17) was observed (Fig. 4.1). In general, parental inbred lines that are good combiners under low-N and /or drought stress are TZEEI-76 and TZEEI-21. TZEEI-6 and TZEEI-29 had similarity in GCA estimates in all environments as both had consistent negative GCA estimates indicating that they are poor combiners. The general combining ability behaviour of TZEEI-38 was not consistent like TZEEI-76. TZEEI-38 only had positive GCA estimates under optimal N and across N environment with others being influenced by negative GCA estimate

(Fig. 4.1). In summary, the *per se* performance of the parental lines across similar environments and stress environments combined showed that TZEEI-76 and TZEEI-38 were the best combiners which can be used for further breeding programs.

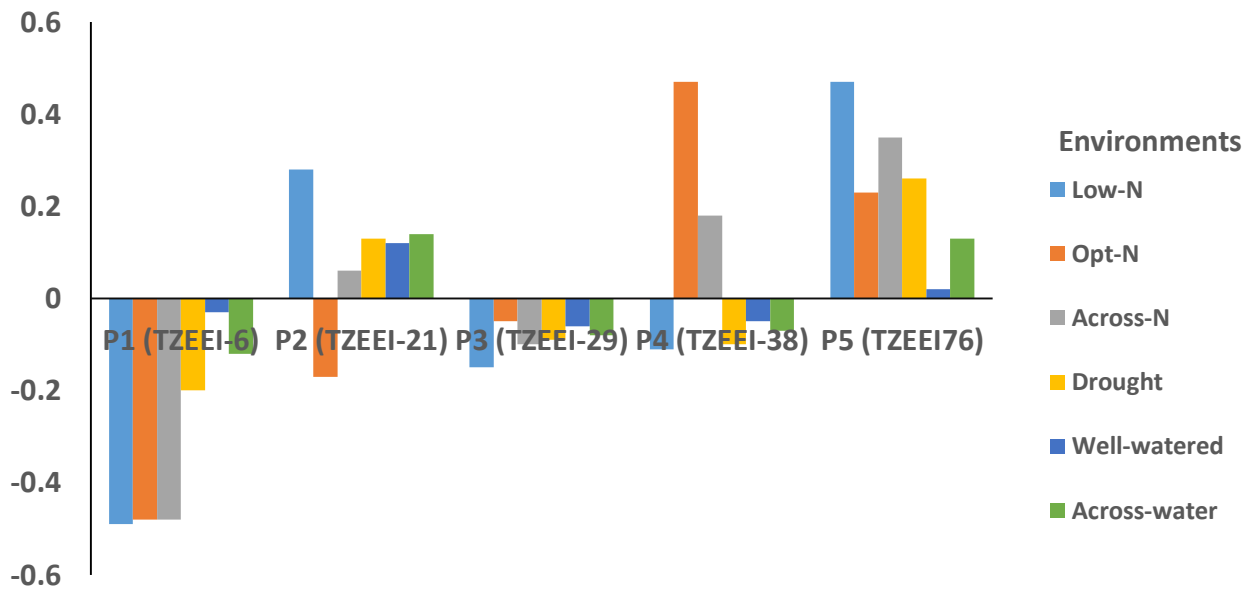


Fig. 4.1 GCA estimates of five parents for yield evaluated under stressed, non-stressed and across environments.

Secondary traits that overlap in the selection of genotypes for low-N stress and drought stress tolerance present in this study (ASI, LFS and CHL) also varied with respect to parental inbred lines GCA estimates. The best combiners for yield under the stress conditions (TZEEI-76 and TZEEI-21) had consistently negative GCA estimates for ASI. TZEEI-76 had significant ($p < 0.05$) estimate (-0.37) under low-N but not significant though negative under drought stress. TZEEI-21 on the other hand had significant ($p < 0.01$) estimate (-0.77) under drought stress but not significant though negative under low-N. TZEEI-38 and TZEEI-6 also showed significant ($p < 0.001$) GCA estimates.

While TZEEI-38 was the best performing (-0.80) parent for ASI, TZEEI-6 was the worst for the same trait (1.43) under low-N stress. Similar effect was observed under drought stress though TZEEI-38 was not the best parent there. Negative GCA estimate indicate quick synchronization of the male and female flowers for successful fertilization (Table 4.9).

With respect to LFS, only TZEEI-6 had significant ($p < 0.01$) positive estimate under both stress conditions indicating high senescence of leaves while other parents had negative GCA estimates with TZEEI-38 and TZEEI-76 being significant ($p < 0.01$) under low-N stress and TZEEI-21 under drought stress condition. Negative estimate for LFS is desirable as a measure of abiotic stress tolerance for a genotype.

CHL does not totally follow the pattern of ASI and LFS. Only TZEEI-21 and TZEEI-29 had significant ($p < 0.001$) positive GCA estimates under low-N with others being negative. Under drought, only TZEEI-76 had significant ($p < 0.05$) negative GCA estimate with others being positive though not significant (Table 4.9).

Parental inbred lines TZEEI-29, TZEEI-38 and TZEEI-76 showed a reduced plant height owing to their negative GCA estimate but not so for TZEEI-6 and TZEEI-21. In addition, TZEEI-6 showed consistent negative estimates for EPP, EDIAM, and 1000KW confirming its poor yielding ability. For ARL and DRW, TZEEI-76 had the highest significant ($p < 0.05$, 0.001) positive GCA estimates whose performance was followed by TZEEI-21 under drought stress. It is not surprising that P5 and P2 had the highest positive GCA estimates for grain yield as well as biomass yield, this may be due to their good performances in yield determining characters (Table 4.9).

Table 4.9. General combining ability effects of parental lines for agronomic traits under low-N (above) and drought stress (below) environments

Parent	DS	ASI	PLHT	LFS	CHL	EPP	EDIAM	EL	1000KW	GY	
TZEEI-6	0.4	1.43***	5.04	0.65**	-3.75***	-0.13**	-0.11*	0.41**	-6.99**	-0.49**	
TZEEI-21	-0.80***	-0.20	4.30	-0.24	3.18***	0.01	0.15**	0.35*	5.67*	0.28	
TZEEI-29	0.20	-0.07	-3.68	-0.39	4.41***	0.03	0.01	-0.40**	11.14***	-0.15	
TZEEI-38	0.50*	-0.80***	-0.05	-0.94***	-1.42	-0.03	-0.11*	-0.30*	-7.46**	-0.11	
TZEEI-76	-0.30	-0.37*	-5.61*	-0.96***	-2.41**	0.11**	0.07	-0.07	-2.36	0.47**	
SE(Gi)	0.21	0.17	2.69	0.15	0.73	0.03	0.04	0.13	2.20	0.13	
SE (Gi – Gj)	0.33	0.28	4.26	0.24	1.15	0.06	0.07	0.20	3.47	0.21	
Parent	DS	ASI	PLHT	LFR	LFS	CHL	TB	ARL	RSR	DRW	BYLD
TZEEI-6	-1.36**	1.56**	-1.08	-0.13	0.49**	0.22	-0.05	-3.50	-0.02	-1.49***	-0.20***
TZEEI-21	1.49**	-0.77**	3.57*	0.09	-0.46**	0.42	-0.19**	1.50	-0.02	0.55*	0.13***
TZEEI-29	-0.74	-0.33	-2.46	0.11	-0.11	0.33	-0.04	-0.25	0.01	-0.12	-0.09*
TZEEI-38	0.84*	-0.11	0.97	-0.10	0.19	0.06	0.14**	-1.65	-0.03	0.12	-0.10**
TZEEI-76	-0.23	-0.36	-1.01	0.04	-0.11	-1.03*	0.15**	3.90*	0.06*	0.95***	0.26***
SE(Gi)	0.44	0.31	1.63	0.09	0.08	0.47	0.04	1.83	0.02	0.20	0.03
SE (Gi – Gj)	0.70	0.48	2.57	0.15	0.13	0.73	0.06	2.88	0.04	0.32	0.05

*, **, *** indicates significance at 0.05, 0.01, and 0.001 probability levels respectively;

DS= days to 50% silking; ASI= anthesis-silking interval; PLHT= plant height; LFS= leaf senescence; CHL= chlorophyll content of leaf; EPP=ears per plant; EDIAM= ear diameter; EL= ear length; 1000KW= weight of a thousand grain; GY= grain yield; LFR= leaf rolling; TB= tassel blasting; ARL= average root length; RSR= root to shoot ratio; DRW= dry root weight; and BYLD= biomass yield.

4.3.1 Specific Combining Ability estimate for yield

The yield of the F1 hybrids varied in magnitude from one environment to another. However, the performance of the F1 hybrids for SCA estimates across all the environments combined (Low-N, Optimal N, Drought and Well-watered) showed that all hybrid crosses with positive SCA values had mean yield greater than 3 t/ha except for hybrid crosses TZEEI-76×TZEEI-21 and TZEEI-76×TZEEI-29 (Table 4.10). These hybrids were found to have the highest yield under low-N (4.82 t/ha and 4.32 t/ha respectively) yet had a negative SCA values (Table 4.10). Of the F1 hybrids identified with positive SCA estimates and grain yield greater than 3 t/ha across all environments, only four hybrids crosses; TZEEI-21×TZEEI-38, TZEEI-21×TZEEI-76, TZEEI-29×TZEEI-76 and TZEEI-38×TZEEI-76 had significant ($p < 0.05$, 0.01, 0.001) positive SCA estimates under the stressed environments (low-N and drought stresses). These four hybrids also have high yield under non-stressed environments (optimal N and well-watered) indicating they are good genotypes for both stress and non-stress conditions (Table 4.3). This might be influenced by the combining power of the parental inbred lines forming these hybrids.

Considering individual environment, hybrid crosses TZEEI-6×TZEEI-29, TZEEI-6×TZEEI-76, and TZEEI-21×TZEEI-29 were found to have significant ($p < 0.05$, 0.01, 0.001) positive SCA estimate under drought stress and well-watered conditions, non-significant but positive under low-N stress and optimal N conditions with the exception of TZEEI-21×TZEEI-29 that was negative under this condition (Table 4.10). Hybrid crosses TZEEI-29×TZEEI-38 and TZEEI-29×TZEEI-6 had positive significant ($p < 0.05$) SCA estimate under low-N and opt-N conditions but negative under drought stress condition indicating they may only do well under low-N stress condition and not drought stress.

Table 4.10 Specific combining ability estimates of F1 hybrids grain and biomass yield evaluated in stressed, non-stressed and across similar environments

Crosses	Low-N	Opt-N	Across-N	Dro-str	Well-W	Across-W	Across-env
TZEEI-6×TZEEI-21	0.01	-0.05	-0.02	-0.05	-0.04	-0.05	-0.04
TZEEI-6×TZEEI-29	0.22	0.22	0.22	0.26***	0.43***	0.34	0.29
TZEEI-6×TZEEI-38	0.54	0.64	0.59*	0.15	0.25**	0.20	0.37*
TZEEI-6×TZEEI-76	1.19	0.85	1.02	0.96***	0.82***	0.89*	0.95**
TZEEI-21×TZEEI-29	-0.21	-0.05	-0.12	0.72***	0.26**	0.49*	0.23
TZEEI-21×TZEEI-38	0.85**	0.38	0.61*	0.54***	0.35***	0.44*	0.52**
TZEEI-21×TZEEI-76	2.53***	3.31***	2.92***	1.19***	1.85***	1.52***	2.12***
TZEEI-29×TZEEI-38	0.63*	1.13*	0.88**	-0.35***	0.16	-0.10	0.32
TZEEI-29×TZEEI-76	2.50***	2.56**	2.53***	0.89***	1.73***	1.31**	1.83***
TZEEI-38×TZEEI-76	2.10**	2.99**	2.55***	1.17***	1.45***	1.31**	1.84***
TZEEI-21×TZEEI-6	0.47	0.14	0.30	-0.15	0.14	-0.01	0.13
TZEEI-29×TZEEI-6	0.91*	0.9	0.90*	-0.03	0.09	0.03	0.41
TZEEI-29×TZEEI-21	-0.16	0.01	-0.06	-0.18*	0.18	0.00	-0.03
TZEEI-38×TZEEI-6	0.28	-0.08	0.10	0.04	0.07	0.05	0.07
TZEEI-38×TZEEI-21	-0.57	0.59	0.01	-0.05	0.15	0.05	0.03
TZEEI-38×TZEEI-29	0.14	0.68	0.41	0.07	-0.10	-0.02	0.17
TZEEI-76×TZEEI-6	0.36	0.23	0.29	0.14	-0.03	0.05	0.16
TZEEI-76×TZEEI-21	-0.35	-0.2	-0.27	0.15	0.09	0.12	-0.05
TZEEI-76×TZEEI-29	-0.29	-0.19	-0.24	0.14	-0.11	0.01	-0.10
TZEEI-76×TZEEI-38	0.12	0.33	0.22	0.14	0.10	0.12	0.16
SE(Sij)	0.28	0.45	0.27	0.07	0.07	0.04	0.20
SE(Sij-Skl)	0.37	0.60	0.36	0.09	0.10	0.07	1.22
SE (Rij)	0.34	0.55	0.32	0.08	0.09	0.06	0.15

*, **, *** indicates significance at 0.05, 0.01, and 0.001 probability levels respectively

Low-N= Low-Nitrogen condition; Opt-N= optimal nitrogen condition; Across-N= across nitrogen combined; Dro-str= drought stress condition; Well-W= well-watered condition; Across-W= across water level combined and Across-env= across all environments combined.

Hybrid crosses TZEEI-6×TZEEI-38, TZEEI-38×TZEEI-29, TZEEI-76×TZEEI-6 and TZEEI-76×TZEEI-38 also had positive but non-significant SCA estimate for both stress conditions indicating potential to do well under both stress conditions. Hybrid crosses TZEEI-29×TZEEI-21 and TZEEI-38×TZEEI-21 had negative SCA estimates under low-N and drought stress indicating a probable potential of poor performance but these genotypes showed good performance under drought stress condition with yield of 2.90 t/ha and 2.58 t/ha respectively.

4.4 Variance components of GCA and SCA for individual environment and across similar environments

To a large extent, non-additive variance was more important than additive variance for the inheritance of almost all the selected traits under both stress environments (Fig. 4.2) and across similar environments as indicated by their high contributions to the entry sum of square (Appendix 13). ASI, LFS and EPP clearly show the importance of additive variance under low-N stress but suppressed by non-additive variance under drought stress. ASI and LFS also show a dominance of additive variance under opt-N environment. GCA was found to contribute a sizable proportion to CHL, 1000KW and GY under low-N and DS, ASI, TB, LFR and BYLD under drought stress condition. Grain yield under low-N stress as well as biomass yield under drought stress were both dominated by non-additive variance with GCA contributing less than 30 % to the total variation. This explains the relative importance of SCA in determining yield of hybrid genotypes in maize. Under Opt-N environment, GCA had immense contribution ($\geq 40\%$) for ASI, LFS and CHL. Under well-watered environment, only LFS had high GCA contribution among the secondary traits (Fig. 4.3). Narrow sense heritability (h^2) estimate across stressed conditions was moderately higher for ASI and LFS than observed for CHL suggesting the importance of ASI and LFS in enhancing

selection than CHL under stressed conditions (Table 4.11). In general, h^2 of the selected secondary traits observed across stressed environments were higher than those across optimal environments.

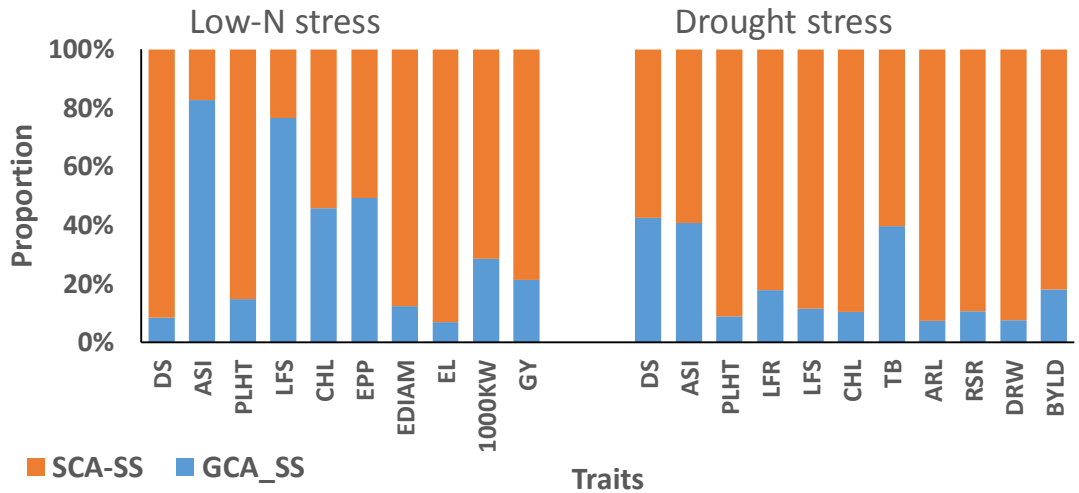


Fig. 4.2 Components of variance for GCA and SCA under low-N and drought stress environments

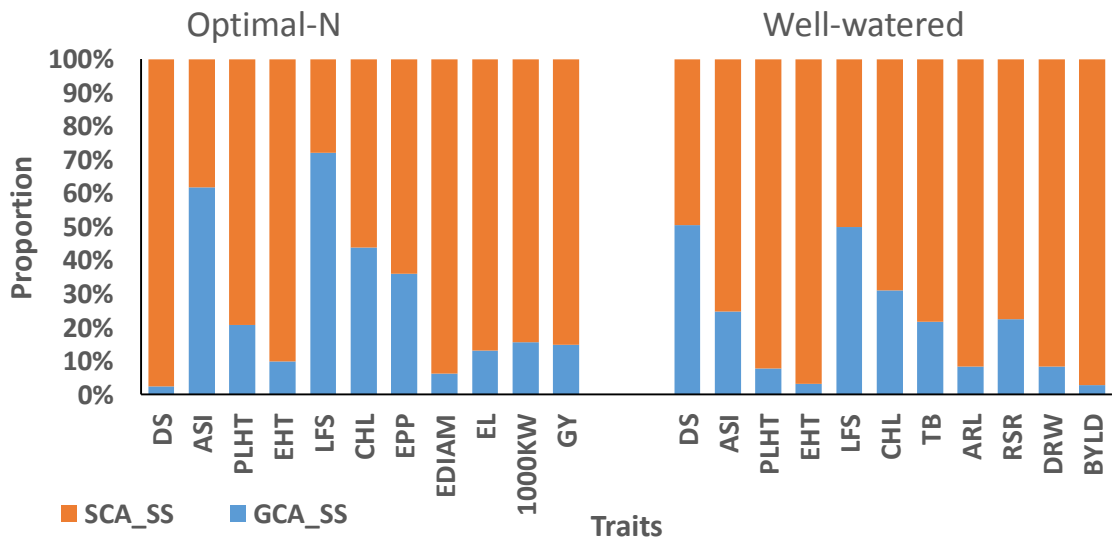


Fig.4.3 Components of variance for GCA and SCA under optimal N and well-watered environments

Table 4.11 Variance components for general and specific combining ability, narrow and broad sense heritability and Baker's ratio for anthesis-silking interval, leaf senescence, leaf chlorophyll content and yield across stressed, optimal and similar environments

Env	Traits	Genetic parameter							
		σ^2_{gca}	σ^2_{sca}	σ^2_e	σ^2_A	σ^2_D	h^2	H^2	BR
Across str									
	ASI	3.43	1.71	2.55	6.86	1.71	51.66	64.53	0.80
	LFS	1.64	1.12	1.21	3.28	1.12	48.19	64.70	0.74
	CHL	1.27	0.62	14.38	2.54	0.62	35.58	44.29	0.80
	YLD	0.94	3.40	2.30	1.87	3.40	22.59	63.58	0.36
Across opt									
	ASI	1.09	1.67	3.51	2.17	1.67	34.23	60.51	0.57
	LFS	1.20	0.85	1.61	2.40	0.85	46.68	63.19	0.74
	CHL	1.33	1.85	13.82	2.67	1.85	29.38	49.74	0.59
	YLD	0.21	8.32	0.79	0.42	8.32	3.18	66.00	0.05
Across-N									
	ASI	7.25	2.67	1.75	14.50	2.67	55.67	65.92	0.84
	LFS	5.88	3.02	1.01	11.77	3.02	52.64	66.16	0.80
	CHL	2.04	36.80	19.25	4.08	36.80	6.33	63.35	0.10
	EPP	2.65	0.69	0.08	5.29	0.69	58.87	66.57	0.88
	YLD	0.37	35.07	1.30	0.74	35.07	1.38	66.40	0.02
Across-W									
	ASI	0.38	3.64	3.88	0.75	3.64	10.40	60.71	0.17
	LFS	0.79	2.34	0.41	1.59	2.34	26.66	65.90	0.40
	CHL	0.95	2.78	10.67	1.89	2.78	21.55	53.16	0.41
	YLD	0.29	8.95	0.05	0.59	8.95	4.12	66.63	0.06

Across str= across all stressed environments; Across opt= across all optimal environments; Across-N= across nitrogen levels; Across-W= across water levels; σ^2_{gca} = variance of general combining ability; σ^2_{sca} = variance of specific combining ability; σ^2_A = additive variance; σ^2_D = dominance variance; σ^2_e = error variance; h^2 = narrow sense heritability; H^2 = broad sense heritability; BR=baker's ratio; ASI= anthesis-silking interval; LFS= leaf senescence; CHL= leaf chlorophyll content; EPP= ears per plant; and YLD= grain and biomass yield combined

Considering similar environments, moderately high h^2 estimates were observed across N environments for ASI, LFS, and EPP. Only CHL and YLD had very low h^2 estimates (Table 4.11). This does not hold true for these traits across moisture environments as they all show low h^2 estimates (< 30%).

The low h^2 estimates of these traits were due to their poor GCA/SCA ratio which in turn results in very low σ^2_{gca} and σ^2_A . Baker's ratio indicates that ASI, LFS and CHL are controlled by additive gene action under all stressed conditions and all non-stressed conditions. Across N environments, ASI, LFS and EPP were controlled by additive gene action owing to their baker's ratio but CHL was influenced by non-additive gene action. Estimates across moisture environments showed the influence of non-additive gene action for all the selected traits. Narrow sense heritability of YLD was low in all the environment combinations used for the study which is an indication of YLD being a polygenic character.

4.5 Heterosis estimate for grain and biomass yield under stressed and non-stressed environments

Heterosis estimated for grain yield under nitrogen variation experiment and biomass yield under moisture variation experiment revealed varying magnitude of hybrid vigor with respect to mid and better parent heterosis. On the average, heterosis estimates were higher under stressed conditions (low-N and drought) than non-stressed conditions (optimal N and well-watered) indicating the concept of better adaptation (increased vigor) of the F1 hybrids to stressed conditions than inbred lines (Table 4.12). Heterosis estimate ranged from approximately 21 % to 368 % under low-N stress. Hybrids with significantly positive SCA effects all had high heterosis estimates with significance ($p < 0.01, 0.001$) under both stress conditions except for TZEEI-29×TZEEI-6 under low-N (Table 4.12). The result further revealed that not all hybrid crosses with positive SCA had high heterosis estimates. Hybrid crosses with negative SCA estimates were also found to have high heterosis value with significant effect (Table 4.10 and Table 4.12). Also, hybrid crosses involving parental lines with negative GCA were also found to have high heterosis (Table 4.9 and Table 4.12). The highest heterosis under low-N was 368 % (MPH) 263 % (BPH) for hybrid cross

Table 4.12 Mean yield and heterosis of F1 hybrid crosses evaluated under stressed and non-stressed conditions

Parent	Low-N			Opt-N			Dro-str			Well-W		
	Mean t/ha	MPH %	BPH %	Mean t/ha	MPH %	BPH %	Mean t/ha	MPH %	BPH %	Mean t/ha	MPH %	BPH %
TZEEI-6×TZEEI-21	3.04	135.66**	73.71*	3.11	82.94	66.31	1.68	77.78**	68.00*	4.06	35.33*	32.25*
TZEEI-6×TZEEI-29	3.27	275.86***	259.34***	4.24	142.98*	126.74*	1.89	100.00***	89.00**	4.28	55.07***	39.41*
TZEEI-6×TZEEI-38	3.00	351.13***	261.45***	4.22	116.41*	107.88*	1.84	94.71***	84.00*	4.10	43.36**	33.55*
TZEEI-6×TZEEI-76	3.33	172.95***	106.83**	3.65	115.34*	95.19	2.44	92.13***	47.88*	3.89	44.07**	26.71
TZEEI-21×TZEEI-29	2.55	91.73*	45.71	3.40	115.87*	109.88	2.53	153.00***	153.00***	4.37	62.45***	49.15**
TZEEI-21×TZEEI-38	3.22	186.22***	84.00*	4.93	176.97***	142.86*	2.48	148.00***	148.00***	4.44	59.14***	51.54**
TZEEI-21×TZEEI-76	4.12	145.24***	135.43***	5.04	230.49***	229.41**	2.48	87.17***	50.30*	4.73	79.85***	61.43***
TZEEI-29×TZEEI-38	3.30	368.09***	262.64***	5.89	222.74***	190.15***	1.49	49.00	49.00	3.80	49.02**	43.40*
TZEEI-29×TZEEI-76	3.73	196.03***	131.68***	4.27	171.97**	163.58*	2.38	79.62***	44.24*	4.13	72.80***	68.57***
TZEEI-38×TZEEI-76	3.30	212.80***	104.97*	5.10	187.32***	151.23**	2.67	101.51***	61.82**	4.25	70.68***	60.38***
TZEEI-21×TZEEI-6	2.11	63.57	20.57	2.82	65.88	50.80	1.99	110.58***	99.00**	3.78	26.00	23.13
TZEEI-29×TZEEI-6	1.45	66.67	59.34	2.45	40.40	31.02	1.95	106.35***	95.00**	4.10	48.55**	33.55*
TZEEI-29×TZEEI-21	2.87	115.79**	64.00	3.38	114.60*	108.64	2.90	190.00***	190.00***	4.01	49.07**	36.86*
TZEEI-38×TZEEI-6	2.45	268.42**	195.18*	4.37	124.10*	115.27*	1.77	87.30**	77.00*	3.95	38.11*	28.66
TZEEI-38×TZEEI-21	3.75	231.86***	114.29***	4.37	145.51**	115.27*	2.58	158.00***	158.00***	4.14	48.39**	41.30*
TZEEI-38×TZEEI-29	3.02	328.37***	231.87***	4.53	148.22**	123.15*	1.35	35.00	35.00	4.00	56.86***	50.94**
TZEEI-76×TZEEI-6	2.62	114.75*	62.73	3.19	88.20	70.59	2.17	70.87***	31.52	3.95	46.30**	28.66
TZEEI-76×TZEEI-21	4.82	186.90***	175.43***	5.42	255.41***	254.25***	2.17	63.77**	31.52	4.56	73.38***	55.63***
TZEEI-76×TZEEI-29	4.32	242.86***	168.32***	4.64	195.54**	186.42**	2.10	58.49**	27.27	4.36	82.43***	77.96***
TZEEI-76×TZEEI-38	3.06	190.05***	90.06*	4.45	150.70**	119.21*	2.39	80.38***	44.85*	4.06	63.05***	53.21**
Mean	3.17	189.15	132.38	4.17	144.58	127.89	2.16	97.18	79.22	4.15	55.20	44.81

*, **, *** indicates significance at 0.05, 0.01, and 0.001 probability levels respectively

Low-N= Low-Nitrogen condition; Dro-str= drought stress condition; Opt-N= optimal nitrogen condition; Well-W= wall-watered condition; MPH= mid-parent heterosis; and BPH=better parent heterosis.

TZEEI-29×TZEEI-38 and the lowest being 64 % (MPH) 21 % (BPH) for hybrid cross TZEEI-21×TZEEI-6. Under drought stress condition, the highest estimate was 190 % (MPH and BPH) for hybrid cross TZEEI-29×TZEEI-21 and the lowest being 35 % (MPH) 27 % (BPH) for the crosses TZEEI-38×TZEEI-29 and TZEEI-76×TZEEI-29 respectively.

4.5.1 Effect of drought stress and low-N on biomass and grain yield

As supposed, drought and low-N stress resulted in reduced yield but the magnitude of yield reduction varied considerably among the entries due to the variability in their level of tolerance to these stresses. On the average, drought stress caused more yield reduction (49 %) than compared to low-N stress (28 %) (Table 4.3). The highest yield loss among the hybrids genotype was found for TZEEI-38×TZEEI-29 (66 %) located under drought stress condition while the lowest was for TZEEI-6×TZEEI-76 (9 %) located under low-N stress condition. Considering each stress effect on the yield outcome, the loss due to drought stress alone ranged from 28 % to 66 % with an average of approximately 48 %. Loss due to low-N ranged from 9 % to 44 % with an average of approximately 24 % which is half of what was observed for drought stress effect. Nevertheless that biomass yield was considered for the drought stress study due to lack of ear bearing kernels from plants, the magnitude of loss if grain yield was considered would still correlates to the trend observed (Table 4.3).

Yield loss measured for local checks range from 29 % to 48 % with an average of 39 % under drought and 34 % to 40% with an average of 37 % under low-N confirming the higher influence of drought stress on yield than low-N. The best five hybrids under drought stress have low to moderate yield loss as compared to the worst five hybrids. Similar trend was observed under low-N condition. The most interesting aspect is that not all the hybrids that fall in the first and last five category under drought were duly represented in the same manner under low-N indicating an overlap in hybrids responses to the two stress conditions. Such

overlap is that of hybrid cross TZEEI-21×TZEEI-29 which combine moderately high yield under drought stress with low yield under low-N stress condition (Table 4.3). Ranking the hybrids and local checks performance based on the combined stress effects, only ten hybrids crosses; TZEEI-29×TZEEI-21, TZEEI-6×TZEEI-76, TZEEI-38×TZEEI-21, TZEEI-29×TZEEI-76, TZEEI-76×TZEEI-29, TZEEI-76×TZEEI-21, TZEEI-21×TZEEI-76, TZEEI-76×TZEEI-6, TZEEI-6×TZEEI-21 and TZEEI-21×TZEEI-29 had lower yield loss than the local checks (Table 4.3). Though the local checks had lower yield, losses due to N deficit and drought stresses are still reduced compared to some hybrids.

Combine effect of drought and low-N stresses showed different magnitude of the hybrids yield loss with an average of approximately 39 %. The magnitude of the combined stress effect ranges from 22 % to 51 %. It was surprising to see that the least and the best performed hybrid analyzed across stress conditions; TZEEI-29×TZEEI-6 and TZEEI-76×TZEEI-21 respectively were not the hybrids with the highest and lowest yield loss (Table 4.3)

4.5.2 Identifying the level of tolerance of hybrids to drought and low-N stresses

In a means to identify the level of tolerance of the F1 hybrids and local checks to drought and/or low-N stress, Genotypes performance was plotted against the two stress environmental conditions in order to identify hybrids with only drought tolerance potential, hybrids with low-N tolerance potential only and hybrids that combine tolerance to both stress conditions. In Fig. 4.4, genotypes lying to the right of the vertical axis performed above average while those on the left had low performance under drought stress condition. Genotypes lying above the horizontal axis had above average performance while those below had below average performance under low-N condition. The diagonal line is the equality measure of tolerance to both stresses and any genotype found on this line performed equally

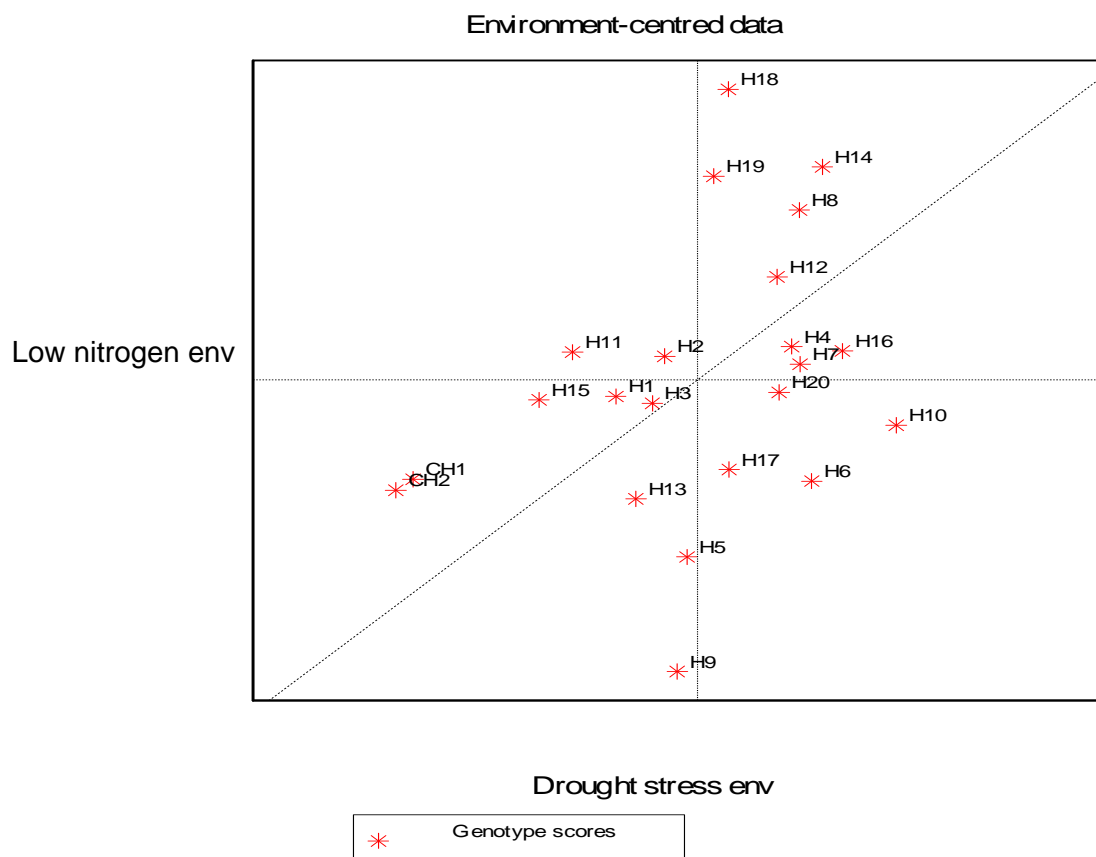


Fig. 4.4 Centered scattered plot of F1 hybrids as influenced by drought and low nitrogen stresses

H1=TZEEI-6×TZEEI-21	H7=TZEEI-21×TZEEI-38	H13=TZEEI-38×TZEEI-6	H19=TZEEI-76×TZEEI-29
H2=TZEEI-6×TZEEI-29	H8=TZEEI-21×TZEEI-76	H14=TZEEI-38×TZEEI-21	H20=TZEEI-76×TZEEI-38
H3=TZEEI-6×TZEEI-38	H9=TZEEI-29×TZEEI-6	H15=TZEEI-38×TZEEI-29	CH1=CHECK-1
H4=TZEEI-6×TZEEI-76	H10=TZEEI-29×TZEEI-21	H16=TZEEI-38×TZEEI-76	CH2=CHECK-2
H5=TZEEI-21×TZEEI-6	H11=TZEEI-29×TZEEI-38	H17=TZEEI-76×TZEEI-6	
H6=TZEEI-21×TZEEI-29	H12=TZEEI-29×TZEEI-76	H18=TZEEI-76×TZEEI-21	

under the two stress conditions indicating tolerance to both stress. In this study, none of the F1 hybrids was exactly on the diagonal line of equality above both drought and low-N axis but the hybrids closest to this line are TZEEI-6×TZEEI-76, TZEEI-21×TZEEI-38, TZEEI-21×TZEEI-76, TZEEI-29×TZEEI-76 and TZEEI-38×TZEEI-76 indicating they possess better adaptation to both stress conditions than other genotypes. Considering drought stress tolerance, hybrids TZEEI-29×TZEEI-21, TZEEI-38×TZEEI-21, TZEEI-21×TZEEI-29,

TZEEI-76×TZEEI-38, TZEEI-76×TZEEI-6, and TZEEI-76×TZEEI-21 had above average performance. Hybrids TZEEI-76×TZEEI-21 and TZEEI-38×TZEEI-21 also show some degree of tolerance to low-N but not as high as those close to the diagonal. Considering low-N tolerance, hybrids TZEEI-76×TZEEI-21 took the lead with others such as TZEEI-38×TZEEI-21, TZEEI-76×TZEEI-29, TZEEI-29×TZEEI-38 and TZEEI-6×TZEEI-29 tailed behind. The local check materials that was used for the study had performances indicating the lack of tolerance to both stress conditions owing to their performance which was below average according to the results.

4.6 Correlation among yield and selected contributing characters

Pearson correlation computed using the observed yield and selected agronomic characters that contributed to yield (YLD) show that, under low-N stress environment, yield had significant ($p < 0.001$) positive correlation with PLHT (0.54) and leaf CHL (0.66). However, YLD showed a significant ($p < 0.001$) negative correlation with LFS (-0.78) and DS (-0.42). Significant ($p < 0.05$) negative correlation was observed with ASI and EPP showing non-significant positive correlation (Table 4.13).

Under drought stress environment, YLD was observed to have varying magnitudes of correlation ranging from $p < 0.05$ to $p < 0.001$ levels of significance with other traits. YLD had significant ($p < 0.05$) negative correlation with ASI (-0.20), significant ($p < 0.01$) negative correlation with DS (-0.27) and significant ($p < 0.001$) negative correlation with LFS (-0.48). The only significant positive correlation observed with yield here occurred with ARL (0.62) at $p < 0.001$ (Table 4.13).

Table 4.13 Correlation matrix of yield and selected agronomic characters evaluated in stress and non-stress environments

Environment	Traits	DS	ASI	PLHT	LFS	CHL	EPP	LFR	ARL	TB
Low-N stress	ASI	0.19								
	PLHT	-0.64***	0.09							
	LFS	0.32**	0.26*	-0.03						
	CHL	-0.59***	-0.33**	0.37***	-0.47***					
	EPP	0.14	-0.15	-0.26*	-0.26*	0.05				
	YLD	-0.78***	-0.25*	0.54***	-0.42***	0.66***	0.20			
Drought stress	ASI	0.24*								
	PLHT	-0.08	-0.12							
	LFS	0.01	0.32***	-0.29**						
	CHL	-0.04	-0.02	0.21*	-0.45***					
	LFR	0.07	-0.09	-0.18	0.07	-0.03				
	ARL	0.22*	-0.12	0.19	-0.30**	-0.05	-	-0.03		
	TB	-0.12	-0.03	-0.40***	0.33***	-0.28**	-	0.13	-0.21*	
YLD	-0.27**	-0.20*	0.14	-0.48***	0.16		0.02	0.62***	-0.16	
Across stress	ASI	0.09								
	PLHT	-0.43***	0.01							
	LFS	0.42***	0.17*	-0.31***						
	CHL	-0.65***	0.05	0.42***	-0.67***					
	YLD	-0.43***	-0.11	0.49***	-0.54***	0.60***				
Across non-stress	ASI	0.33***								
	PLHT	-0.39***	0.12							
	LFS	0.24**	0.15*	-0.17*						
	CHL	-0.64***	-0.09	0.33***	-0.52***					
	YLD	-0.30***	-0.09	0.61***	-0.27***	0.28***				

*, **, *** indicates significance at 0.05, 0.01, and 0.001 probability levels respectively; DS= days to 50% silking; ASI=anthesis-silking interval; PLHT= plant height; LFS= leaf senescence; CHL= leaf chlorophyll content; EPP= ears per plant; LFR= leaf rolling; ARL= average root length; TB=tassel blasting; YLD= yield

When drought and low-N stresses were combined, DS and LFS maintained a highly significant negative correlation with yield with correlation coefficient of -0.43 and -0.54 respectively while leaving ASI behind with non-significant negative correlation. PLHT and CHL also maintained highly significant positive correlation with YLD with coefficient of 0.49 and 0.60 (Table 4.13). The trend of relationship between yield and contributing characters observed for across non-stress environment was similar to that of across stress environment. CHL and PLHT had significant ($p < 0.001$) positive correlation with yield while DS and LFS had significant ($p < 0.001$) negative correlation with yield. ASI was also found to negatively correlated but without any significance.

CHAPTER FIVE

5.0 DISCUSSION

5.1 Yield and agronomic performance across all environments and similar environments

When analyzed across all environments, the significance of the entry, environment and the interaction of the entry by environment indicated that environmental and genotypic variations have resulted in the observable differences in the yield and other traits performance of the genotypes. This is so because crop phenotypic expression is dependent on the genotype involved and the environment in which such genotype is grown (Badu-Apraku *et al.*, 2011b). This indicates the uniqueness of the environments where the genotypes were evaluated and the presence of sufficient genetic variability among the genotypes which will allow selection for improvement to be possible for most of the traits considered for the study. The significant interaction of entry by environment is a measure of the similarity or dissimilarity in the entry performance across the environments. This explains how the genetic system controls the physiological processes that confer adaptation to the environmental differences (Cooper and Byth, 1996).

Considering N variation environments, there was a high degree of similarity in the genes that influence the genetic variation for grain yield in the two environments which was observed as the lack of genotype by environment interaction. This means that alleles controlling grain yield under low-N are similar to those under opt-N. This is in contradiction to the observations made by Worku *et al.* (2008), and Cooper and Byth (1996) that different genetic variability in grain yield is expressed under high and low-N conditions. The observed entry performance across these environments was slightly lower than that of the local checks

probably due to the diallel mating type used for the study. The complete model of Griffing (1956) diallel methodology permits the inclusion of parental inbreds which by convention will have a drastically reduced yield performance as a result of inbreeding depression when compared to their F1 progeny. Fehr (1993) explained that continuous segregation of heterozygous loci by mating of genetically similar genotypes will result in fixation of the alleles in the population. This will permit deleterious alleles to show their phenotypic influence which is termed inbreeding depression. This depression which is common in parental inbreds could have accounted for the reduction in the average performance of the entry as was observed.

Across N variation environments, GCA and SCA were both found to be important in controlling grain yield and other agronomic characters. Kamara (2015) reported that when both GCA and SCA are found to be significant for a quantitative character, it is an indication of Additive-dominance gene action. In order to clear the misconception as to which of the gene action predominates, GCA/SCA ratio was used to identify the predominance of non-additive gene action for grain yield. This result is in contradiction with the findings of Worku *et al.* (2008) who reported the predominance of additive gene action for grain yield across low and high N environments but in agreement with that of Katta *et al.* (2013) and Kamara (2015).

In the moisture variation environments, entry performance varied with environmental differences as observed by the significant genotype by environment interaction for biomass yield (BYLD) and other agronomic characters. The magnitude of additive and dominance gene actions also varied with environmental variations. Both additive and non-additive genetic control were observed for BYLD though there was preponderance of non-additive gene action. The agronomic characters considered showed predominance of non-additive

genetic control except for DS. This is in contradiction to the findings of Pavan *et al.* (2011) who reported a preponderance of non-additive genetic effect for days to silking in maize. The results also corroborate the findings of the same authors for PLHT and EHT. The average entry performance across moisture environment was 32 % greater than the average performance of the local checks, despite the inclusion of parental inbred lines.

5.2 Yield and agronomic performance under individual environment

5.2.1 Low-N stress and optimal N environments

The intensity of stress as conditioned by the N stress level chosen for this study resulted in yield loss ranging from 2.25 - 75 % with an average of 28 % which is lesser/greater than the yield reduction reported in some other studies. In reality, yield loss attributable to N deficit varies with variations in soil N deficiency as many studies have reported different levels of yield loss owing to the different levels of soil N deficit. To mention a few, Badu-Apraku *et al.* (2011b) observed that the yield of the best performing inbred under low-N (30 kg/ha) was just 5 % less than that of the best performing inbred under high-N (90 kg/ha) though the least performing under low-N was 21 % of the yield observed for the least performing under high-N condition. Meseke *et al.* (2013) reported a yield loss of 56 % following application of 20 kg/ha of N fertilizer in a low-N study with twenty-four maize inbred lines crossed in a design II mating system; Worku *et al.* (2008) also reported reduction of 64 % in grain yield under low-N compared to high-N condition in a study involving 635 lines (S₂-S₇) following 0 kg/ha in a field with highly depleted soil nitrogen.

Nitrogen deficit has been reported to affect inbred lines than hybrids due to the presence of very low/zero heterozygous loci (Badu-Apraku *et al.*, 2011c) which explains the higher

magnitude of mean yield loss (39 %) observed for the inbred lines compared to that of the F1 hybrids (24 %).

The agronomic traits contributing to grain yield vary in performances under low-N and opt-N environments. Low-N stress resulted in delayed DS which in turn resulted in the extension of the ASI observed in this study. Banziger *et al.* (2000) explained the delay in silk emergence as a cause of ineffective pollination and fertilization, reduced kernel weight as observed for this study, as well as increased kernel and ear abortions which might have resulted in the reduced number of ear per plant observed for the study.

The increased leaf senescence observed under low-N is an indication of high rate of remobilization of N from the lower/older leaves to the upper leaves for photosynthesis and from the stem to the growing sinks. According to Banziger *et al.* (2000), low-N stress induces premature leaf yellowing which progresses to leaf death. This increases the rate of leaf senescence through N remobilization and reduces the rate of photoassimilate production and partitioning into growing sinks which in turn affect kernel weight.

Nitrogen is an important part of chlorophyll making it a key determinant in the photosynthetic life of maize plant and consequently a factor for high yield performance. The reduced leaf chlorophyll content conditioned by low-N stress used in this study showed the high sensitivity of maize plant to soil N as reported by Banziger and Diallo (2004). Pale green coloration of leaves is a well-known sign of N stress in maize plant. Leaf chlorophyll content has a positive correlation with yield and hence can be used to determine yield potential of maize indirectly as reported by Tittonell *et al.* (2004). Reduced leaf chlorophyll content caused by N stress translates to reduced photo assimilate production which is evident in the reduced plant height observed for this study.

The genetic effect controlling grain yield under low-N stress and opt-N environments observed for this study were highly similar. Both GCA and SCA mean squares were found significant indicating that the presence of additive and non-additive genetic influence as compared to the opt-N environment where only non-additive genetic influence was significant. According to Diallo *et al.* (2003), when maize is subjected to environmental stress, its performance is dictated by those genes that are stress responsive as well as stress adaptive. With this understanding, the N stress must have activated some genes responsible for tolerance to act in an additive manner leading to the significant additive genetic contribution observed under low-N stress which was totally absent under opt-N environment. Though this contribution was low as the predominant gene action controlling grain yield was still found to be non-additive gene action. It might have been possible that if the N stress becomes more severe the gene action controlling grain yield could also change. This proposition was made due to the findings of Gallais and Hirel (2004) who reported that in maize, genetic variation expression differs under low-N and high N conditions due to the detection of different Quantitative Traits Loci (QTLs) conditioning yield under the two conditions. The non-additive gene action controlling grain yield found in this study corroborates the findings of Kamara (2015), Abdel-Moneam *et al.* (2014), Meseka *et al.* (2013), Gouda *et al.* (2013), Zare *et al.* (2011), Worku *et al.* (2008), and Betran *et al.* (2003). This result also contradicts the report made by Kling *et al.* (1997) that grain yield is influenced by predominance of additive gene action under low-N condition. The significant SCA mean square for grain yield coupled with high magnitude of non-additive genetic effect and high heterotic values observed under low-N condition suggests that hybrid development could be possible to exploit non-additive genetic influence in N stress growing environments.

In selection for N stress tolerant genotypes, Banziger *et al.* (2000) highlighted some secondary traits which can enhance selection efficiency due to their strong association with grain yield as well as high heritability maintained under N stress condition. The use of secondary traits was proposed by Badu-Apraku *et al.* (2004 and 2005) due to low heritability of grain yield under low-N stress making it difficult for appropriate selection to be carried out. In this study, the secondary traits considered (ASI, LFS, EPP, CHL) were controlled by additive gene action which can make selection to be carried out on these traits of high adaptive values in order to strengthen the decision on the selection of any genotype for tolerance to low-N stress.

Other agronomic characters (DS, PLHT, EDAIM, and EL) showed a predominance of non-additive gene action under both N conditions with the exception of thousand kernel weight (1000KW) where the magnitude of influence of both gene actions were equal under N stress. The observation for PLHT and EDIAM contradicts the findings of Kamara (2015), Worku *et al.* (2008) Abd-El-Hadi *et al.* (2004) and Nigussie and Zelleke (2001) where preponderance of additive gene action was reported. The results also corroborates the findings of Abdel-Moneam *et al.* (2014), El-Badawy (2013), Barakat and Osman (2008), and Katta *et al.* (2007) for EL where non additive genetic effect was also reported to condition the trait.

5.2.2 Drought stress and well-watered environment

Tittonell *et al.* (2004) reported that assessment of a crop performance can either be done directly through grain yield or biomass production at harvest or indirectly through the use of yield correlated agronomic characters such as chlorophyll color intensity (cci). Therefore, the use of biomass yield/production at the expense of grain yield for drought stress evaluation should not be underestimated as it does not in any way change the performance of the genotypes from that obtained when grain yield is considered.

The intensity of drought stress used in this study resulted in biomass yield reduction ranging from 28 % to 71 % with average of about 50 %. The high reduction in biomass yield observed was partly due to ineffective photo assimilate production capacity resulting from the reduced activities of enzyme invertase under drought stress (Banziger *et al.*, 2000). Effect of drought stress translated into yield loss varies with the timing and duration of the stress as reported by many findings. Denmead and Shaw (1960) explained that when drought stress strikes at flowering stage, it can reduce the yield of maize by 50 % which perfectly explains the possibility of the results observed in this study as drought stress was imposed just one week to flowering period. Badu-Apraku *et al.* (2010) also reported 44 % yield loss in the early maturing cultivars evaluated under drought from 2007-2009 and attributed this to mild drought. In another study from the same authors, drought stress was observed to have caused yield reduction by 59 % (Badu-Apraku *et al.*, 2011b). Drought stress caused a greater yield loss (50 %) in both F1 hybrids and parental inbred lines than low-N stress (25 %) as observed in this study. This explains why Banziger *et al.* (2000) ranked drought stress above N stress in abiotic stress ranking.

In selecting for drought tolerant genotypes, Banziger *et al.* (2000) also proposed some secondary traits which could be helpful in the selection process. Of these traits, ASI, LFR, LFS and CHL were considered for this study. The drought intensity observed for this study caused a slight delay in the silk emergence (DS) which in turn resulted in the slight extension of the ASI. Maize silk is more affected under drought stress than tassel which may not be favorable for effective fertilization. This could partially explain the kernel bareness observed in the study.

The significant increase in leaf senescence observed under drought stress compared to well-watered environments is due to disruption of the normal physiological activities in the plants.

Under drought stress, abscisic acid (ABA) is synthesized in the root portion of the plant and increases in magnitude with increasing drought intensity which are transported to the above ground part where it causes leaf stomata closure and leaf rolling (LFR) to prevent excessive evapotranspiration. Banziger *et al.* (2000) reported that as the intensity of drought increases, leaf water loss increases and ultimately results in increased leaf senescence as observed for this study.

The reduction in the CHL under drought stress is attributable to loss of chlorophyll in the plants which directly affects photo-assimilate production (Tittonell *et al.*, 2004). When maize is exposed to drought stress, chlorophyll oxidation occurs due to the free electrons liberated in the photosynthetic pathway. This causes the break of the linkage between the photosystem I and II which is expressed as leaf bleaching as reported by Banziger *et al.* (2000). This result agrees with that of Mansouri-Far *et al.* (2010) who observed reduced leaf greenness in a study comparing the performance of maize genotype under drought and well-watered conditions.

Plant height (PLHT) significantly reduced under drought compared to well-watered environment due to reduced photo assimilate production. Under drought stress, Maize tends to increase root length down the soil profile in search for water. In doing this, photo assimilate produced which should have been used to increase the internode length for increased plant height are used for root growth and extension in an attempt to tolerate the stress condition (Khan *et al.*, 2001). This in turn results in heavier root system as more solute are absorbed during the search for water as well as increased root to shoot ratio (RSR) as observed for this study. This findings agrees with that of Akinwale *et al.* (2016) who proposed the use root length and root weight as parameters that should be considered in selecting for drought tolerant maize genotypes at seedling and vegetative phase rather than waiting till flowering.

Furthermore, Camacho and Caraballo (1994) in a study considered dried root weight as the best indicator for drought tolerance in maize.

Tassel blasting (TB) is a phenomenon that can occur in maize when temperature goes beyond 38 °C as reported by Banziger *et al.* (2000). In this study, the nature of the structure (plant house) could have led to the excessive increase in the observed temperature beyond 38 °C required for TB to occur. This must have been the major reason why the genotypes under drought stress experienced more tassel blasting than well-watered condition genotypes consequently leading to ineffective pollination and complete bareness of some genotypes.

The gene action controlling biomass yield under drought and well-watered environments were similar to that observed under low-N and opt-N environments. Both additive and non-additive genetic influence were also involved in the control of the trait though magnitude of influence of additive gene was higher under drought stress compared to well-watered environments. In spite of the contribution of GCA, non-additive gene action was found to predominate the control of the trait. This study finds it difficult to be compared to previous findings as reports on genetic action conditioning biomass yield was not found. Of the secondary traits that were proposed by Banziger *et al.* (2000), anthesis-silking interval (ASI) showed a preponderance of additive gene action. ASI was consistent with additive gene influence under both stress conditions which was also reflected in the higher narrow sense heritability estimate compared to others secondary traits. This means that ASI will be a trait can be highly depended upon for selection for maize genotype that combines tolerance to low-N and drought stresses. ASI has been generally considered by maize breeders as the secondary trait that is most indicative of drought tolerance. This result is consistent with the previous findings of Messmer *et al.* (2009), Edmeades *et al.* (1993), and Bolanos and

Edmeades (1993), that ASI is a reliable indicator of drought tolerance in maize and Banziger *et al.* (2000) who also proposed ASI for low-N tolerance selection.

Leaf senescence (LFS) and leaf chlorophyll content (CHL) and leaf rolling (LFR) on the other hand were influenced by non-additive gene action under drought stress but LFS and CHL were influenced by additive gene action under well-watered condition. The change in the genetic effect influencing LFS and CHL follows the earlier reports of Blum (1997) who said that adaptation to environmental stress in maize is based on genes that are stress responsive and adaptive.

Of the other agronomic traits, days to silking (DS) and tassel blasting (TB) showed preponderance of additive gene effect under drought stress and well-watered conditions for DS only since TB was not taken for well-watered condition. Plant height (PLHT), root length (ARL), root to shoot ratio (RSR) and dry root weight (DRW) all showed dominance of non-additive gene action under both drought stress and well-watered environments. This results contradicts the findings of Pavan *et al.* (2011) for DS who reported non-additive genetic influence but agrees with the findings of Dawod *et al.* (2012) for PLHT.

5.3 Combining Ability

5.3.1 General Combining Ability (GCA) effect for yield and agronomic traits

General Combining Ability (GCA) revealed parental line TZEEI-76 and TZEE-21 as the best combiners for both grain and biomass yield under both stress conditions considered for the study indicating these lines possessed tolerance to both stresses but TZEEI-76 was undesirable for leaf chlorophyll content. These lines were previously reported by Badu-Apraku and Oyekunle (2012) and Badu-Apraku *et al.* (2011a) to possess drought tolerant

gene which must have influenced their N deficit tolerance as explained by Banziger *et al.* (2000).

On the other hand, PZEEI-6 and TZEEI-29 performed poorly under both stress conditions for both grain and biomass yield. It was surprising to see these lines performed poorly under both stress conditions as their poor performance was in contradiction to the recommendation of Badu-Apraku *et al.* (2011a) as drought tolerant lines.

Earliest silk emergence was observed for TZEEI-21 under N stress but under drought stress was TZEEI-6. Synchronization of the male and female flowers expressed as ASI was earliest for TZEEI-38 under N stress but under drought stress was TZEEI-21. Early nicking of flowers reduces ASI and hence increased pollination and fertilization efficiency as reported by Banziger *et al.* (2000).

Increased plant height has been found to correlate positively with yield as reported by Tittonell *et al.* (2004) which is a character of TZEEI-21 under both stress conditions. This maybe good from agronomic point of view but not desirable from breeding perspective as genotypes with increased height are liable to lodging and ultimately reduces yield. TZEEI-76 and TZEEI-29 with consistently reduced height will be a better choice in plant breeding.

The best combiner for leaf senescence was TZEEI-76 under N stress but TZEEI-21 under drought stress while that of leaf chlorophyll content was TZEEI-29 under N stress and TZEEI-21 under drought stress. A combination of negative ASI, LFS and positive CHL could become a morpho-physiological basis for assessing maize crop yield indirectly. Edmeades *et al.* (1993) proposed the development of selection index with these traits in order to enhance selection under abiotic stress conditions.

Some morphological adaptations of maize plant conditioned by its genetics to drought tolerance have been reported to include increased root length and dry root weight (Akinwale *et al.*, 2016; Camacho and Caraballo (1994). This was true for inbred line TZEEI-76 which had the longest root length, heaviest dry root weight and increased root to shoot ratio. It is still unclear to find this line rated as drought susceptible according to Badu-Apraku *et al.* (2011b).

5.3.2 Specific Combining Ability (SCA) effect for grain and biomass yield

With respect to all environmental conditions observed for this study, crosses TZEEI-6×TZEEI-38, TZEEI-6×TZEEI-76, TZEEI-21×TZEEI-38, TZEEI-21×TZEEI-76, TZEEI-29×TZEEI-76, and TZEEI-38×TZEEI-76 showed stable performance. Among these crosses TZEEI-76 and TZEEI-38 were more prevalent.

Under low-N and drought stress environments as targeted for this study, crosses TZEEI-21×TZEEI-38, TZEEI-21×TZEEI-76, TZEEI-29×TZEEI-76, and TZEEI-38×TZEEI-76 were consistent in performance with higher yield. Of these crosses, TZEEI-76 was more prevalent than any other line indicating its superior yield ability. This may be due to its combining ability which enables the transfer of the high yielding character (genes) into the F1 during hybridization. These crosses could be selected to improve the performance for grain production in the growing environments where both stresses occur.

In addition to the crosses that showed better performance under both stressed conditions, crosses TZEEI-21×TZEEI-38, TZEEI-29×TZEEI-6 showed promising performance under low-N stress indicating they are better adapted to N stress condition and TZEEI-6×TZEEI-29, TZEEI-6×TZEEI-76, and TZEEI-21×TZEEI-29, showed better adaptability to drought

stress condition. Badu-Apraku *et al.* (2011a) also observed the hybrid combination involving TZEEI-29 and TZEEI-21 to be adapted to drought stress condition only.

For the purpose of breeding using indirect selection methods (normal conditions), crosses TZEEI-21×TZEEI-76, TZEEI-29×TZEEI-76, and TZEEI-38×TZEEI-76 showed desirable performance just like under stressed conditions. This confirmed the superiority of TZEEI-76 compared to other lines in hybrid combinations. It is not surprising to see crosses originating from two lines with poor combining ability expressing superior hybrid combination as observed in this study as this has been reported in many genetics studies such as Kamara (2015), Mousa (2014), Katta *et al.* (2013) and Abuali *et al.* (2012). This could be attributable to dominance effect of one line over the expression of the other line for the specified traits.

Estimates of specific combining ability effect for agronomic traits was used to assess the F1 hybrids with respect to different agronomic characters. In this session, only secondary traits were reported due to their importance in enhancing selection in breeding for tolerance to abiotic stress conditions. Of these traits, ASI, LFS and CHL are common for selection for tolerance to both stress conditions used for the study.

The cross TZEEI-29×TZEEI-21 was the only stable genotype with faster synchronization of male and female flowers under both stress conditions compared to other crosses whose performance vary with environmental differences. TZEEI-29×TZEEI-38 was identified for stable performance for reduced leaf senescence under both stress conditions. Reduced senescence is desirable as a requirement for increased photosynthetic capability in maize plant (Tittonell *et al.* 2004).

The cross TZEEI-21×TZEEI-76 showed stable performance for leaf chlorophyll content which is an expression of the stay green properties of the plant. This trait can be used to

indirectly assess the yield potential of maize plant according to Tiftonell *et al.* (2004). This could be due to its important role in photo-assimilates production particularly solar radiation absorption and electron transport chain reaction for carbohydrate/sugar production for assimilate partitioning into growing sinks (Banziger *et al.*, 2000).

5.4 Heterosis for yield

Heterosis is the major factor responsible for the better adaptability of hybrids to stress conditions compared to the inbred lines. Heterosis is a measure of the heterozygous loci present in a hybrid genotype and it increases with increase in the genetic distance between two genotype in a cross combination e.g. between two different heterotic pools (Falconer and Mackay, 1996). In this study, heterosis was expressed more under stress environments compared to the non-stress environments. This was partly due to the poor performance of the parental lines under stressed conditions thereby leaving a wide gap in the average heterotic values.

The magnitude of heterosis varied from hybrid to hybrid indicating a greater potential for increased grain yield. Under low-N and opt-N stress environments, TZEEI-29×TZEEI-38, TZEEI-6×TZEEI-29, TZEEI-6×TZEEI-38 and TZEEI-38×TZEEI-29 were among the crosses that expressed the highest significant heterosis. Under drought and well-watered environments, TZEEI-21×TZEEI-29, TZEEI-21×TZEEI-38, TZEEI-29×TZEEI-21 and TZEEI-38×TZEEI-21 were among the crosses that expressed the highest heterosis. Some hybrid combination were found to have low heterotic values such as TZEEI-21×TZEEI-6 and TZEEI-29×TZEEI-38 under both low and opt-N environments and TZEEI-29×TZEEI-38 under both drought and well-watered environments. This could be attributed to narrow genetic base of the population from which the parents are derived (Hallauer, 1999).

The high heterosis observed in some crosses is attributable to dominance or dominance type of epistasis resulting from variations in allelic frequencies of dominant alleles in the parents forming the hybrids (Falconer and Mackay, 1996). Schon *et al.* (2010) compared QTLs mapping results for grain yield in three population of maize and concluded that high heterosis observed in the hybrids was due to combination of alleles which were originally fixed in each opposite pool. Hallauer and Miranda (1988) suggested a criterion for selecting parental lines with high heterotic response and consequently produce hybrid with high yield performance to be GCA effect. In the case of single cross hybrid, positive GCA may satisfy this criterion of selection.

In this study, consistency was found between SCA effect and mean yield of the hybrids. Corresponding hybrid mean to significant positive SCA effect were found to have high MPH and BPH though some few crosses do not follow the pattern. This explains that GCA effect of the parental lines can predicts the performance of the hybrid possibly better than heterosis as the latter depends largely on the performance of the parental lines. Furthermore, differences in the response of the parental lines to different stress conditions which may be larger than the hybrids will allow heterosis to be inconsistent across the varying stress conditions and environments. Hence, heterosis alone cannot justify the performance of the hybrids used for this study as it was observed that some better yielding hybrids did not show very high heterosis but rather showed their quality from their reduced yield loss when compared to average or low yielding ones with high heterotic values. Very high heterotic value could be as a result of high level of inbreeding in the parental lines as reported by Falconer and Mackay (1996). Hence, hybridization program targeting high yield should consider the use of the GCA effect more than the use of heterosis.

CHAPTER SIX

6.0 CONCLUSIONS AND RECOMMENDATIONS

6.1 Conclusions

The main objective of this study was to improve maize production and productivity in the low rainfall zones of Ghana.

Based on the findings observed from this study, the following conclusions were made;

1. Grain yield reduced by 24 % due to N stress and biomass yield reduced by 48 % due to drought stress.
2. The inbred lines TZEEI-76 and TZEEI-21 were the best combiners for grain and biomass yield under both stress conditions. The best combiner for ASI was TZEEI-38, for PLHT and LFS was TZEEI-76, and for CHL was TZEEI-29 under low-N stress. Under drought stress, the best combiner was for ASI, LFS, CHL and TB was TZEEI-21.

The best crosses that combined tolerance to both stress conditions were TZEEI-21×TZEEI-38, TZEEI-21×TZEEI-76, TZEEI-29×TZEEI-76 and TZEEI-38×TZEEI-76. For tolerance to N stress, crosses TZEEI-29×TZEEI-38 and TZEEI-29×TZEEI-6 were the best alongside the aforementioned crosses.

For drought stress, crosses TZEEI-6×TZEEI-29, TZEEI-6×TZEEI-76 and TZEEI-21×TZEEI-29 were the best alongside those combining tolerance to both stresses.

Non-additive gene action controlled grain and biomass yield under both stress and non-stress conditions, additive gene action controlled all the secondary traits (ASI, LFS, EPP, and CHL) under both N levels and non-additive gene action controlled other agronomic traits. Non-additive gene controlled all the selected traits except for

DS and ASI that are controlled by additive gene action under drought condition. Additive gene action controlled DS, LFS, and CHL while non-additive gene action was responsible for the rest of the traits under well-watered condition.

3. Heterosis was higher under stressed conditions than non-stressed conditions. Low heritability and baker's additive genetic ratio for yield indicated low additive gene action. Hence, selection could be improved by combining yield with secondary traits ASI, LFS and CHL due to their significant genetic correlation with yield as well as moderately high heritability estimates under stressed conditions.
4. Hybrids selection combining tolerance to both stress conditions should focus on using yield in combination with negative LFS and positive CHL.

6.2 Recommendations

1. Further study should be done to identify the gene action that controls grain yield and yield determining characters under more severe condition of low-N such as 0 kg/ha and 30 kg/ha to support the claim of yield variability with low-N variability made in this study.
2. Consideration should be given to determination of genetic differences in the nitrogen uptake efficiency as well as nitrogen use efficiency in the selection of those hybrids that exhibit tolerance to low-N stress.
3. Consideration should be given to evaluation for drought tolerance under field conditions with increased replications and environments to observe the pattern of $G \times E$ and reduce error variance for better heritability estimation.

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APPENDICES

Appendix 1: Weekly mean temperature, relative humidity and rainfall data during the period of evaluation of the genotypes for tolerance to low-N and drought stresses

Period	Max_T	Min_T	RH	Prep.	Period	PH_tempt	NE_tempt
(°C)	(°C)	(%)	(mm)	(°C)	(°C)	(°C)	
wk1	31.23	21.99	86.43	10.60	wk1	37.85	35.14
wk2	31.44	21.40	86.00	4.71	wk2	37.52	34.59
wk3	32.31	21.75	82.13	3.23	wk3	36.94	33.70
wk4	32.00	21.86	85.00	3.40	wk4	37.25	35.36
wk5	31.20	21.87	87.71	2.07	wk5	37.64	35.69
wk6	32.54	21.98	83.88	0.68	wk6	39.86	36.21
wk7	32.23	22.07	86.00	0.24	wk7	40.03	36.33
wk8	32.26	22.33	84.50	0.00	wk8	39.23	36.00
wk9	31.90	20.29	72.57	0.00	wk9	38.20	34.93
wk10	33.81	20.81	71.38	0.00	wk10	37.44	34.69
wk11	33.06	19.81	58.75	0.00	wk11	36.43	34.14
wk12	33.69	20.88	50.50	0.00	wk12	36.83	34.00
Mean	32.31	21.42	77.90	2.08		38.04	35.16

Max_T= maximum temperature; Min_T=minimum temperature; RH=relative humidity; Prep=precipitation (rainfall); PH_tempt=temperature in the plant house between 12 and 3pm; NE_tempt=normal environmental temperature; and wk=week.

Appendix 2: Mean square from ANOVA for yield and agronomic character across all environments combined for the study

Source	DF	DA	DS	ASI	PLHT	EHT	EPO	CHL	LFS	PASP	MSV	YLD
Env (E)	3	872.58***	827.92***	1.58***	4215***	413.33***	0.18***	13525***	6.42***	1.22***	0.15***	72.98***
Entry (G)	24	45.68***	24.14***	1.08***	2952***	931.80***	0.02***	252.85***	0.46***	0.35***	0.35***	9.26***
GCA	4	101.96***	17.47*	2.46***	2070***	237.15**	0.06***	517.21***	1.16***	0.60***	1.30***	5.02***
SCA	10	56.10***	41.46***	1.30***	5879***	1866***	0.02***	235.68***	0.41***	0.46***	0.25***	17.59***
REC	10	10.22	5.68	0.40*	249	192.28**	0.01*	64.36**	0.27***	0.18***	0.08**	0.78
G x E	72	21.79***	20.18***	0.45***	384***	214.59***	0.01***	74.40***	0.20***	0.11***	0.02	1.10***
GCA x E	12	36.86***	33.68***	0.79***	221	193.16***	0.01***	117.08***	0.36***	0.15***	0.01	1.65***
SCA x E	30	29.27***	25.17***	0.39***	516***	286.07***	0.01***	86.99***	0.18***	0.10***	0.02	1.43***
REC x E	30	8.29	9.79	0.36*	316*	151.68***	0.01**	44.64**	0.13*	0.11***	0.02	0.56
Error	240	6.13	7.09	0.17	180.21	58.43	0.00	14.10	0.07	0.05	0.02	0.55
CV%		4.59	4.54	18.71	13.36	13.97	10.15	15.63	12.34	11.59	10.18	24.49
Mean		53.92	58.65	4.73	100.51	54.72	0.55	24.02	4.41	3.07	1.85	3.04
GCA/SCA		1.82	0.42	1.89	0.35	0.13	3.00	2.19	2.83	1.30	5.20	0.29
h ²		10.25	21.32	31.38	1.75	2.50	48.48	29.65	39.97	18.10	27.03	3.41
H		63.62	52.04	63.46	25.30	34.66	66.67	53.11	64.77	66.02	66.59	66.36
BR		0.78	0.47	0.82	0.41	0.20	0.88	0.81	0.82	0.70	0.91	0.36

*, **, *** probability levels at 0.05, 0.01 and 0.001 respectively

Env=environment; GCA=general combining ability; SCA= specific combining ability; DA=days to 50% anthesis; DS=days to 50% silking; ASI= anthesis-silking interval; PLHT=plant height; EHT= ear height; EPO= ear position; CHL= leaf chlorophyll content; LFS= leaf senescence; PASP= plant aspect; MSV= maize streak virus disease; and YLD= yield.

**Appendix 3: General combining ability effects of parental lines for agronomic traits
under optimal N (above) and well-water (below) environments**

Parent	DS	ASI	PLHT	EHT	LFS	CHL	EPP	EDIAM	EL	1000KW	GY
TZEEI-6	0.20	1.41***	2.16	-0.76	0.65***	-4.20***	0.06	-0.1	0.42	-2.41	-0.48*
TZEEI-21	-0.07	0.15	7.23*	2.72	-0.17	2.80***	-0.08	0.02	-0.03	-0.55	-0.17
TZEEI-29	0.00	-0.05	-5.18	-1.87	-0.52**	4.36***	-0.09	-0.04	-0.30	8.39**	-0.05
TZEEI-38	-0.37	-0.82**	3.47	2.56	0.50**	-0.07	0.07	0.12	0.054*	0.42	0.47*
TZEEI76	0.23	-0.69**	-7.68**	-2.65	-0.46**	-2.90***	0.04	0.00	-0.63*	-5.85*	0.23
SE(Gi)	0.36	0.24	2.77	1.32	0.17	0.70	0.05	0.06	0.23	2.57	0.22
SE (Gi – Gj)	0.57	0.39	4.39	2.09	0.27	1.10	0.08	0.10	0.37	4.06	0.35

Parent	DS	ASI	PLHT	EHT	LFS	CHL	TB	ARL	RSR	DRW	BYLD
TZEEI-6	-0.88	-0.18	0.88	-0.76	0.39*	-0.17	-0.06*	1.03	0.00	0.78***	-0.03
TZEEI-21	0.59	0.74*	6.20***	2.72*	-0.16	-0.30	0.01	-0.12	-0.04*	0.15	0.12**
TZEEI-29	1.22*	0.32	-0.47	0.14	-0.36	1.92***	0.04	0.26	0.02	-0.41*	-0.06
TZEEI-38	-1.56**	-0.85**	-1.39	-0.79	-0.31	0.35	0.09***	-2.22	-0.01	-0.14	-0.05
TZEEI76	0.64	-0.03	-5.23**	-1.31	0.44*	-1.79***	-0.06*	1.06	0.04*	-0.39*	0.02
SE(Gi)	0.56	0.33	1.83	1.17	0.11	0.53	0.02	1.19	0.01	0.13	0.03
SE (Gi – Gj)	0.88	0.53	2.90	1.85	0.17	0.84	0.04	1.88	0.02	0.21	0.05

*, **, *** probability levels at 0.05, 0.01 and 0.001 respectively

DS= days to 50% silking; ASI= anthesis-silking interval; PLHT= plant height; EHT= ear height; LFS= leaf senescence; CHL= chlorophyll content of leaf; EPP=ears per plant; EDIAM= ear diameter; EL= ear length; 1000KW= weight of a thousand grain; GY= grain yield; TB= tassel blasting; ARL= average root length; RSR= root to shoot ratio; DRW= dry root weight; and BYLD= biomass yield

Appendix 4: General combining ability effects of parental lines for agronomic traits across moisture variation experiment (above) and across nitrogen variation experiment (below)

Parent	DS	ASI	PLHT	EHT	LFS	CHL	TB	ARL	RSR	DRW	BYLD
TZEEI-6	-1.12**	0.69**	-0.1	-2.97**	0.44**	0.03	-0.05	-1.24	-0.01	-0.36	-0.12
TZEEI-21	1.05**	-0.01	4.89**	0.5	-0.31*	0.06	-0.10**	0.69	-0.03	0.35	0.14
TZEEI-29	0.24	-0.01	-1.46	0.36	-0.24	1.12*	0.00	0.01	0.01	-0.27	-0.08
TZEEI-38	-0.36	-0.48*	-0.21	1.2	-0.06	0.2	0.11**	-1.94	-0.02	-0.01	-0.07
TZEEI76	0.2	-0.2	-3.12*	0.9	0.17	-1.42**	0.04	2.48	0.05*	0.28	0.13
SE(Gi)	0.75	0.56	1.38	1.93	0.09	0.43	0.06	1.26	0.01	0.60	0.06
SE (Gi – Gj)	1.19	0.89	2.18	3.05	0.15	0.69	0.10	1.99	0.02	0.95	0.11

Parent	DS	ASI	PLHT	EHT	LFS	CHL	EPP	EDIAM	EL	1000KW	GY
TZEEI-6	0.3	1.42***	3.6	-0.37	0.65***	-3.97***	-0.10*	-0.11**	0.41**	-4.70**	-0.48***
TZEEI-21	-0.43	-0.03	5.76**	1.85	-0.2	2.99***	-0.04	0.09*	0.16	2.56	0.06
TZEEI-29	0.1	-0.06	-4.43*	-1.2	-0.45***	4.39***	0.05	-0.02	-0.35*	9.76***	-0.1
TZEEI-38	0.07	-0.81***	1.71	1.67	0.72***	-0.75	0.01	0.01	0.12	-3.52*	0.18
TZEEI76	-0.03	-0.53**	-6.64***	-1.94	-0.71***	-2.66***	0.08	0.03	-0.35*	-4.10*	0.35*
SE(Gi)	0.28	0.10	1.33	0.72	0.15	0.34	0.03	0.06	0.24	2.65	0.17
SE (Gi – Gj)	0.45	0.16	2.11	1.15	0.24	0.54	0.05	0.09	0.38	4.20	0.27

*,**,*** probability levels at 0.05, 0.01 and 0.001 respectively

DS= days to 50% silking; ASI= anthesis-silking interval; PLHT= plant height; EHT= ear height; LFS= leaf senescence; CHL= chlorophyll content of leaf; EPP=ears per plant; EDIAM= ear diameter; EL= ear length; 1000KW= weight of a thousand grain; GY= grain yield; TB= tassel blasting; ARL= average root length; RSR= root to shoot ratio; DRW= dry root weight; and BYLD= biomass yield

Appendix 5: Specific combining ability effect of F1 hybrid crosses for yield and agronomic traits evaluated under low-N environment

Crosses	DS	ASI	PLHT	EHT	LFS	CHL	EPP	EDIAM	EL	1000KW	GY
TZEEI-6×TZEEI-21	0.067	-0.10	9.34	1.93	-0.93*	-0.24	0.06	0.08	0.54	-6.74	0.01
TZEEI-6×TZEEI-29	-1.10*	0.60	0.72	2.54	0.70	-1.07	-0.02	0.09	0.27	25.63***	0.22
TZEEI-6×TZEEI-38	-1.57***	-0.17	12.19*	3.67	-0.17	1.89	0.09	0.25*	1.74***	2.06	0.54
TZEEI-6×TZEEI-76	-5.13***	-0.87	36.51*	24.78***	-0.06	3.29	-0.32*	0.83***	2.71***	33.53***	1.19
TZEEI-21×TZEEI-29	0.10	0.40	-1.67	5.10	0.02	1.08	-0.06	-0.03	0.11	-7.55	-0.20
TZEEI-21×TZEEI-38	-0.87	0.47	15.33**	10.67***	0.72	-0.29	-0.01	0.40***	1.72***	15.73**	0.85**
TZEEI-21×TZEEI-76	-4.50***	-0.50	36.09**	19.28***	-0.48	11.51**	0.01	0.71***	4.47***	29.70**	2.53***
TZEEI-29×TZEEI-38	-1.37**	-0.17	5.66	4.67	-0.89*	5.68**	0.04	0.28**	0.97**	8.59	0.63*
TZEEI-29×TZEEI-76	-6.17***	1.30	31.24**	29.26***	0.01	13.51***	-0.26	0.84***	4.27***	46.33***	2.50***
TZEEI-38×TZEEI-76	-5.53***	-0.27	26.46*	12.93*	-1.52	13.04***	0.07	1.14***	3.98***	24.90**	2.10**
TZEEI-21×TZEEI-6	-1.33*	-0.33	19.65**	11.32**	-0.19	3.57	-0.14	0.15	1.22**	13.17*	0.47
TZEEI-29×TZEEI-6	-0.83	-1.17*	3.72	0.38	-0.56	5.43*	-0.02	0.18	0.74	16.00**	0.91*
TZEEI-29×TZEEI-21	-0.50	-1.00*	-0.15	-6.27	0.28	2.88	0.01	-0.02	-0.36	4.17	-0.16
TZEEI-38×TZEEI-6	-0.67	0.00	3.68	6.68*	-0.09	0.43	0.04	0.11	-0.15	-0.83	0.28
TZEEI-38×TZEEI-21	-0.17	0.00	0.08	1.23	0.28	-5.17*	-0.15	-0.09	-0.37	-4.17	-0.57
TZEEI-38×TZEEI-29	-0.67	-0.83	4.40	-0.40	0.00	-1.08	-0.05	0.11	0.45	7.50	0.14
TZEEI-76×TZEEI-6	-1.17*	0.33	5.27	2.78	-0.19	1.87	0.02	0.22	0.40	11.17*	0.36
TZEEI-76×TZEEI-21	1.00	0.33	0.30	1.99	-0.09	-1.38	-0.09	-0.06	-0.43	-2.00	-0.35
TZEEI-76×TZEEI-29	-0.33	0.67	-3.68	2.38	0.19	-5.92**	0.02	-0.04	0.03	1.83	-0.29
TZEEI-76×TZEEI-38	0.00	-0.17	0.73	-4.39	-0.46	-0.92	0.02	0.07	-0.26	-2.33	0.12
SE(S_{ij})	0.43	0.36	5.56	2.71	0.31	1.50	0.07	0.09	0.26	4.53	0.28
SE(S_{ij}-S_{ki})	0.57	0.48	7.39	3.60	0.42	2.00	0.10	0.12	0.35	6.02	0.37
SE (R_{ij})	0.52	0.44	6.74	3.29	0.38	1.82	0.09	0.11	0.32	5.50	0.34

*, **, *** probability levels at 0.05, 0.01 and 0.001 respectively
DS= days to 50% silking; ASI= anthesis-silking interval; PLHT= plant height; EHT= ear height; LFS= leaf senescence; CHL= chlorophyll content of leaf; EPP=ears per plant; EDIAM= ear diameter; EL= ear length; 1000KW= weight of a thousand grain; and GY= grain yield

Appendix 6: Specific combining ability effect of F1 hybrid crosses for yield and agronomic traits evaluated under Optimal N environment

Crosses	DS	ASI	PLHT	EHT	LFS	CHL	EPP	EDIAM	EL	1000KW	GY
TZEEI-6×TZEEI-21	-0.27	1.19*	13.43*	10.12**	0.07	-1.32	0.01	-0.00	0.22	-5.02	-0.05
TZEEI-6×TZEEI-29	-1.50	0.22	0.95	-2.02	0.15	-0.25	-0.17	0.19	0.36	10.21	0.22
TZEEI-6×TZEEI-38	-0.80	-0.01	10.25	3.76	-0.22	3.17*	0.05	0.17	1.42**	2.68	0.64
TZEEI-6×TZEEI-76	-3.7*	1.43	24.45*	14.60*	-0.74	9.23**	0.18	0.59*	1.64	0.10	0.85
TZEEI-21×TZEEI-29	0.43	0.82	5.54	3.68	-0.61	0.19	0.15	-0.01	0.40	-6.32	-0.05
TZEEI-21×TZEEI-38	-0.20	0.09	4.39	4.89	0.22	0.07	-0.10	0.14	0.59	12.81*	0.38
TZEEI-21×TZEEI-76	-5.30***	-0.33	54.20***	28.91***	-0.35	5.23	-0.29	0.82**	5.03***	34.63**	3.31***
TZEEI-29×TZEEI-38	-1.43	-0.21	11.58	3.07**	-0.35	4.96**	0.06	0.38**	0.87	16.71**	1.13*
TZEEI-29×TZEEI-76	-4.57**	3.47**	39.75**	28.27***	-0.89	15.71***	-0.11	1.46***	4.59***	58.57***	2.56**
TZEEI-38×TZEEI-76	-4.10**	-1.30	25.44*	14.61*	-1.35	13.45***	0.42	0.72**	2.87**	22.43*	2.99**
TZEEI-21×TZEEI-6	-1.00	-1.67*	9.08	3.77	-0.74	5.30**	-0.27	0.32	1.10	19.17**	0.14
TZEEI-29×TZEEI-6	0.17	0.17	14.12	6.50	-0.65	8.77***	-0.09	0.16	0.62	11.67	0.90
TZEEI-29×TZEEI-21	0.50	-0.20	-14.08	-9.71*	0.37	-3.33	0.18	-0.25	-0.32	-12.00	0.01
TZEEI-38×TZEEI-6	0.17	1.50*	8.40	8.75*	0.56	-4.82**	0.13	0.12	-0.23	-0.83	-0.08
TZEEI-38×TZEEI-21	-1.50	-0.67	7.05	5.23	0.19	-1.98	0.06	0.11	-0.16	2.83	0.59
TZEEI-38×TZEEI-29	-0.67	-0.83	1.80	2.57	-0.19	0.57	-0.04	0.01	0.38	5.33	0.68
TZEEI-76×TZEEI-6	-2.67**	0.00	11.92	5.48	-0.37	2.07	-0.25	0.25	0.47	13.67*	0.23
TZEEI-76×TZEEI-21	0.33	0.50	14.27*	8.46*	0.65	0.93	-0.05	0.07	0.12	10.00	-0.20
TZEEI-76×TZEEI-29	0.67	0.83	-1.98	2.13	-0.28	-4.02*	0.05	-0.02	0.20	-6.00	-0.19
TZEEI-76×TZEEI-38	0.50	0.17	4.57	0.83	-0.09	1.80	-0.27	0.02	0.04	0.83	0.33
SE(S_{ij})	0.75	0.51	5.73	2.73	0.35	1.44	0.10	0.13	0.49	5.30	0.46
SE(S_{ij}-S_{ki})	0.99	0.67	7.61	3.63	0.47	1.92	0.14	0.17	0.65	7.04	0.61
SE (R_{ij})	0.91	0.62	6.94	3.31	0.43	1.75	0.13	0.16	0.59	6.42	0.56

*, **, *** probability levels at 0.05, 0.01 and 0.001 respectively

DS= days to 50% silking; ASI= anthesis-silking interval; PLHT= plant height; EHT= ear height; LFS= leaf senescence; CHL= chlorophyll content of leaf; EPP=ears per plant; EDIAM= ear diameter; EL= ear length; 1000KW= weight of a thousand grain; and GY= grain yield

Appendix 7: Specific combining ability effect of F1 hybrid crosses for yield and agronomic traits evaluated across N variation environments

Crosses	DS	ASI	PLHT	EHT	LFS	CHL	EPP	EDIAM	EL	1000KW	GY
TZEEI-6×TZEEI-21	-0.10	0.54	11.39**	6.03**	-0.43	-0.78	0.04	0.04	0.38	-5.88	-0.02
TZEEI-6×TZEEI-29	-1.30**	0.41	0.83	0.26	0.43	-0.66	-0.10	0.14	0.31	17.92***	0.22
TZEEI-6×TZEEI-38	-1.18*	-0.09	11.22**	3.72	-0.19	2.53*	0.07	0.21*	1.58***	2.37	0.59*
TZEEI-6×TZEEI-76	-4.42***	0.28	30.48***	19.69***	-0.39	6.27*	0.07	0.71***	2.17***	16.82*	1.02
TZEEI-21×TZEEI-29	0.57	0.61	1.94	4.39*	-0.29	0.64	0.05	-0.02	0.25	-6.93	-0.12
TZEEI-21×TZEEI-38	-0.53	0.28	9.86*	7.78***	0.47	-0.11	-0.06	0.27**	1.16***	14.27***	0.61*
TZEEI-21×TZEEI-76	-4.90***	-0.42	45.14***	24.09***	-0.42	8.37**	-0.14	0.77***	4.75***	32.17***	2.92***
TZEEI-29×TZEEI-38	-1.40**	-0.19	8.62*	6.92**	-0.62*	5.32***	0.05	0.33***	0.92**	12.65***	0.88**
TZEEI-29×TZEEI-76	-5.37***	2.38**	35.50***	28.77***	-0.44	14.61***	-0.18	1.15***	4.58***	52.45***	2.53***
TZEEI-38×TZEEI-76	-4.82***	-0.78	25.95***	13.77**	-1.44**	13.24***	0.24	0.93***	3.43***	23.67***	2.55***
TZEEI-21×TZEEI-6	-1.17	-1.00*	14.37**	7.54**	-0.46	4.43**	-0.20	0.23*	1.16**	16.17***	0.30
TZEEI-29×TZEEI-6	-0.33	-0.50	8.92	3.44	-0.60	7.10***	-0.05	0.17	0.68	13.83**	0.90*
TZEEI-29×TZEEI-21	0.00	-0.75	-7.12	-7.99**	0.32	-0.23	0.09	-0.13	-0.34	-3.91	-0.06
TZEEI-38×TZEEI-6	-0.25	0.75	6.04	7.72**	0.23	-2.19	0.08	0.11	-0.19	-0.83	0.10
TZEEI-38×TZEEI-21	-0.83	-0.33	3.57	3.23	0.23	-3.58*	-0.05	0.01	-0.26	-0.67	0.01
TZEEI-38×TZEEI-29	-0.67	-0.83	3.10	1.08	-0.09	-0.26	-0.04	0.06	0.41	6.42	0.41
TZEEI-76×TZEEI-6	-1.92**	0.17	8.59	4.13	-0.28	1.97	-0.11	0.23*	0.43	12.41**	0.29
TZEEI-76×TZEEI-21	0.33	0.42	7.28	5.23*	0.28	-0.23	-0.07	0.01	-0.15	4.00	-0.27
TZEEI-76×TZEEI-29	0.17	0.75	-2.83	2.26	-0.05	-4.97**	0.03	-0.03	0.11	-2.08	-0.24
TZEEI-76×TZEEI-38	0.25	0.00	2.65	-1.78	-0.28	0.44	-0.14	0.05	-0.11	-0.75	0.22
SE(S_{ij})	0.43	0.31	3.99	1.92	0.23	1.04	0.06	0.08	0.27	3.48	0.27
SE(S_{ij}-S_{kl})	0.57	0.41	5.30	2.56	0.31	1.38	0.09	0.10	0.37	4.63	0.36
SE (R_{ij})	0.52	0.38	4.84	2.33	0.28	1.26	0.08	0.09	0.33	4.23	0.32

*, **, *** probability levels at 0.05, 0.01 and 0.001 respectively DS= days to 50% silking; ASI= anthesis-silking interval; PLHT= plant height; EHT= ear height; LFS= leaf senescence; CHL= chlorophyll content of leaf; EPP=ears per plant; EDIAM= ear diameter; EL= ear length; 1000KW= weight of a thousand grain; and GY= grain yield

Appendix 8: Specific combining ability effect of F1 hybrid crosses for yield and agronomic traits evaluated under drought stress environments

Crosses	DS	ASI	PLHT	EHT	LFR	LFS	CHL	TB	ARL	RSR	DRW	BYLD
TZEEI-6×TZEEI-21	-0.49	2.33***	-6.55*	-5.47*	-0.39*	0.51	-0.27	-0.14	2.10	0.06	-2.98***	-0.05
TZEEI-6×TZEEI-29	0.98	1.97***	2.23	-1.14	-0.04	-0.84*	1.36	-0.23**	-0.90	-0.03	3.32***	0.26***
TZEEI-6×TZEEI-38	0.11	-0.38	9.56**	0.26	-0.14	-0.89*	1.03	0.09	0.88	-0.06	-0.97*	0.15
TZEEI-6×TZEEI-76	2.54	0.92	-10.20	-8.68*	0.03	-1.90**	-1.28	-0.01	27.23***	0.41***	7.29***	0.96***
TZEEI-21×TZEEI-29	0.81	-0.33	5.33	3.14	0.32	0.11	-1.59	0.14	15.35***	0.13*	6.22***	0.72***
TZEEI-21×TZEEI-38	2.43	-0.01	2.91	0.79	0.43*	-1.69***	4.06**	-0.20*	10.63**	0.10*	4.72***	0.54***
TZEEI-21×TZEEI-76	0.51	2.14*	44.45***	11.43**	-0.76*	-4.85***	6.66***	-1.01***	29.73***	0.02	7.27***	1.19***
TZEEI-29×TZEEI-38	-3.18	-2.03***	3.43	3.49	-0.06	-0.79*	-1.35	-0.11	7.25	0.06	-2.60***	-0.35***
TZEEI-29×TZEEI-76	2.66	-0.68	5.05	2.10	-0.05	-1.00	-2.02	-0.06	30.60***	0.31**	4.15***	0.89***
TZEEI-38×TZEEI-76	2.58	-1.42	7.85	4.95	-0.14	0.05	-3.08	-0.35*	29.45***	0.29**	9.96***	1.17***
TZEEI-21×TZEEI-6	0.50	2.00**	-7.38*	-6.25*	-0.31	-0.50	0.40	-0.06	-6.00	-0.01	-0.45	-0.15
TZEEI-29×TZEEI-6	1.17	1.50*	-6.38	-3.63	0.31	0.00	-0.79	0.13	-4.50	-0.01	0.71	-0.03
TZEEI-29×TZEEI-21	-0.67	-1.96**	2.13	-1.63	-0.13	-1.00*	0.72	0.03	-4.75	-0.06	-1.24*	-0.18*
TZEEI-38×TZEEI-6	-2.54**	-0.04	-5.13	-5.88*	0.38	0.25	-0.76	0.13	3.38	0.06	0.69	0.04
TZEEI-38×TZEEI-21	-1.13	-0.75	1.13	-1.38	-0.13	0.00	0.31	0.06	-2.88	-0.04	-1.16*	-0.05
TZEEI-38×TZEEI-29	0.21	-1.17	1.13	-2.88	-0.06	0.75	-0.59	-0.31**	-0.25	-0.02	-0.12	0.07
TZEEI-76×TZEEI-6	2.00*	1.67*	-6.63	-7.38**	-0.06	1.00*	1.24	0.19	-3.34	0.06	1.02	0.14
TZEEI-76×TZEEI-21	-1.25	-1.08	6.13	4.75	-0.69**	-1.50***	-0.04	0.00	2.63	-0.02	0.50	0.15
TZEEI-76×TZEEI-29	-1.33	0.54	-2.00	-2.63	0.00	-1.00*	-1.25	0.25*	-4.00	-0.03	0.34	0.14
TZEEI-76×TZEEI-38	-0.83	-1.33*	-2.88*	-1.38	-0.25	-1.75***	0.24	0.00	1.50	0.10	1.17*	0.14
SE(S_{ij})	0.91	0.62	3.35	2.51	0.19	0.14	0.95	0.07	3.76	0.04	0.41	0.06
SE(S_{ij}-S_{kl})	1.21	0.83	4.45	3.34	0.26	0.23	1.26	0.10	5.00	0.06	0.54	0.08
SE (R_{ij})	1.10	0.75	4.06	3.04	0.24	0.21	1.15	0.09	4.56	0.06	0.49	0.08

*, **, *** probability levels at 0.05, 0.01 and 0.001 respectively

DS= days to 50% silking; ASI= anthesis-silking interval; PLHT= plant height; EHT= ear height; LFS= leaf senescence; CHL= chlorophyll content of leaf; TB= tassel blasting; ARL= average root length; RSR= root to shoot ratio; DRW= dry root weight; and BYLD= biomass yield

Appendix 9: Specific combining ability effect of F1 hybrid crosses for yield and agronomic traits evaluated under well-watered environment

Crosses	DS	ASI	PLHT	EHT	LFS	CHL	TB	ARL	RSR	DRW	BYLD
TZEEI-6×TZEEI-21	-1.14	-0.52	6.25	2.71	-0.09	-1.43	0.01	3.30	0.00	-0.55	-0.04
TZEEI-6×TZEEI-29	-0.14	2.03**	8.67*	3.53	0.61	2.18*	-0.04	1.05	-0.04	1.56***	0.43***
TZEEI-6×TZEEI-38	1.13	0.83	9.72**	4.33*	-0.44	0.02	-0.09	1.15	-0.04	0.99**	0.25**
TZEEI-6×TZEEI-76	-2.77	0.61	15.23*	11.30**	0.20	3.05	-0.00	7.48	-0.01	4.35***	0.82***
TZEEI-21×TZEEI-29	2.14*	0.61	4.98	3.31	0.16	-2.22*	-0.09	-3.81	-0.07*	-0.06	0.26**
TZEEI-21×TZEEI-38	-2.34*	1.03	8.65*	2.73	-0.14	3.75***	-0.14*	5.67*	0.01	3.35***	0.35***
TZEEI-21×TZEEI-76	-1.21	4.50	48.25***	19.55***	-1.84*	5.48**	-0.20	4.20	-0.21**	6.63***	1.85***
TZEEI-29×TZEEI-38	-1.97	-2.17***	16.82***	12.32***	-0.44	3.80***	-0.19**	3.17	-0.07*	-0.93	0.16
TZEEI-29×TZEEI-76	-1.04	0.98	56.01***	32.82***	0.95	0.48	-0.40**	21.45**	-0.17*	4.09***	1.73***
TZEEI-38×TZEEI-76	-2.95	-1.94	48.58***	39.40***	0.50	3.92*	-0.60**	10.97*	-0.18**	1.02	1.45***
TZEEI-21×TZEEI-6	-0.63	-1.63*	0.25	3.13	0.00	-0.65	0.00	-1.88	-0.02	0.35	0.14
TZEEI-29×TZEEI-6	-0.75	-0.75	2.50	4.38	0.00	-2.68*	0.00	0.75	-0.02	0.35	0.09
TZEEI-29×TZEEI-21	0.00	1.00	2.88	0.88	0.50	-1.18	0.00	1.25	0.00	-0.50	0.18
TZEEI-38×TZEEI-6	-0.25	0.63	1.38	6.00*	0.50	-2.23	0.00	2.37	0.01	0.55	0.07
TZEEI-38×TZEEI-21	0.00	-0.25	4.38	1.63	0.25	-0.20	0.00	-1.75	-0.03	-0.31	0.15
TZEEI-38×TZEEI-29	0.00	0.13	-0.88	4.88	0.75	0.69	0.00	3.88	0.04	0.29	-0.10
TZEEI-76×TZEEI-6	0.75	-1.25	-2.63	-0.75	0.25	2.80*	0.00	2.75	0.04	-0.20	-0.03
TZEEI-76×TZEEI-21	2.59*	-0.03	4.08	7.27**	-1.74***	-2.00	0.00	-2.63	-0.04	0.25	0.09
TZEEI-76×TZEEI-29	1.38	1.13	-5.00	-2.13	0.25	0.64	0.00	-2.25	0.01	-0.29	-0.11
TZEEI-76×TZEEI-38	1.50	0.63	5.75	3.13	-0.75	0.45	0.00	1.75	-0.02	0.52	0.10
SE(S_{ij})	1.15	0.69	3.78	2.41	0.23	1.10	0.06	2.45	0.02	0.27	0.07
SE(S_{ij}-S_{ki})	1.54	0.92	5.02	3.21	0.31	1.46	0.07	3.26	0.04	0.36	0.09
SE (R_{ij})	1.40	0.84	4.58	2.93	0.28	1.33	0.07	2.97	0.03	0.33	0.08

*, **, *** probability levels at 0.05, 0.01 and 0.001 respectively

DS= days to 50% silking; ASI= anthesis-silking interval; PLHT= plant height; EHT= ear height; LFS= leaf senescence; CHL= chlorophyll content of leaf; TB= tassel blasting; ARL= average root length; RSR= root to shoot ratio; DRW= dry root weight; and BYLD= biomass yield

Appendix 10: Specific combining ability effect of F1 hybrid crosses for yield and agronomic traits evaluated across moisture variation environments

Crosses	DS	ASI	PLHT	EHT	LFS	CHL	TB	ARL	RSR	DRW	BYLD
TZEEI6×TZEEI-21	-0.81	0.90	-0.15	-1.14	0.21	-0.85	-0.07	2.69	0.03	-1.77**	-0.05
TZEEI6×TZEEI-29	0.42	2.00***	5.45	1.20	-0.12	1.77	0.14	0.07	-0.04	2.44***	0.34
TZEEI6×TZEEI-38	0.62	0.23	9.64**	2.30	-0.66	0.53	-0.00	1.01	-0.05	0.01	0.20
TZEEI6×TZEEI-76	-0.11	0.76	2.51	1.31	-0.85	0.89	-0.01	17.35**	0.19*	5.82***	0.89*
TZEEI21×TZEEI29	1.47*	0.14	5.15	3.22	0.13	-1.90	0.02	5.77*	0.03	3.08***	0.49*
TZEEI21×TZEEI38	0.05	0.51	5.78	1.76	-0.92**	3.91***	-0.17*	8.15**	0.05	4.04***	0.44*
TZEEI21×TZEEI76	-0.35	3.32**	46.35***	15.49***	-3.34**	6.07**	-0.60**	16.96**	-0.09	6.94***	1.52***
TZEEI29×TZEEI38	-2.57***	-2.10**	10.12**	7.90***	-0.61	1.23	-0.15*	5.21	-0.00	-1.76**	-0.10
TZEEI29×TZEEI76	0.81	0.15	30.52***	17.46***	-0.03	-0.77	-0.23	26.03***	0.07	4.12**	1.31**
TZEEI38×TZEEI-76	-0.18	-1.68	28.22***	22.17***	0.27	0.42	-0.47**	20.21***	0.05	5.49***	1.31**
TZEEI21×TZEEI-6	-0.06	0.19	-3.56	-1.56	-0.25	-0.13	-0.03	-3.94	-0.02	-0.05	-0.01
TZEEI-29×TZEEI6	0.21	0.38	-1.94	0.38	0.00	-1.73	0.06	-1.88	-0.01	0.53	0.03
TZEEI29×TZEEI21	-0.33	-0.48	2.50	-0.38	-0.25	-0.23	0.01	-1.75	-0.03	-0.87	-0.00
TZEEI-38×TZEEI6	-1.40	0.29	-1.88	0.06	0.38	-1.49	0.06	2.88	0.03	0.62	0.05
TZEEI38×TZEEI21	-0.56	-0.50	2.75	0.13	0.13	0.06	0.03	-2.31	-0.03	-0.73	0.05
TZEEI38×TZEEI29	0.10	-0.52	0.13	1.00	0.75	0.05	-0.16	1.81	0.01	0.08	-0.02
TZEEI76×TZEEI-6	1.38	0.21	-4.63	-4.06	0.63	2.02	0.09	-0.31	0.05	0.41	0.05
TZEEI76×TZEEI21	0.67	-0.56	5.10	6.01*	-1.62***	-1.02	0.00	0.00	-0.03	0.37	0.12
TZEEI76×TZEEI29	0.02	0.83	-3.50	-2.38	-0.38	-0.31	0.13	-3.13	-0.01	0.03	0.01
TZEEI76×TZEEI38	0.33	-0.35	-1.56	0.88	-1.25**	0.34	0.00	1.63	0.04	0.85	0.12
SE(S_{ij})	0.63	0.40	2.18	1.51	0.32	0.67	0.05	2.24	0.02	0.24	0.04
SE(S_{ij}-S_{kl})	0.84	0.53	2.90	2.00	0.35	0.89	0.07	2.98	0.03	0.32	0.07
SE (R_{ij})	0.77	0.49	2.65	1.83	0.32	0.81	0.06	2.72	0.03	0.30	0.06

***, ***, ** probability levels at 0.05, 0.01 and 0.001 respectively

DS= days to 50% silking; ASI= anthesis-silking interval; PLHT= plant height; EHT= ear height; LFS= leaf senescence; CHL= chlorophyll content of leaf; TB= tassel blasting; ARL= average root length; RSR= root to shoot ratio; DRW= dry root weight; and BYLD= biomass yield

Appendix 11: Mean yield, heterosis and SCA effects of F1 hybrid crosses evaluated under stressed and non-stressed conditions

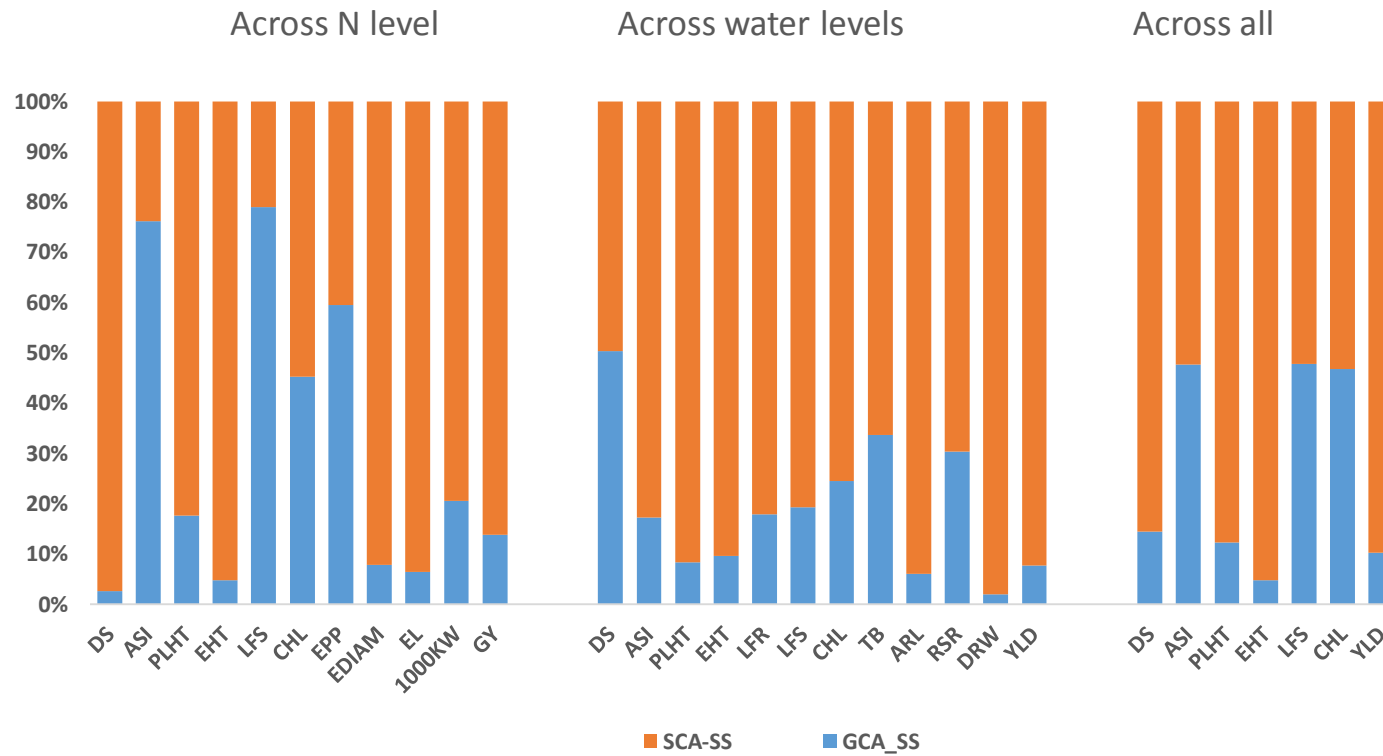
Parent	Grain Yield								Biomass Yield								
	Mean	Low-N			Opt-N				Mean	Dro-str				Well-W			
		MPH	BPH	SCA	Mean	MPH	BPH	SCA		Mean	MPH	BPH	SCA	Mean	MPH	BPH	SCA
TZEEI6×TZEEI21	3.04	135.66	73.71	0.01	3.11	82.94	66.31	-0.05	1.68	77.78	68.00	-0.05	4.06	35.33	32.25	-0.04	
TZEEI6×TZEEI29	3.27	275.86	259.34	0.22	4.24	142.98	126.74	0.22	1.89	100.00	89.00	0.26	4.28	55.07	39.41	0.43	
TZEEI6×TZEEI38	3.00	351.13	261.45	0.54	4.22	116.41	107.88	0.64	1.84	94.71	84.00	0.15	4.10	43.36	33.55	0.25	
TZEEI6×TZEEI-76	3.33	172.95	106.83	1.19	3.65	115.34	95.19	0.85	2.44	92.13	47.88	0.96	3.89	44.07	26.71	0.82	
TZEEI21×TZEEI76	2.55	91.73	45.71	-0.21	3.40	115.87	109.88	-0.05	2.53	153.00	153.00	0.72	4.37	62.45	49.15	0.26	
TZEEI21×TZEEI38	3.22	186.22	84.00	0.85	4.93	176.97	142.86	0.38	2.48	148.00	148.00	0.54	4.44	59.14	51.54	0.35	
TZEEI21×TZEEI76	4.12	145.24	135.43	2.53	5.04	230.49	229.41	3.31	2.48	87.17	50.30	1.19	4.73	79.85	61.43	1.85	
TZEEI29×TZEEI38	3.30	368.09	262.64	0.63	5.89	222.74	190.15	1.13	1.49	49.00	49.00	-0.35	3.80	49.02	43.40	0.16	
TZEEI29×TZEEI76	3.73	196.03	131.68	2.50	4.27	171.97	163.58	2.56	2.38	79.62	44.24	0.89	4.13	72.80	68.57	1.73	
TZEEI38×TZEEI76	3.30	212.80	104.97	2.10	5.10	187.32	151.23	2.99	2.67	101.51	61.82	1.17	4.25	70.68	60.38	1.45	
TZEEI-21×TZEEI-6	2.11	63.57	20.57	0.47	2.82	65.88	50.80	0.14	1.99	110.58	99.00	-0.15	3.78	26.00	23.13	0.14	
TZEEI29×TZEEI-6	1.45	66.67	59.34	0.91	2.45	40.40	31.02	0.90	1.95	106.35	95.00	-0.03	4.10	48.55	33.55	0.09	
TZEEI29×TZEEI21	2.87	115.79	64.00	-0.16	3.38	114.60	108.64	0.01	2.90	190.00	190.00	-0.18	4.01	49.07	36.86	0.18	
TZEEI38×TZEEI-6	2.45	268.42	195.18	0.28	4.37	124.10	115.27	-0.08	1.77	87.30	77.00	0.04	3.95	38.11	28.66	0.07	
TZEEI38×TZEEI21	3.75	231.86	114.29	-0.57	4.37	145.51	115.27	0.59	2.58	158.00	158.00	-0.05	4.14	48.39	41.30	0.15	
TZEEI38×TZEEI29	3.02	328.37	231.87	0.14	4.53	148.22	123.15	0.68	1.35	35.00	35.00	0.07	4.00	56.86	50.94	-0.1	
TZEEI76×TZEEI-6	2.62	114.75	62.73	0.36	3.19	88.20	70.59	0.23	2.17	70.87	31.52	0.14	3.95	46.30	28.66	-0.03	
TZEEI76×TZEEI21	4.82	186.90	175.43	-0.35	5.42	255.41	254.25	-0.20	2.17	63.77	31.52	0.15	4.56	73.38	55.63	0.09	
TZEEI76×TZEEI29	4.32	242.86	168.32	-0.29	4.64	195.54	186.42	-0.19	2.10	58.49	27.27	0.14	4.36	82.43	77.96	-0.11	
TZEEI76×TZEEI38	3.06	190.05	90.06	0.12	4.45	150.70	119.21	0.33	2.39	80.38	44.85	0.14	4.06	63.05	53.21	0.10	
Mean	3.17	197.25	132.38	0.56	4.17	144.58	127.89	0.72	2.16	97.18	79.22	0.29	4.15	55.20	44.81	0.39	

Low-N=Low-Nitrogen stress; Opt-N= optimal nitrogen; Dro-str=drought stress; Well-W=well-watered; MPH=mid-parent heterosis; BPH=better parent heterosis; SCA=specific combining ability

Appendix 12: Pearson correlation of Mean yield, heterosis and SCA effects of hybrid crosses evaluated under stress and non-stressed environments

Environment		Mean	MPH	BPH
Low—N stress environment	MPH	0.34		
	BPH	0.37	0.92***	
	SCA	0.06	-0.07	-0.04
Optimal-N environment	MPH	0.94***		
	BPH	0.87***	0.98***	
	SCA	0.33	0.36	0.34
Drought stress environment	MPH	0.70***		
	BPH	0.46*	0.95***	
	SCA	0.51*	0.05	-0.16
Well-watered environment	MPH	0.77***		
	BPH	0.64**	0.94***	
	SCA	0.38	0.47*	0.41
Across stress environment	MPH	0.64***		
	BPH	0.53***	0.911***	
	SCA	0.24	0.08	0.02
Across non-stress environment	MPH	0.63***		
	BPH	0.61***	0.99***	
	SCA	0.32*	0.38*	0.37*
Across all environments	MPH	0.19		
	BPH	0.27*	0.93***	
	SCA	0.25*	0.18	0.18

*, **, *** probability levels at 0.05, 0.01 and 0.001 respectively
 MPH=mid-parent heterosis; BPH=better parent heterosis



Appendix 13: Components of variance for GCA and SCA across N variation and moisture variation environments