Combining Ability and Genotype-by-Environment Interaction Analyses among Early-to-Medium Maturing Maize Hybrids under Drought and non-Drought Environments

by

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PREFACE

The research contained in this dissertation was completed by the candidate while based in the Discipline of Plant Breeding, School of Agricultural, Earth and Environmental Sciences, of the College of Agriculture, Engineering and Science, University of KwaZulu-Natal, Pietermaritzburg, South Africa. The research was financially supported by the Alliance for a Green Revolution in Africa (AGRA).

The contents of this work have not been submitted in any form to another university and, except where the work of others is acknowledged in the text, the results reported are due to investigations by the candidate.

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Signed: Professor Julia Sibiya

Date: 28/01/2021

ABSTRACT

Developing high yielding early to medium maturing maize hybrids for Southern Africa represents an effective way to contribute to improving crop productivity in the face of climate change and unpredictable weather patterns. The objectives of this study were: (i) to determine combining ability and gene action among germplasm lines for grain yield (GY) and other traits under drought and non-drought conditions using the line x tester mating design (ii) to explore genotype-by-environment interaction (GEI) patterns of the developed hybrids and identify broadly and specifically adapted entries, with the intention of developing early to medium maturing hybrids for South Africa and the sub region.

Twenty-three white maize inbred lines sourced from the International Maize and Wheat Improvement Center (CIMMYT) were crossed in a line x tester mating design involving 13 lines (females) and 10 testers (males), resulting in 122 successful single-cross (SC) hybrids. The SC hybrids and six commercial hybrid checks were evaluated in a 13x10 alpha lattice design, replicated twice under drought and non-drought conditions across three sites *viz*: Cedara Research Station, Ukulinga Research Farm and Makhathini Research Station over three seasons, (2018-2019 summer growing season, the 2019 offseason, and 2019-2020 summer growing season). Data for grain yield and its related traits was collected. Genetic analysis of the line x tester data followed a fixed effects model. The parents differed in general combining ability (GCA) effects for GY and other traits under drought and non-drought conditions. Likewise, the crosses varied in specific combining ability (SCA) effects for GY and other traits under the drought and non-drought regimes. Line CZL1380 and tester CML539 were good general combiners for GY under drought. Lines CML568, CKDHL0378, CKDHL0467, CML672, and CZL1380 and testers CML312 and CML547 had good GCA effect across non-drought regime. Two crosses, CML540 x CML547 and CKDHL0467 x CML312 had high SCA values for GY under drought and

non-drought regimes. The additive type of gene action was predominant for days to anthesis (AD), days to silking (SD), anthesis-silking interval (ASI) plant height (PH), ear position (EPO), ears per plant (EPP), ear aspect (EA), grain texture (GTX), grain moisture (GMH), kernel row number (KRN), and shelling percentage (SHL) under drought, and for AD, SD, ear height (EH), EPO, EPP, EA, GTX, GMH, ear length (EL), kernels per ear row (KER), ear weight (EW), and hundred kernel weight (HKW) across non-drought conditions. Non-additive gene action prevailed for EH, EL, ear diameter (ED), KER, EW, HKW, and GY under drought and for ASI, PH, ED, KRN, SHL, and GY across non-drought conditions. The identified hybrids could be targeted for release as cultivars, and the types of gene action are practically relevant for improvement of early to medium maturing maize germplasm for Southern Africa.

Grain yield data from the five environments was analysed to explore genotype by environment (GEI) among the developed hybrids and checks. Analysis of variance across all the environments showed huge environmental, genotypic and GEI effects, with the environment contributing the largest proportion of the variation followed by genotype and lastly GEI. The additive main and multiplicative interaction effects (AMMI) and the genotype and genotype-by-environment interaction (GGE) methods were employed on selected 62 entries to visualize the GEI patterns. The AMMI revealed that two interaction principal component axes (IPCA1 and IPCA2) were significant, and these contributed 50.32 % and 20.84%, respectively, to the total GEI variation. The AMMI1 revealed that hybrid MAK1-122 x CML545 was specifically adapted to drought conditions whereas hybrids CKDHL0467 x CML312 and CZL1380 x CML547 were broadly adapted. The identified two high yielding and broadly adapted experimental hybrids were superior to the best check WE3127 across all environments. Hybrids CML569 x CML566 and CKDHL0467 x CML547 were specifically adapted to irrigated conditions. The GGE-biplots had two principal components, PC1 and PC2, which together explained 69.87% of variation due

to genotype and GEI. The GGE-biplots showed similar GEI patterns as AMMI, with the same hybrids identified as broadly and specifically adapted. The identified hybrids could be assessed further in multi-environmental and multiple stress trials to confirm their suitability under high and low input production systems in South Africa and the sub-region.

DECLARATION: PLAGIARISM

I, **Mandisa Noxolo Dlamini,** declare that,

1. The research reported in this thesis, except where otherwise indicated is my original work.
2. This thesis has not been submitted for any degree or examination at any university.
3. This thesis does not contain other person' data, pictures, graphs or other information unless specifically acknowledged as being sourced from other persons.
4. This thesis does not contain other persons' writing unless specifically acknowledged as being sourced from other researchers.

a) Where other written sources have been quoted, then their words have been rewritten or rephrased, but the general information attributed them has been referenced.

b) Where exact words have been used, then their writing has been placed in italics and inside quotation marks and referenced.

5. This thesis does not contain text, graphic or tables copied from the internet unless specifically acknowledged and the source being detailed in the thesis and in the reference sections

Mandisa Noxolo Dlamini (Candidate)

As the candidate's supervisors, we agree the submission of this thesis:

Prof Julia Sibiya (Main Supervisor)

Dr Cousin Musvosvi (Co-Supervisor)

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DEDICATION

This work is dedicated to:

1. My God Almighty and ancestors, who constantly give me strength each day, nothing is impossible through them.
2. My mother Mrs. Philisiwe Dlamini and to the memory of my late father, Mr. Zwelibi Dlamini who unfortunately did not live to see my work.
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LIST OF ABBREVIATIONS

ABF:	abscisic acid-responsive elements binding factor
AD:	Days to anthesis
AEC:	Average Environment Coordinate
AGRA:	Alliance for Green Revolution in Africa
AMMI:	Additive Main and Multiplicative Interaction
ANOVA:	Analysis of Variance
ARC:	Agricultural Research Council
ASI:	Anthesis-Silking Interval
CBF:	C-repeat binding factor
CIMMYT:	International Maize and Wheat Improvement Center
CovHS:	Covariance of Half-Sib families
CovFS:	Covariance of Full-Sib families
Ced2018-19:	Cedara research station in the 2018- 2019 season
Ced2019-20:	Cedara research station in the 2019- 2020 season
DREB:	drying out responsive component binding
EA:	Ear Aspect
EL:	Ear Length
EH:	Ear Height
EPO:	Ear Position
EPP:	Ears Per Plant
EW:	Ear Weight
FW:	Field Weight
GCA:	General Combining Ability

GEI:	Genotype by Environment Interaction
$GEI_{n:}$	Genotype by Environment Interaction (noise)
$GEI_{s:}$	Genotype by Environment Interaction (signal)
GGE:	Genotype and Genotype by Environment interaction
GMH:	Grain Moisture at Harvest
GTX:	Grain Texture
GY:	Grain Yield
HKW:	Hundred Kernel Weight
IPCA:	Interaction Principal Component Axes
KER:	Kernels per Ear Row
KRN:	Kernel Row Number
KZN:	KwaZulu-Natal
Mak2019:	Makathini Research Station under drought in 2019 season
MSE:	Managed Stress Environment
OPV	Open-Pollinated Varieties
PC1:	Primary Principal Component
PC2:	Secondary Principal Component
PH:	Plant Height
SCA:	Specific Combining Ability
SD:	Days to Silking
SHL:	Shelling Percentage
Ukul2018-19:	Ukulinga research farm in the 2018-2019 season
Ukul2019-20:	Ukulinga research farm in the 2019-2020 season
WCA:	West and Central Africa
WEMA:	Water Efficient Maize for Africa

CHAPTER 1: GENERAL INTRODUCTION

1.1 Importance of maize

Maize (*Zea mays* L.) is the most cultivated staple crop in sub-Saharan Africa (Santpoort, 2020), having a prominent productive potential among other cereals (Gómez, 2018). It is the third most important grain crop after wheat and rice, having numerous uses as determined by the kernel endosperm qualities. The main endosperm types are flint, dent, flour, pop, pod, waxy, and sweet. Flint and dent maize types are the widely cultivated types and they have been substantially improved by introgression of both temperate and tropical exotic germplasm (Abe and Adelegan, 2019).

The demand for maize is continuously increasing because of its multiple uses (Meseka et al., 2018), but the production still cannot meet the demand (Maphumulo et al., 2015). Worldwide demand in 2020 was estimated to reach a 45% increase, which reflected a critical increase of 72% for maize in developing countries, and an 18% increase in industrial countries between 1997 and 2020. All things considered, an increase in maize demand requires an outstanding increase in maize production in both developing and industrial countries. Undeniably, in developing countries, especially in Africa, there is a limited access to improved technologies and weak infrastructure to utilize for production improvement, whereas industrial countries have capabilities to enhance production excessively (Murdia et al., 2016).

Improved technologies are the basis for national strategies that target increased production of maize (James, 2017). In several developing countries, there are notable challenges in acquiring the new technologies to increase production, hence they still experience constraints in accessing improved conventional technologies (OECD, 2001). There are numerous new

technologies adopted to increase maize productivity, such as the use of biotechnology, the use of improved hybrids, and changes in production management (Andorf et al., 2019).

1.2. Factors affecting crop yields

Crop yield is influenced by the direct impact of weather and environmental conditions on plant growth and development during growing period of a crop. The primary environmental conditions that influence plant development are photoperiod and temperature, but modern maize hybrids are less dependent on photoperiod and respond more to temperature (Moeletsi, 2017a). The duration for a particular growth stage to be completed by the plant is connected to temperature and especially the total daily temperature. Adams et al. (2001) mentioned that hot temperatures accelerate maturity whilst cool temperatures slow plant growth. This provides a route for indices based on air temperature to be used in regulating the phenological attributes of crops viz, growing degree days (GDD), photothermal units, photothermal index, and heat use (Amrawat et al., 2013). It is thus important to determine the duration of phenological stage and maturity of crops in various locations so that proper planting dates can be implemented for cultivars that match the growing period length of the specific locations to ensure optimum production. The crop will always require the same heat unit, but number of days to attain maturity or another development stage are not necessarily the same (Moeletsi, 2017b).

Maize production under dry environments has been hindered by drought stress indices which results in massive yield losses (Larson, 1993). According to Amrawat et al. (2013), yield loss in maize is due to the developmental stage at which water stress occurs, with the greatest yield decreases resulting from drought stress at or near anthesis. Hence, water deficit and high temperatures often coexist with significant growth stages. In areas where water is limited, breeding programs usually focus on drought-tolerant hybrids to mitigate the impact of drought stress. The drought stress of a specific crop is primarily regulated by the coexistence of its

reproductive growth stages with unfavourable environmental circumstances (Korres et al., 2016). Maize experiences drought stress because it is a summer annual with a relatively long growing season. This can result in high evaporative demand for water during the growth duration characterized by water deficit. The management tools which could be used to better water use efficiency would be very valuable for maize growers where there is potential for drought stress. Therefore, the adoption and development of early to medium maturing maize hybrids are of importance to escape late-season drought stress (Larson, 1993).

Maize hybrids are improved to be resilient to different environmental conditions, such as drought tolerance, meaning compared to OPVs they tend to yield better since they are genetically improved to develop in unfavourable environments. Maize yield is unfortunately impacted by the changes in climatic conditions; therefore, it is important to promote climate-resilient maize hybrids (Meseka et al., 2018), especially drought-tolerance and early to medium maturing hybrids. In maize improvement, it is of high importance to improve quality and grain yield potential. Hence, genetic improvement in attributes that are economically important with maintaining an adequate amount of variability is the appropriate objective in maize breeding programs (Gómez, 2018). Therefore, through the adoption of hybrid maize, the yield can be enhanced significantly (Meseka et al., 2018).

In Africa, particularly the eastern and southern African regions, high-yielding maize hybrids with intermediate to late maturity are grown. However, due to unreliable rainfall patterns experienced in these regions, there has been a switch towards hybrids that are early in maturity (Maphumulo et al., 2015). Pswarayi and Vivek (2008) described that these varieties provide an early harvest to overpass the “hunger season” (time of year between planting and harvest, when food runs out) before the period of a full-season crop, particularly in arid environments (Noëlle et al., 2018). Besides, they are less competitive for natural light, water, and nutrients than varieties

that are late maturing, therefore for this reason, they are able to escape late season drought and are perfect for intercropping (Noëlle et al., 2018).

1.3 Combining ability analysis

The concept of combining ability is particularly important regarding “testing” methodology, in which it is necessary to study and examine the performance of lines in hybrid combination (Griffing, 1956). The productivity or combining ability of parents is their ability to combine with one another throughout the process of hybridization, with the end goal that genes that are desired are inherited by the progenies (Biotech et al., 2005). Therefore, identifying lines that perform better as parents in future crosses is the fundamental objective for many breeding programs (Oakey et al., 2006). The selection of parental lines can be achieved through the use of specific mating designs, for example, the line by tester, North Carolina design I, II and III, and diallel, where the genetic impacts of the lines can be apportioned into additive and non-additive constituents (Fasahat, 2016; Oakey et al., 2006). Combining ability has been demonstrated as essential in plant breeding through several studies that have been previously conducted in numerous crops like cereals, and roots to legumes.

Sprague and Tatum, (1942) originally interpreted general combining ability (GCA) as a concept utilized to assign the mean performance of a line in hybrid combinations and specific combining ability (SCA) as dealing with specific combinations that relatively do better or worse than would be expected based on the mean performance of the lines used (Griffing, 1956). In plant breeding, vital decisions could be made based on the relative contribution of GCA and SCA effects. Early generation selection of genotypes turns out to be more effective, and promising hybrids can be chosen based on GCA effect predictions when the GCA variances prevail over SCA variances (Melchinger et al., 1988; Smith et al., 2008). Line x tester analysis is one of the most essential methodologies utilized to choose appropriate parents and crosses with good

GCA and SCA, respectively (Rashid et al., 2007). The analysis of line x tester furnishes information on the genotype and the gene action controlling the yield components.

1.4 Significance of genotype x environment interaction (GEI)

The genotype by environment interaction (GEI) is a phenomenon referring to the interplay of genetic and nongenetic effects resulting in varying genotype performances across environments. Hence, the relative performance of an attribute of two or more genotypes that were assessed in different environments would vary and consequently impact on the effectiveness of genotype selections in breeding programs (Adiloğlu et al., 2012). To reduce the effect of GEI, crops need to be subjected to numerous environments for testing to measure their specific and broad adaptation (Adiloğlu et al., 2012). Maize hybrids that are recently improved by the private seed companies are required to be tested in many areas for many years before being released for growing in each location (Tonk et al., 2011). Therefore, huge attention is required to ensure consistently performing genotypes under various environments are identified to develop genotypes with high yielding ability and optimum performance (Adiloğlu et al., 2012).

The adaptability and stability of maize hybrids can be identified from the significant information obtained from evaluating the genotypic performance in various locations (Haruna et al., 2017). A study conducted by Kang et al. (1991) revealed that selection dependent only on yield may not be always adequate when GEI is significant. Thus, in plant breeding, the fundamental activity is to identify genotypes that have high yield potential and yield stability (Araus et al., 2008). Moreover, the presence of GEI makes the process of identifying stable genotypes very difficult because it causes the relative ranking of genotype performance to change across environments that affect the breeding process. Consequently, genotypes having a wide range of adaptation are rarely identified because of the GEI effect (Masila and Langat, 2020).

The genotype by environment interactions in multi-environment trials affect the phenotypic and genotypic values observed, thus decreases gain from selection (Kumar et al., 2017). High-yielding and stable hybrids across various locations are highly desirable for commercial maize production. Scott (1967) explained that in maize, stability of yield is under genetic control and hence, appropriate to be considered for selection. Registered maize hybrids have been used in numerous attempts for analysing GEI in different environments (Badu-Apraku et al., 2011). For instance, GEI was investigated for grain yield on 132 early maturing hybrids of maize in 229 environments, and that was interpreted by Epinat-Le Signor et al. (2001) over 12 years. Consequently, it was revealed that the contributing factors of the genotypic and environment interaction for grain yield were early maturity of hybrids, water balance around flowering, and average temperature from the 12 leaf stage to the end of grain-filling phase in the given location (Tonk et al., 2011).

1.5 Importance of maize hybrid production

The use of maize hybrids has been reported to contribute to yield increase, for example a 25 to 50% increase was observed by Ahmad (2018). A study conducted by Abera et al (2016) revealed up to 250% high parent heterosis in maize hybrids. Additionally, hybrids can tolerate different stresses that are biotic and abiotic such as drought, salinity, diseases and pests, than open-pollinated genotypes (Ahmad, 2018). Recently, farmers have also been using the single cross hybrids as they are superior yielding hybrids than three-way and double cross hybrids. Hybridization of inbred lines between differing heterotic groups results in higher heterosis compared to hybridization within the same heterotic group.

1.6 Specific objectives of the study

Given the foregoing, the objectives of this study were:

i) to determine combining ability and gene action among germplasm lines for grain yield (GY) and other traits under drought and non-drought conditions using the line x tester mating design,

ii) to explore genotype-by-environment interaction (GEI) patterns of the developed crosses and identify broadly and specifically adapted entries, intending to develop early to medium maturing hybrids for South Africa and the sub-region.

1.7 Research hypotheses

The following research hypotheses were tested:

I. The parents (lines and testers) differ in general combining ability under drought and across non-drought environments.

II. The cross combinations between lines and testers differ in specific combining ability under drought and non-drought conditions.

III. The additive and non-additive type of gene action are equally important for all considered traits under drought and non-drought conditions.

IV. Genotype by environment (GEI) has a significant effect on grain yield performance of the crosses arising from the line x tester hybridization.

V. High potential crosses superior to standard check hybrids, that are specifically adapted, and those that are broadly adapted are available in the hybrid progeny population.

1.8 Dissertation outline

This thesis is organised into four chapters based on specific objectives with a journal paper design. Therefore, with such format some of the information on introduction and materials & methods is a repetition. The referencing style used is based on the Crop Science journal style. The dissertation has the following structure:

Chapter 1: General introduction

This section gives a setting by presenting brief background information of the study. Research aims, specific objectives and research hypotheses are presented in this section.

Chapter 2: Literature review

This chapter creates a frame of reference for the study. The chapter provides description and important evaluation of the major concepts including, genotype by environment interaction and stability analysis and combining analysis in relation to breeding for drought tolerance. The progress made in breeding for drought and genes controlling drought is reviewed. Selection methods and technologies for drought tolerance in maize are also outlined in this chapter.

Chapter 3: Combining Ability Analysis for Grain Yield and related traits of Early to Medium Maturing Maize Hybrids under Drought and Non-Drought Environments

This chapter focuses on combining ability of the inbred lines and crosses and understanding the type of gene action that controls the inheritance of major yield traits under drought and non-drought environments. The yield potential of the experimental hybrids is highlighted, and the major findings of the study are presented and discussed.

Chapter 4: Genotype by Environment Interaction Analysis of Grain Yield of Early to Medium Maturing Maize Hybrids Across Non-Drought and Drought Environments

The chapter focuses on determining the genotype by environment interaction among experimental hybrids evaluated and identifying varieties that are adapted across ideal and stressed environments. Suitable environments for testing, high yielding and stable varieties across drought and non-drought environments are identified.

Chapter 5: The general overview

This chapter outlines the general review of the research findings to the major objectives, implications of findings, and recommendations for future research.

References

Abe, A., and C.A. Adelegan. 2019. Genetic variability , heritability and genetic advance in shrunken-2 super-sweet corn (Zea mays L . saccharata) populations. 11: 100–105. doi: 10.5897/JPBCS2018.0799.

Abera, W., S. Hussein, J. Derera, M. Worku, and M. Laing. 2016. Heterosis and combining ability of elite maize inbred lines under northern corn leaf blight disease prone environments of the mid-altitude tropics. Euphytica 208(2): 391–400. doi: 10.1007/s10681-015-1619-5.

Adams, S.R., K.E. Cockshull, and C.R.J. Cave. 2001. Effect of temperature on the growth and development of tomato fruits. Ann. Bot. 88(5): 869–877. doi: 10.1006/anbo.2001.1524.

Adiloğlu, S., I, C. Yu, R. Chen, J.J. Li, et al. 2012. We are IntechOpen , the world ' s leading publisher of Open Access books Built by scientists , for scientists TOP 1 %. Intech i(tourism): 13. doi: 10.1016/j.colsurfa.2011.12.014.

Ahmad, M. 2018. Heritability and trait association studies in maize F1 hybrids. Int. J. Biosci. 12(1): 18–26. doi: 10.12692/ijb/12.1.18-26.

Amrawat, T., N.S. Solanki, S.K. Sharma, D.K. Jajoria, and M.L. Dotaniya. 2013. Phenology growth and yield of wheat in relation to agrometeorological indices under different sowing dates. African J. Agric. Res. 8(49): 6366–6374. doi: 10.5897/AJAR2013.8019.

Andorf, C., W.D. Beavis, M. Hufford, S. Smith, W.P. Suza, et al. 2019. Technological advances in maize breeding: past, present and future. Springer Berlin Heidelberg.

Araus, J.L., G.A. Slafer, C. Royo, and M.D. Serret. 2008. Breeding for yield potential and stress

adaptation in cereals. CRC. Crit. Rev. Plant Sci. 27(6): 377–412. doi: 10.1080/07352680802467736.

Badu-Apraku, B., M. Oyekunle, R.O. Akinwale, and A. Fontem Lum. 2011. Combining ability of early-maturing white maize inbreds under stress and nonstress environments. Agron. J. 103(2): 544–557. doi: 10.2134/agronj2010.0345.

Biotech, J.C.S., I. Bertan, F.I.F. De Carvalho, and A.C. De Oliveira. 2005. Parental Selection Strategies in Plant Breeding Programs. 10(4): 211–222.

Epinat-Le Signor, C., S. Dousse, J. Lorgeou, J.B. Denis, R. Bonhomme, et al. 2001. Interpretation of genotype X environment interactions for early maize hybrids over 12 years. Crop Sci. 41(3): 663–669. doi: 10.2135/cropsci2001.413663x.

Fasahat, P. 2016. Principles and Utilization of Combining Ability in Plant Breeding. Biometrics Biostat. Int. J. 4(1): 1–22. doi: 10.15406/bbij.2016.04.00085.

Gómez, R.L. 2018. Research Article Research Article. Arch. Anesthesiol. Crit. Care 4(4): 527–534. http://www.globalbuddhism.org/jgb/index.php/jgb/article/view/88/100.

Griffing, B. 1956. Concept of General and Specific Combining Ability in Relation to Diallel Crossing Systems. Aust. J. Biol. Sci. 9(4): 463. doi: 10.1071/bi9560463.

Haruna, A., G.B. Adu, S.S. Buah, A.L. Roger, A.I. Kudzo, et al. 2017. Analysis of genotype by environment interaction for grain yield of intermediate maturing drought tolerant top-cross maize hybrids under rain-fed conditions Analysis of genotype by environment interaction for grain yield of intermediate maturing drought to. Cogent Food Agric. 98(1). doi: 10.1080/23311932.2017.1333243.

James, C. 2017. Global Status of Commercialized Biotech/GM Crops in 2017: Biotech Crop Adoption Surges as Economic Benefits Accumulate in 22 years. Isaaa (Brief 53): 1–143. https://www.isaaa.org/resources/publications/briefs/53/download/isaaa-brief-53-2017.pdf%0Ahttp://library1.nida.ac.th/termpaper6/sd/2554/19755.pdf.

Kang, M.S., D.P. Gorman, and H.N. Pham. 1991. Application of a stability statistic to international maize yield trials. Theor. Appl. Genet. 81(2): 162–165. doi: 10.1007/BF00215718.

Korres, N.E., J.K. Norsworthy, P. Tehranchian, T.K. Gitsopoulos, D.A. Loka, et al. 2016. Cultivars to face climate change effects on crops and weeds: a review. Agron. Sustain. Dev. 36(1): 1–22. doi: 10.1007/s13593-016-0350-5.

Kumar, R.S., S.G. Kant, R. Kharel, K.C. Bahadur, M. Sapkota, et al. 2017. Study of yield and yield attributing traits of maize. Am. J. Food Sci. Heal. 3(6): 123–129.

Larson, E.M. 1993. Feeding value of wet distillers by product for finishing ruminants. *Journal of Animal Science,* 71(8), pp.2228-2236.

Masila B., and C. Langat. 2020. Multilocation Evaluation for yield and Yield Related Traits in Three- Way Multilocation Evaluation for yield and Yield Related Traits in Three- Way Cross Maize Hybrids in Kenya. (February). doi: 10.6084/m9.figshare.11994459.

Maphumulo, S.G., J. Derera, F. Qwabe, P. Fato, E. Gasura, et al. 2015. Heritability and genetic gain for grain yield and path coefficient analysis of some agronomic traits in early-maturing maize hybrids. Euphytica 206(1): 225–244. doi: 10.1007/s10681-015-1505-1.

Melchinger, A.E., W. Schmidt, and H.H. Geiger. 1988. Comparison of Testcrosses Produced from F 2 and First Backcross Populations in Maize . Crop Sci. 28(5): 743–749. doi: 10.2135/cropsci1988.0011183x002800050004x.

Meseka, S., A. Menkir, B. Bossey, and W. Mengesha. 2018. Performance assessment of drought tolerant maize hybrids under combined drought and heat stress. Agronomy 8(12). doi: 10.3390/agronomy8120274.

Moeletsi, M.E. 2017a. Mapping of Maize Growing Period over the Free State Province of South Africa: Heat Units Approach. Adv. Meteorol. 2017(January). doi: 10.1155/2017/7164068.

Moeletsi, M.E. 2017b. Mapping of Maize Growing Period over the Free State Province of South

Africa : Heat Units Approach. 2017.

Murdia, K., R. Wadhwani, N. Wadhawan, P. Bajpai, and S. Shekhawat. 2016. Maize Utilization in India: An Overview. Am. J. Food Nutr. 4(6): 169–176. doi: 10.12691/ajfn-4-6-5.

Noëlle, H., A. Mafouasson, V. Gracen, M.A. Yeboah, G.N. Id, et al. 2018. Genotype-by-Environment Interaction and Yield Stability of Maize Single Cross Hybrids Developed from Tropical Inbred Lines. : 1–17. doi: 10.3390/agronomy8050062.

Oakey, H., A. Verbyla, W. Pitchford, B. Cullis, and H. Kuchel. 2006. Joint modeling of additive and non-additive genetic line effects in single field trials. Theor. Appl. Genet. 113(5): 809–819. doi: 10.1007/s00122-006-0333-z.

OECD. 2001. Adoption of Technologies for Sustainable Farming Systems. Wageningen Work. Proc.: 149.

Pswarayi, A., and B.S. Vivek. 2008. Combining ability amongst CIMMYT's early maturing maize (Zea mays L.) germplasm under stress and non-stress conditions and identification of testers. Euphytica 162(3): 353–362. doi: 10.1007/s10681-007-9525-0.

Rashid, M., A.A. Cheema, and M. Ashraf. 2007. Line X tester analysis in basmati rice. Pakistan J. Bot. 39(6): 2035–2042.

Santpoort, R. 2020. THE drivers of maize area expansion in sub-Saharan Africa. How policies to boost maize production overlook the interests of smallholder farmers. Land 9(3). doi: 10.3390/land9030068.

Scott, G.E. 1967. Selecting for Stability of Yield in Maize 1 . Crop Sci. 7(6): 549–551. doi: 10.2135/cropsci1967.0011183x000700060001x.

Smith, J.S.C., T. Hussain, E.S. Jones, G. Graham, D. Podlich, et al. 2008. Use of doubled haploids in maize breeding: Implications for intellectual property protection and genetic diversity in hybrid crops. Mol. Breed. 22(1): 51–59. doi: 10.1007/s11032-007-9155-1.

Sprague, G.F., and L.A. Tatum. 1942. General vs. Specific Combining Ability in Single Crosses

of Corn. Am. Soc. Agron.

Tonk, F.A., E. Ilker, and M. Tosun. 2011. Evaluation of genotype x environment interactions in maize hybrids using GGE biplot analysis. Crop. Breed. Appl. Biotechnol. 11: 1–9.

CHAPTER 2: LITERATURE REVIEW

2.1. Introduction

This review focuses on topics that give a theoretical foundation for the study. The topics include a brief description of the biology of maize, production of maize in South Africa, importance of early to medium maturing varieties, impact of drought stress on maize production and useful germplasm resources to mitigate drought, selection for drought tolerance and a discussion on genotype x environment interaction and stability analysis. The importance of combining ability analysis in maize hybrid production is also reviewed.

2.2. Significance of maize

Maize production in South Africa is governed by practices that end up degrading soil, such as severe tillage, monoculture of maize, and fallow periods, consequently the organic matter and nutrients in the soil are depleted(Haarhoff et al., 2020). However, many sustainable practices have been proposed to resolve soil degradation issues, but the adoption of these practices has been slow. Moreover, despite severe soil losses due to these production practices, maize grain yields increased over the years (Haarhoff et al., 2020). As previously mentioned, maize is the most excessively produced crop in South Africa and globally, however there are huge differences in yield from location to location.

In previous years, in 2012 particularly, maize production was estimated to be 875 226 630 tons, with the United States, China, and Brazil contributing 31%, 24%, and 8%, respectively, to the production. In South Africa, approximately 16.7 teragram (Tg) of maize grain was produced from 2.6 million ha during the 2016-2017 production season (Nagy and Széles, 2018). The quantity of food supply, specifically maize and its products, ranges from 250-300 g/capita/day in South

Africa indicating its importance in the daily diet of South Africans (Schönfeldt et al., 2013). In addition, 40% of maize is used as livestock feed, accounting for approximately 4.5 Tg per year (Haarhoff et al., 2020).

Maize producers have been able to achieve profitable yields because of modern drought-tolerant and genetically modified maize hybrids, which expectedly eased the effect of soil degradation. Despite the increase in maize grain yield in recent decades, the sustainability of the improved grain yield is doubtful. This is due to erratic rainfall patterns and frequent drought periods that continue to make the rainfed production systems of maize vulnerable. There are three distinct rainfed regions in South Africa based on climate and soil type, viz, the Western region (35% total production), Eastern region (45% total production), and Kwa-Zulu-Natal region (10% total production). Climatic differences among the production locations are predominantly due to the influence of oceans surrounding South Africa (Ziervogel et al., 2014). South Africa is located between the cold Atlantic Ocean to the west, which induces a drier climate, and the warm Indian Ocean to the east, with the latter ocean creating a warm and humid climate in Kwa-Zulu Natal regions. The western region is classified as cold semiarid (BSk) having a mean annual rainfall ranging from 400 mm in the most western regions to 550 mm in the northern regions (Kotey et al., 2016**;** Ranum et al., 2014).

The Eastern and Kwa-Zulu Natal regions receive an annual rainfall ranging from 600-700 mm and 700-900, respectively, with humid subtropical (Cwa) and subtropical highland (Cwb) climatic zone in both regions (Ranum et al., 2014). In Western and Eastern regions, the variability of rainfall patterns between growing seasons affects maize yields, while in the Kwa-Zulu Natal region variability in temperature is more crucial (Khan et al., 2016). Gouse et al. (2005), reported that there was lower variability in maize grain produced from 2000-2001 to 2017-2018 in all three rainfed regions of South Africa because of improved crop breeding, where plants became more

drought and disease tolerant. Rainfed maize is produced on deep sandy oxisols of Aeolian origin with a clay content between 5 and 20% in the Western region. Soil types found in the Eastern and KwaZulu-Natal regions have textures of loamy sands, clay loams, and clay and are classified as Oxisols, Vertisols, Ultisols, and Mollisols (Ranum et al., 2014).

2.3. Impact of drought stress on maize production

Drought as a sole abiotic element severely affects crops negatively more than some other abiotic elements and it is becoming more crucial in various regions in the world (Khan et al., 2016). The dryness that mainly affects crops is defined based on the degree of dryness in comparison to average amounts of rainfall for an area and the duration of the dry period. The quantity of water utilized by maize to finish off growth and development during its life cycle is 350-450 mm of water. Each millimeter of moisture results in production of 10-16 kg grain, hence 250 liters of water are needed by a single maize plant at maturity (Tandzi and Mutengwa, 2020). Climate changes in the production environments impacts food production around the world significantly, with droughts having the greatest impact (Haarhoff et al., 2020; Ranum et al., 2014).

2.4. Breeding of maize under drought conditions

The fundamental impact of drought in maize crop is reflected by delayed silking, thus increasing the anthesis-silking interval (ASI), leading to extreme yield reductions (Sayadi Maazou et al., 2016). The distinctive phenotypic expression of drought stress in maize includes changes in color from green to green-grey, leaf rolling of lower leaves followed by the upper canopy leaves, resulting in stomatal closure that inhibits photosynthesis and consequently growth is slowed. The importance and the impact of drought stress in maize prompted maize breeders to develop maize germplasm that can tolerate drought stress. Traits that are receptive to inadequate moisture and mechanisms that can be used to adapt in drought should be known in order to develop suitable

drought-tolerant maize germplasm (Amalero et al., 2003). A variety of genes based on mechanisms such as drought-escape, drought avoidance, and drought tolerance are present in maize genotypes (Aslam et al., 2015).

The useful tool used to combat drought stress is the selection of genotypes that have improved yield and yield potential under dry conditions. Plants have different traits that enable them to adapt to drought conditions ranging from morphological, physiological and biochemical. Breeding for genetic resistance is the mechanism that is currently being used to realise yields during severe dry season (Aslam et al., 2015). During growth and development of crops, drought resistance is defined as the potential of a crop to survive and reproduce under limited water availability. However, in the context of agriculture, drought is defined as the potential of a crop to yield economically under limited water conditions (Fahad et al., 2017). Generally, any plant mechanism that contributes to minimal yield loss during severe dry season is included as a drought resistance mechanism (Byrne et al., 2018).

There are numerous adaptive drought mechanisms in maize production, i.e. (i) density of plants during inbred line development, (ii) the extent of drought and heat stress in nurseries with insufficient water, (iii) use of high yielding and stable germplasm for breeding programs, and (iv) adequate testing and evaluation of the progenies in several locations (Sayadi Maazou et al., 2016). The significant breeding objectives in maize production are to grow maize hybrids with substantial yield potential, and enhanced grain and related traits for users, however, growing hybrids that have improved resistance against different stresses is an extra demand to most programs. All the mentioned objectives being considered while developing new maize hybrids assists in overcoming water stress by decreasing the loss of yield. This significantly indicates that maize hybrids should have crucial levels of drought resistance (Aslam et al., 2015).

2.5. Exploiting drought tolerance germplasm

Agriculture today relies on movement of germplasm that furnishes the required genetic traits by plant breeder to enhance yield, quality, resistance to pest, diseases, and tolerance to various abiotic stresses (Ghimiray and Vernooy, 2017). Unfavorable environmental conditions include drought, extreme temperature, and low soil fertility. The germplasm is exploited to develop these improved crop varieties to cope under the changing demands and environments. In economically advanced countries, public funds support the collections, evaluations and maintenance of the genetic resources (Carlson, 1994). Various public bodies such as Consultative Group for International Agriculture Research (CGIAR) support regional, international and few national collections so that industrialized and developing countries can benefit. In economically advanced countries the public and private sectors produces the new varieties to meet farmers' requirements (Ghimiray and Vernooy, 2017).

The accessibility of developing maize hybrids that are early maturing significantly contributed to an increase in maize production in West and Central Africa (WCA), particularly in areas where shortage of water was experienced (Boakyewaa et al., 2012). Ndebeh et al. (2017) investigated and evaluated single cross hybrids demonstrating their superiority over double-crosses and open-pollinated varieties (OPV). According to Badu-Apraku et al. (2012), as a result of severe drought stress in least developed countries, about 15% yield losses of maize are experienced yearly. Several farmers chose to grow maize hybrids that are early maturing in WCA since they performed well during the off-season planting and gave an early harvest. This reduced the "hunger gap" before the harvest of late-maturing full season crops, particularly where there are two growing seasons per year (Pswarayi and Vivek, 2008). The maize hybrids that mature early, likewise, warrant various planting dates as a measure to adapt to the vulnerability of the precipitation pattern. They also give farmers flexibility in planting dates to avoid known terminal droughts during the cropping season. Early maturing hybrids are also acceptable for intercropping and off-season

planting in dry riverbed since they compete less for moisture, light, and nutrients than late-maturing varieties (Ngie et al., 2014).

Larson (1999) established that the use of hybrids that are well adapted and early maturing could enhance yield stability and identified an early maturing hybrid (Pioneer 3737) that produced yield comparable to those of late-maturing hybrids. Larson therefore conclude that all adapted early maturing hybrids could produce yields comparable to late maturing hybrids in environments where late-season droughts were predominant (Mabhaudhi, 2013). Annor et al. (2019) assessed three drought tolerant maize varieties on a farmer's field for two years. The farmers were able to select extra-early maturing varieties, setting great emphasis on the earliness of crop maturity than on yield.

2.6. Genotype x environment interaction and stability analysis

There is a growing requirement to identify maize hybrids that perform consistently and reliably for yield regardless of the environment. Eberhart and Russell (1966) expressed that a desirable cultivar ought to have an average yield performance that is higher under ideal conditions and less fluctuating under unfavourable conditions than that of a group of cultivars when tested in numerous environments (Raj, 2019). The genetic variability of maize, as a cross-pollinated crop requires a huge management for it genetic improvement due to the high genetic and phenotypic differences and allelic polymorphism (Mohammed, 2020). Therefore, evaluating trials in diverse locations is fundamental in assessing interaction of genotypes and the environments and precisely identifying superior genotypes.

Analysing GEI effects during maize hybrid evaluation for grain yield is significant because of the large differences that exist in soil and climatic conditions in growing locations. These differences can result in changes in yield ranking of genotypes over different environments making it

difficult to select for better genotypes (Miah et al., 2016). The GEI can be explained through various statistical models. Models that have been used include joint regression, multivariate clustering techniques, multiplicative formulations such as additive main effect, and multiplicative interaction (AMMI) (Gauch and Zobel, 1988). In plant breeding, phenotypic selection is used and determines the quality of yield predictions (Malosetti et al., 2013). In plant breeding, the phenotype is known to be a function of the genotype and environmental interaction. Therefore different components which include statistical, genetic, and physiological are used in the phenotypic prediction models (Liu et al., 2010). Even though various strategies are utilized for GEI and phenotypic stability analysis, the AMMI model is the most commonly used and appropriate for identifying ideal genotypes for stability. The model gives the relationship between genotypes, environment, and their interaction (Giridhar et al., 2016).

2.7. Selection methods and technologies for drought tolerance

Breeding programs that conduct trials in dry locations or in off-season are frequently increasing, hence in these trials, different water regime treatments should be strictly managed through frequent irrigation treatments. Nonetheless, a dry season period is significantly required to be longer and cover the whole growth cycle (Rauf et al., 2016). The use of proper experimental designs allows control between replicate variability and lowers spatial trends ensuring good field experiments, management, and interpretations of the phenotypic information (Rauf et al., 2016). This is useful because the heritability of secondary traits for maize drought tolerance varies according to the genetic makeup of each variety evaluated, the conditions where the varieties are evaluated and precision of phenotypic information. Moreover, the genetic variation observed in drought tolerance studies results from the interaction of a multitude of quantitatively inherited morphological traits whose effects on yield differ both in terms of magnitude and direction

depending on the prevailing drought scenarios in the managed stress environments (MSE) (Rauf et al., 2016).

Plants respond to the changing environment in a complex, integrated way leading them to react to a particular set of conditions and constraints during a specific period. Nevertheless, the genetic control for stress tolerance is furthermore complicated, yet highly affected by other environmental components and developmental stage of the crop (Nasser et al., 2020). In general, species grown in dry conditions are well adapted to drought stress, therefore such species, viz crop wild relatives, may be suggested for drought susceptible environments (Akram et al., 2010).

The use of hybrids that show superiority for yield over open-pollinated varieties under drought is highly important. The potential yield of hybrids is determined by the magnitude of heterosis that is sequentially regulated by the genetic combining ability of parental lines. Combining ability is generally referred to as the potential of lines involved in the breeding program to produce superior progeny (Rauf et al., 2016). Phenotypic selection is vital for developing inbred lines that are drought-tolerant but the primary effect on the performance of the hybrids is more crucial. Additionally, the proceeding testing and hybrid evaluation over inclusive range of environments leads to genetic gains under the optimum and drought stress conditions, however the gain is relatively lower under drought conditions (Hossain et al., 2016).

2.8. Genes controlling drought tolerance

Plants respond to environmental stresses through diverse physiological and biochemical changes. Exposure to environmental stresses such as drought, high salinity, and low temperature leads to cellular dehydration. Plants simultaneously respond and adapt to water stress at both the cellular and molecular levels, by accumulating osmolytes and proteins particularly involved in stress tolerance (Rauf et al., 2016). Some of the effects of drought on crop species could be

reduced by utilizing genetic variation for drought tolerance to create genotypes better adapted to cope with water stress (Frova et al., 1999).

To adapt to drought stress, plants have developed versatile mechanisms, including physiological and metabolic reprogramming, that guide gene expression. Diverse qualities are expressed and understood under water deficiency conditions (Langner et al., 2019). Many investigations conducted to understand the molecular mechanisms of drought stress have recognized species-specific, conserved drought response genes, and proteins involved in stabilizing membranes and expanding the cells' water restricting limit (Deng et al., 2009). Additionally, drought generates the biosynthesis of the phytohormone abscisic in stress tolerance, in turn causes stomata to close and prompt expression of stress-related genes (Rauf et al., 2016). Several factors that control and provide an adaptive response under drought stress were acknowledged, including myeloblastosis (MYB), drying out responsive component binding (DREB), C-repeat binding factor (CBF), abscisic acid-responsive elements binding factor (ABF), ABRE binding (AREB), (NAM, ATAF1/2, and CUC2 containing protein), WRKY, and SNF1-related kinase 2 (SnRK2). Regardless of these findings, nevertheless, the gene network of the drought stress response is yet not completely clarified (Deng et al., 2009**;** Zenda et al., 2019).

2.9. Analysis of combining ability and heterosis for drought tolerance in maize

Maize hybrids play a vital role in maize production improvement and food security (Rudolf-Pilih et al., 2019). Choosing acceptable parents (inbred lines) is extremely important for the development of hybrid maize varieties. Thus, the identification and critical selection of hybrids that are high yielding ought to be supported by the parent's combining ability and genetic structure (Fasahat, 2016). It is very important to test newly developed inbred lines with testers (inbred lines, single crosses, and open-pollinated varieties) in order predict their performance in their hybrids (Feher

et al., 2014). The combining ability is defined as the potential of an inbred line to transmit favourable traits to its hybrid progenies (Elmyhun et al., 2020a).

Analysis of the combining ability is important in assessing the general combining ability (GCA) of parents and specific combining ability (SCA) of crosses; thus facilitating the selection of parents and crosses to utilize in the breeding program (Nasser et al., 2020). Additionally, Griffing (1956) affirmed that GCA is the mean performance of parents in a series of hybrid combinations, whilst SCA is the performance of a specific hybrid combination, either better or worse than expected, based on GCA effect. Hence, GCA is associated with the additive gene effects or main effects in factorial mating designs and SCA as a non-additive gene effect or interaction effects (Elmyhun et al., 2020b). As a result, new inbred lines should be assessed for their performance in hybrid combinations for the traits the breeding program is focusing on, and in this case drought tolerance. This information is essential for hybrid and open-pollinated variety development (Yu et al., 2020).

Furthermore, Darwin (1876) coined the phenomenon of heterosis or hybrid vigor when he observed that the F_1 hybrid produced from inbred lines were phenotypically superior over both their parents. In plant breeding, heterosis could be the result of the interaction among multiple loci, depending on hybrids and traits, as proclaimed in the magnitude of heterosis for biomass, flowering related traits, yield, and resistance to abiotic and biotic constraints (Yu et al., 2020). The use of the phenomenon of heterosis is extremely vital for agricultural production and has been successfully useD in maize hybrid production (Fasahat, 2016). Additionally, breeding practices reveal that the performance of parents alone is not predictive of hybrid performance, hence superior hybrids are not only obtained because of the best parents (Nasser et al., 2020). Therefore, breeders should consider a parental line based on its potential to produce superior hybrids, not only based on the performance of the parents. Also, in breeding ideal hybrids that produce high grain yield, good quality, tolerant to abiotic and biotic stresses, combining ability is

analyzed along with heterosis for germplasm with limited numbers of parental lines (Yu et al., 2020; Elmyhun et al., 2020a).

2.10. Conclusion

The review has shown that crops are greatly affected by different environmental stresses, with drought becoming increasingly common in several regions of the world due to climate change. Maize as a drought-sensitive crop, is affected by water stress in most growth stages of development Therefore, in countries where maize is the major staple crop, particularly in Africa, solutions must be found to combat drought stress which results in maize yield reductions. However, maize producers can achieve profitable production because of modern drought-tolerant and genetically modified maize hybrids. The choice of genotypes with improved yield under drought conditions is thus useful. Currently, maize producers and breeders are aiming to cultivate maize hybrids with larger yield potential, stable yield and increased grain traits, however, it is also imperative that these hybrids be resistant against environmental challenges. The common effect that often arises from drought stress is physiological and causes extreme changes in cellular gene expression profile and several genes are induced by the exposure to dry conditions.

Screening for drought tolerance using conventional breeding methods is complicated by the low genetic variance of yield components under stress conditions, thus effective screening procedures are important. Some of the negative effects of drought on crops could be reduced by exploiting existing genetic variation in drought tolerance to develop genotypes better adapted to cope with water deficiency. For hybrids to be adapted to drought stress, plants have developed complex adaptive mechanisms, including physiological and metabolic. Maize hybrids have a significant role in maize production improvement and food security. Selecting appropriate parents is very critical for the development and success of maize hybrid

development. Parents' combining ability and gene action are useful when identifying and selecting high-yielding hybrids, thus analysing combining ability is an important genetic tool in the selection of ideal parents and crosses. The phenomenon of heterosis is very important for agricultural production and has exploited with success mostly in maize.

References

Akram, M., M.Y. Ashraf, R. Ahmad, E.A. Waraich, J. Iqbal, et al. 2010. Screening for salt tolerance in maize (Zea mays L.) hybrids at an early seedling stage. Pakistan J. Bot. 42(1): 141–154. doi: 10.5897/ajar11.1475.

Amalero, E.G., G.L. Ingua, G.B. Erta, and P.L. Emanceau. 2003. Review article Methods for studying root colonization by introduced. Agronomie 23: 407–418. doi: 10.1051/agro.

Annor, B., B. Badu-Apraku, D. Nyadanu, R. Akromah, and M.A.B. Fakorede. 2019. Testcross performance and combining ability of early maturing maize inbreds under multiple-stress environments. Sci. Rep. 9(1): 1–11. doi: 10.1038/s41598-019-50345-3.

Aslam, M., M. Maqpool, and R. Cengiz. 2015. Drought stress in maize (Zea Mays L.): effects, resistance mechanisms, global achievements, and biological strategies.

Badu-Apraku, B., M. Oyekunle, K. Obeng-Antwi, A.S. Osuman, S.G. Ado, et al. 2012. Performance of extra-early maize cultivars based on GGE biplot and AMMI analysis. J. Agric. Sci. 150(4): 473–483. doi: 10.1017/S0021859611000761.

Boakyewaa, G., A.C. Agricultural, and R. Institu. 2012. Genotype by Environment Interaction and Grain Yield Stability of Extra-early Maize (Zea mays L .) Hybrids Evaluated at three locations in Ghana. doi: 10.13140/2.1.3983.1364.

Byrne, P.F., G.M. Volk, C. Gardner, M.A. Gore, P.W. Simon, et al. 2018. Sustaining the future of plant breeding: The critical role of the USDA-ARS national plant germplasm system. Crop Sci. 58(2): 451–468. doi: 10.2135/cropsci2017.05.0303.

Carlson, T.B. 1994. Rejoinder reproduced with permission of the copyright owner . Further reproduction prohibited without permission . Prev. Sch. Fail. 51(3): 49–51. https://search.proquest.com/docview/220297257?accountid=12834.

Darwin, C. (1876). The effects of cross and self fertilisation in the vegetable kingdom.London. pgs 1-505.

Deng, X. feng, F. ling FU, N. NI, and W. chen LI. 2009. Differential Gene Expression in Response to Drought Stress in Maize Seedling. Agric. Sci. China 8(7): 767–776. doi: 10.1016/S1671-2927(08)60277-1.

Eberhart, S.A., and W.A. Russell. 1966. Stability Parameters for Comparing Varieties 1 . Crop Sci. 6(1): 36–40. doi: 10.2135/cropsci1966.0011183x000600010011x.

Elmyhun, M., C. Liyew, A. Shita, and M. Andualem. 2020a. Combining ability performance and heterotic grouping of maize (Zea mays) inbred lines in testcross formation in Western Amhara, North West Ethiopia. Cogent Food Agric. 6(1). doi: 10.1080/23311932.2020.1727625.

Elmyhun, M., C. Liyew, A. Shita, and M. Andualem. 2020b. Cogent Food & Agriculture Combining ability performance and heterotic grouping of maize (Zea mays) inbred lines in testcross formation in Western Amhara , North West Ethiopia Combining ability performance and heterotic grouping of maize (Zea mays) inb. Cogent Food Agric. 6(1). doi: 10.1080/23311932.2020.1727625.

Fahad, S., A.A. Bajwa, U. Nazir, S.A. Anjum, A. Farooq, et al. 2017. Crop production under drought and heat stress: Plant responses and management options. Front. Plant Sci. 8: 1–16. doi: 10.3389/fpls.2017.01147.

Fasahat, P. 2016. Principles and Utilization of Combining Ability in Plant Breeding. Biometrics Biostat. Int. J. 4(1): 1–22. doi: 10.15406/bbij.2016.04.00085.

Feher, K., J. Lisec, L. Römisch-Margl, J. Selbig, A. Gierl, et al. 2014. Deducing hybrid

performance from parental metabolic profiles of young primary roots of maize by using a multivariate diallel approach. PLoS One 9(1). doi: 10.1371/journal.pone.0085435.

Frova, C., M. Villa, M. Sari-Gorla, P. Krajewski, and N. Di Fonzo. 1999. Genetic analysis of drought tolerance in maize by molecular markers. I. Yield components. Theor. Appl. Genet. 99(1–2): 280–288. doi: 10.1007/s001220051233.

Gauch, H.G., and R.W. Zobel. 1988. Predictive and postdictive success of statistical analyses of yield trials. Theor. Appl. Genet. 76(1): 1–10. doi: 10.1007/BF00288824.

Ghimiray, M., and R. Vernooy. 2017. The importance and challenges of crop germplasm interdependence: the case of Bhutan. Food Secur. 9(2): 301–310. doi: 10.1007/s12571-017-0647-5.

Giridhar, K., S. Surya Kumari, C. Sarada, and L. Naram Naidu. 2016. Stability for seed yield in ajwain based on Gentoype Selection Index. Indian J. Agric. Res. 50(3): 244–248. doi: 10.18805/ijare.v50i3.10745.

Gouse, M., C.E. Pray, J. Kirsten, and D. Schimmelpfennig. 2005. A GM subsistence crop in Africa: The case of Bt white maize in South Africa. Int. J. Biotechnol. 7(1–3): 84–94. doi: 10.1504/IJBT.2005.006447.

Griffing, B. 1956. A Generalised Treatment of the use of Diallel Crosses in Quantitative Inheritance. (Department of Genetics, Cambridge): 31–50.

Haarhoff, S.J., T.N. Kotzé, and P.A. Swanepoel. 2020. A prospectus for sustainability of rainfed maize production systems in South Africa. Crop Sci. 60(1): 14–28. doi: 10.1002/csc2.20103.

Hossain, M.A., S.H. Wani, S. Bhattacharjee, D.J. Burritl, and L.S.P. Tran. 2016. Drought stress tolerance in plants, vol 1: Physiology and biochemistry.

Khan, I.A., G. Hassan, N. Malik, R. Khan, H. Khan, et al. 2016. Efeito dos herbicidas no rendimento e nos componentes de rendimento do milho híbrido (Zea mays). Planta

Daninha 34(4): 729–736. doi: 10.1590/S0100-83582016340400013.

Kotey, D.A., Y. Assefa, A. Obi, and J. van den Berg. 2016. Disseminating Genetically Modified (GM) maize technology to smallholder farmers in the Eastern Cape province of South Africa: Extension personnel's awareness of stewardship requirements and dissemination practices. South African J. Agric. Ext. 44(1): 59–74. doi: 10.17159/2413-3221/2016/v44n1a370.

Langner, J.A., A.J. Zanon, N.A. Streck, L.R.S. Reiniger, M.P. Kaufmann, et al. 2019. Maize: Key agricultural crop in food security and sovereignty in a future with water scarcity | Milho: Cultura agrícola chave na segurança e soberania alimentar num futuro com escassez de água. Rev. Bras. Eng. Agric. e Ambient. 23(9): 648–654.

Larson, E.J. 1999. Using corn maturity to maintain grain yield in the presence of late-season drought. J. Prod. Agric. 12(3): 400–405. doi: 10.2134/jpa1999.0400.

Liu, Y.J., C. Duan, M.L. Tian, E.L. Hu, and Y.B. Huang. 2010. Yield stability of maize hybrids evaluated in maize regional trials in Southwestern China using nonparametric methods. Agric. Sci. China 9(10): 1413–1422. doi: 10.1016/S1671-2927(09)60232-7.

Mabhaudhi, T. 2013. Water use and drought tolerance of selected traditional and indigenous crops.

Malosetti, M., J.M. Ribaut, and F.A. van Eeuwijk. 2013. The statistical analysis of multi-environment data: Modeling genotype-by-environment interaction and its genetic basis. Front. Physiol. 4 MAR(March): 1–17. doi: 10.3389/fphys.2013.00044.

Miah, M., S. Ahmed, and M. Uddin. 2016. Assessment of yield stability of maize inbred lines in multi-environment trials. Bangladesh J. Sci. Ind. Res. 51(1): 61–68. doi: 10.3329/bjsir.v51i1.27064.

Mohammed, A. 2020. Genotype by environment interaction and yield stability analysis of open pollinated maize varieties using AMMI model in Afar Regional State, Ethiopia. J. Plant

Breed. Crop Sci. 12(1): 8–15. doi: 10.5897/jpbcs2019.0839.

Nagy, J., and A. Széles. 2018. The scientific background of competitive maize production. Acta Agrar. Debreceniensis: 33–46. doi: 10.34101/actaagrar/150/1700.

Nasser, L.M., B. Badu-Apraku, V.E. Gracen, and H.N.A. Mafouasson. 2020. Combining ability of early-maturing yellow maize inbreds under combined drought and heat stress and well-watered environments. Agronomy 10(10 October). doi: 10.3390/agronomy10101585.

Ndebeh, J., R. Akromah, and K. Obeng-Antwi. 2017. Genotype by environment interaction and yield stability of early maturing maize single cross hybrids at three locations in Southern Ghana. African J. Agric. Res. 12(37): 2807–2816. doi: 10.5897/ajar2017.12570.

Ngie, A., F. Ahmed, and K. Abutaleb. 2014. Remote sensing potential for investigation of maize production: review of literature. South African J. Geomatics 3(2): 163. doi: 10.4314/sajg.v3i2.4.

Pswarayi, A., and B.S. Vivek. 2008. Combining ability amongst CIMMYT's early maturing maize (Zea mays L.) germplasm under stress and non-stress conditions and identification of testers. Euphytica 162(3): 353–362. doi: 10.1007/s10681-007-9525-0.

Raj, R.N. 2019. G × E interaction and Stability Analysis of Maize Hybrids Using Eberhart and Russell Model. Int. J. Agric. Environ. Biotechnol. 12(01). doi: 10.30954/0974-1712.03.2019.1.

Ranum, P., J.P. Peña-Rosas, and M.N. Garcia-Casal. 2014. Global maize production, utilization, and consumption. Ann. N. Y. Acad. Sci. 1312(1): 105–112. doi: 10.1111/nyas.12396.

Rauf, S., J.M. Al-Khayri, M. Zaharieva, P. Monneveux, and F. Khalil. 2016. Breeding strategies to enhance drought tolerance in crops. Adv. Plant Breed. Strateg. Agron. Abiotic Biot. Stress Trait. 2(December 2014): 397–445. doi: 10.1007/978-3-319-22518-0_11.

Rudolf-Pilih, K., M. Petkovšek, J. Jakše, N. Štajner, J. Murovec, et al. 2019. Proposal of a New

Hybrid Breeding Method Based on Genotyping, Inter-Pollination, Phenotyping and Paternity Testing of Selected Elite F1 Hybrids. Front. Plant Sci. 10(September): 1–12. doi: 10.3389/fpls.2019.01111.

Sayadi Maazou, A.-R., J. Tu, J. Qiu, and Z. Liu. 2016. Breeding for Drought Tolerance in Maize (<i>Zea mays</i> L.). Am. J. Plant Sci. 07(14): 1858–1870. doi: 10.4236/ajps.2016.714172.

Schönfeldt, H.C., N. Hall, and M. Bester. 2013. Relevance of food-based dietary guidelines to food and nutrition security: A South African perspective. Nutr. Bull. 38(2): 226–235. doi: 10.1111/nbu.12027.

Smith O.S. 1986. Covariance between line per se and testcross performance. Crop Sci 26:540–543

Tandzi, L.N., and C.S. Mutengwa. 2020. Estimation of Maize (Zea mays L.) Yield Per Harvest Area: Appropriate methods. Agronomy 10(1): 1–18. doi: 10.3390/agronomy10010029.

Yu, K., H. Wang, X. Liu, C. Xu, Z. Li, et al. 2020. Large-Scale Analysis of Combining Ability and Heterosis for Development of Hybrid Maize Breeding Strategies Using Diverse Germplasm Resources. 11(June): 1–16. doi: 10.3389/fpls.2020.00660.

Zenda, T., S. Liu, X. Wang, G. Liu, H. Jin, et al. 2019. Key maize drought-responsive genes and pathways revealed by comparative transcriptome and physiological analyses of contrasting inbred lines. Int. J. Mol. Sci. 20(6): 1–30. doi: 10.3390/ijms20061268.

Ziervogel, G., M. New, E. Archer van Garderen, G. Midgley, A. Taylor, et al. 2014. Climate change impacts and adaptation in South Africa. Wiley Interdiscip. Rev. Clim. Chang. 5(5): 605–620. doi: 10.1002/wcc.295.

CHAPTER 3: Combining Ability Analysis for Grain Yield and related traits of Early to Medium Maturing Maize Hybrids under Drought and Non-Drought Environments

Abstract

Knowledge of combining ability of parents and gene action, especially for economically important quantitative traits, influences both the choice of parents and breeding methodology to efficiently select superior cultivars. This study determined combining ability and gene action of maize inbred lines for grain yield (GY) and other traits under drought and non-drought conditions using the line x tester mating design with the intention of developing early to medium maturing hybrids for the region. The parents differed significantly in general combining ability (GCA) effects for GY and other traits under drought and non-drought conditions. Likewise, the crosses varied in specific combining ability (SCA) effects for GY and other traits under the drought and non-drought regimes. Line CZL1380 and tester CML539 were good general combiners for GY under drought. Lines CML568, CKDHL0378, CKDHL0467, CML672, and CZL1380 and testers CML312 and CML547 had good GCA effect across non-drought regime. Crosses CML540 x CML547 and CKDHL0467 x CML312 had high SCA values for GY under drought and non-drought regimes. Additive gene action was predominant for days to anthesis (AD), days to silking (SD), anthesis-silking interval (ASI) plant height (PH), ear position (EPO), ears per plant (EPP), ear aspect (EA), grain texture (GTX), grain moisture (GMH), kernel row number (KRN), and shelling percentage (SHL) under drought, and for AD, SD, ear height (EH), EPO, EPP, EA, GTX, GMH, ear length (EL), kernels per ear row (KER), ear weight (EW), and hundred kernel weight (HKW) across non-drought conditions. Non-additive gene action prevailed for EH, EL, ED, KER, EW, HKW, and GY under drought and for ASI, PH, ED, KRN, SHL, and GY across non-drought conditions. The identified hybrids could be targeted for release as cultivars, and the types of gene action are practically relevant for improvement of early to medium maturing maize germplasm for Southern Africa

3.1. Introduction

The main step in developing maize hybrids is to develop and test lines involved at early or late generation of breeding (Hallauer et al., 2015). The choice of best parents and appropriate breeding methodology is crucial for the success of a crop breeding program. Combining ability analysis gives valuable information that is used by breeders in choosing parents, and in addition it gives data concerning the predominant gene action governing quantitative traits that guides the breeder's choice of appropriate breeding methodology (Das et al., 2016).

Combining ability has two components, the general combining ability (GCA) and specific combing ability (SCA) (El-Hosary and Elgammaal, 2013). The GCA estimates the mean performance of lines in different cross combinations, whereas SCA is outlined as the deviation in performance of a specific cross from the expected based on the GCA of the parents (Fasahat, 2016). For better understanding of genetic architecture of quantitative traits, the estimation of GCA and SCA is highly essential and relevant in the establishment of coherent breeding programs (Fasahat, 2016).

Smith (1986) suggested that testing both the lines and their testcrosses is essential to determine superior lines *per se* and high hybrid performance. Hayes and Johnson (1939) indicated that the combining ability of inbred lines is a heritable trait. The selection of parents is vital in the development of hybrids. In this context, a line x tester design was used and is one of the most powerful tools that has been widely used for analysis of inbred lines in respect to predicting the combining ability (GCA) of parents and the selection of appropriate parents (Rashid et al., 2007; Istipliler et al., 2015). This design can evaluate a larger number of inbred lines than diallel designs. The ability of any inbred line to effectively combine with different lines is a valuable character in hybrid breeding to produce hybrids that are superior. Therefore,

combining ability analysis is a useful biometric tool for crop breeders to predict the potential of their breeding lines in hybrid prediction (Amin et al., 2015).

The line x tester analysis gives information about combining ability effect of genotypes and the gene action controlling the traits under investigation, especially yield (Fasahat, 2016). Understanding genetic and specific combining abilities for yield and its components has become increasingly valuable to plant breeders to select suitable parents for developing hybrid cultivars (Istipliler et al., 2015). From statistical perspective, the GCA is the main effect and SCA is an interaction effect. Supported by Sprague and Tatum (1942) the main-effect (GCA) is attributed to the action of genes that are mostly additive in their effects furthermore as additive x additive interaction (Griffing, 1956). Specific combining ability reveals loci with dominance variance (non-additive effect) and all the three types of epistatic interaction elements, if epistasis is present. They incorporate additive x dominance and dominance x dominance interaction (Fasahat, 2016).

Additionally, the magnitude of gene action for a specific trait is affected by the environment in which the crop is grown. Subsequently, much effort has been given by maize breeders to estimate the interaction between genetic components and environmental conditions (Ahmed et al., 2000**;** Mosa and Motawei, 2005)**.** This shows the importance of evaluating the hybrids in multi-environmental trials to determine the importance of genotype x environment interaction. The objective of this study was to determine combining ability and gene action for grain yield (GY) and other traits among maize inbred lines under drought and non-drought conditions using the line x tester mating design with the intention of developing early to medium maturing hybrids for the region.

3.2. Materials and Methods

3.2.1. Development of single crosses hybrids

Twenty-three parental inbred lines of the early (varieties that need 89 to 90 days to mature) and medium (varieties that need 95 to 100 days to mature) maturity groups (Table 3.1) were used in a hybridization scheme to develop experimental hybrids. Among these germplasm lines, 20 were sourced from the International Maize and Wheat Improvement Center – Harare, Zimbabwe (CIMMYT-Harare); two were obtained from the University of KwaZulu-Natal's maize breeding programme at Ukulinga Research Farm, and only one was acquired from the Agricultural Research Council of South Africa (ARC). A line by tester mating design was followed during hybridization, wherein 13 inbred lines were used as females (lines) and ten inbred lines were used as males (testers). Nine of the lines used as testers had already been used as such by CIMMYT maize breeders, and one tester was a South African inbred line. All the inbred lines were fully inbred; thus, the inbreeding coefficient *F* was 1. Out of the 130 crosses expected from the 13 x 10 line by tester mating, only 122 crosses were successful.

Table 3.1. Parental inbred lines used to develop crosses

	Lines			Testers	
	Parents	Origin		Parents	Origin
1	A1220-4CYL	Ukulinga Research Farm	1	CML442	CIMMYT – Zimbabwe
2	CML550	CIMMYT – Zimbabwe	2	CML312	CIMMYT – Zimbabwe
3	CML568	CIMMYT – Zimbabwe	3	CML537	CIMMYT – Zimbabwe
4	CML571	CIMMYT – Zimbabwe	4	CML539	CIMMYT – Zimbabwe
5	CKDHL0378	CIMMYT – Zimbabwe	5	CML545	CIMMYT – Zimbabwe
6	CML569	CIMMYT – Zimbabwe	6	CML444	CIMMYT – Zimbabwe
7	CZL0919	CIMMYT – Zimbabwe	7	CML566	CIMMYT – Zimbabwe
8	CML440	CIMMYT – Zimbabwe	8	CML547	CIMMYT – Zimbabwe
9	CKDHL0467	CIMMYT – Zimbabwe	9	CML395	CIMMYT - Zimbabwe
10	CML540	CIMMYT – Zimbabwe	10	K64R	ARC – South Africa
11	CML572	CIMMYT – Zimbabwe			
12	CZL1380	CIMMYT - Zimbabwe			
13	MAK1-122	Ukulinga Research Farm			

3.2.2. Field evaluation trials

The 122 crosses accompanied by eight commercial checks (130 hybrids) were evaluated in 5 diverse environmental trials in the KwaZulu-Natal maize production region (region4). The trials were: (i) Trial I – Rain-fed trial planted on 13 December 2018 at Cedara Research Station (latitude 29º 32'S, longitude 30º 16'E, altitude 1400m, with average rainfall of 900mm) (ii) Trial II - Rain-fed trial planted on 20 December 2019 at Cedara Research Station (latitude 29º 32'S, longitude 30º 16'E, altitude 1400m, with average rainfall of 900mm) (iii) Trial III – Irrigated trial planted on 6 December 2018 at Ukulinga Research Farm (latitude 29º 40'S, longitude 30º 24'E, altitude 800m, with average rainfall of 750mm) (iv) Trial IV – Irrigated trial planted on 18 December 2019 at Ukulinga Research Farm (latitude 29º 40'S, longitude 30º 24'E, altitude

800m, with average rainfall of 750mm) (v) Trial V – Managed drought trial planted in the offseason on 12 April 2019 at Makhathini Research Station (latitude 27º 23'S, longitude 32º 10'E, altitude 450 m, with average rainfall of 635 mm). The environments could be grouped into non-drought (four environments) and drought (one environment).

3.2.3. Design and field cultural practices

After preparing the field to a fine tilth by disc ploughing and harrowing, experimental units (plots) were marked for planting. The experimental unit consisted of one row which was 5 m in length. Distance from row to row was 0.8 m and 0.3 m from one planting station to the other within a row. The evaluation design used in each environment was a 13x10 alpha lattice in two replicates. Fertiliser application was done at the following rates: 70 kg N ha^{-1}, 40 kg P_2O_5 ha^{-1}, and 30 kg K_2O ha^{-1}. All the P_2O_5 and K_2O were incorporated into the soil at planting. However, a fifth of the required amount of N was applied at planting, and the remainder was applied in three splits 28 days after planting (DAP), 42 DAP and 55 DAP. A pre-emergence herbicide, Dual® was used to control broad leaved weeds. Gramoxone® was applied post-emergence between the rows of the maize crop, and hand weeding was done to get rid of weeds within the rows. Insecticide Karate® was applied pre-emergence to control cutworms. The insects including the maize stalk borer control were controlled by application of Coragen®.

3.2.4. Trait measurement and observation

The following is a description of the traits and how they were assessed and recorded.

Days to anthesis (AD) were the number of days from planting to that time when 50% of plants in a plot had fully emerged tassels that were shedding pollen.

Days to silking (SD) was the count of days from planting to that time when 50% of plants in a plot had emerged silks that were at least 2 cm long.

Anthesis-Silking interval was determined by subtracting AD from SD.

Plant height (PH) refers to the tallness of plants, the distance from soil level to the tip of the plant measured from the hard dough stage till physiological maturity; average of five plants was recorded in centimeters (cm).

Ear height (EH) refers to the distance from soil level to the insertion of the top ear in centimetres; and average of five plants was recorded.

Ear position (EPO) was obtained by dividing EH by PH.

Ear aspect (EA) was the visual impression of ears scored on a 1 to 5 scale with score 1 given to large, uniform, well-filled and clean ears, and score 5 given to ears with most undesirable attributes.

Grain texture (GTX) was scored visually on a 1 to 5, where 1 = dent and 5 = dent; 2, 3, and 4 are intermediate classes.

Grain percent moisture content at harvesting (GMH) was measured soon after harvesting using a grain moisture meter, Dole® E.T.N model 500.

Field weight (FW) was the weight of all the harvested ears in a plot.

Ears per plant (EPP) was determined by dividing the number of harvested ears with at least one kernel in a plot by the number of plants in the same plot at harvesting.

Ear length (EL) introduced the average length of an ear, where average length of ten ears were recorded in centimetres (cm).

Ear diameter (ED) was the average diameter of ten ears was recorded in centimetres (cm), measured at the widest part of the ear.

Number of kernel rows per ear (KRN) is the average number of kernel rows on an ear; the average was recorded from six ears.

Number of kernels per ear row (KER) is the average of kernels in a row on the ear; the average was recorded from six ears.

Ear weight (EW) refers to the weight of dry ears before shelling; an average was recorded from ten ears.

Shelling % (SHL) was obtained by dividing the weight of kernels from ten ears by weight of ten ears and multiplying the quotient by 100.

Hundred kernel weight (HKW) was determined by weighing a random sample of 100 kernels, a mean of three independent samples per plot was recorded in grams (g).

Grain yield (GY) refers to weight of shelled kernels per hectare (t ha^{-1}), at 12.5% moisture level. The components FW, GMH, and SHL were used to estimate GY in tons per hectare (t ha^{-1}) and values were adjusted to 12.5% moisture content (Equation 3.1).

$$Grain\ \ yield\,(t/ha) = \frac{FW(kg) * 10000\,(m^2) * (100 - GMH) * SHL}{1000\,(kg) * plot\ area\,(m^2) * (100 - 12.5) * 100} \qquad \text{Equation 3.1}$$

3.2.5. Statistical and genetic analyses

All analyses were done using SAS version 9.4 (SAS Institute Inc, 2018) and AGD-R (Rodríguez et al. 2015) version 5 (2018) statistical software. The check hybrids were excluded from genetic analysis; thus, the alpha lattice design was no longer valid, and the analysis followed a simple randomized block design. In addition, all crosses involving lines A1220-4CYL and CML569 were excluded to make the dataset balanced, since some of their crosses were not successful. The combining ability effects at a single environment were estimated following Kempthorne (1957) model, as presented in Equation 3.2 and Table 3.2.

$$X_{ijk} = \mu + r_k + g_i + g_j + s_{ij} + e_{ijk}$$ Equation 3.2

Where,

X_{ijk} = value of the *ijk*th observation, μ = population mean effect, g_i = general combining ability effect of the *i*th line, g_j = general combining ability effect of the *j*th tester, s_{ij} = specific combining ability effect of the cross combination involving the *i*th line and *j*th tester, e_{ijk} = random error effect associated with th *ijk*th observation, *I* = number of lines, *j* = number of testers, *k* = number of replications, and r_k = *k*th replication effect.

3.2.6. Combining Ability Analysis of Variance

Table 3.2. Skeleton analysis of variance (ANOVA) of Line x Tester design for Combining ability effects at one environment

Source	Degrees of Freedom	Mean Squares	Expected Mean Square Components of Variance	Covariance of Relatives
Replications	$r-1$			
Crosses	$lt-1$			
Lines	$l-1$	M_l	$\sigma_e^2 + r\sigma_{lt}^2 + rt\sigma_l^2$	$\sigma_e^2 + r\left(Cov\,FS - Cov\,HS_t - Cov\,HS_l\right) + rt\,Cov\,HS_l$
Testers	$t-1$	M_t	$\sigma_e^2 + r\sigma_{lt}^2 + rl\sigma_t^2$	$\sigma_e^2 + r\left(Cov\,FS - Cov\,HS_t - Cov\,HS_l + rl\,Cov\,HS_t\right)$
Lines x Testers	$(l-1)(t-1)$	M_{lt}	$\sigma_e^2 + r\sigma_{lt}^2$	$\sigma_e^2 + r\left(Cov\,FS - Cov\,HS_t - CovHS_l\right)$
Error	$(r-1)(lt-1$	M_{error}	σ_e^2	σ_e^2
Total	$rlt-1$			

3.2.7. Estimation of genetic variance components

At a single environment, the covariance of relatives and combining ability variances were estimated as given by Singh and Chaudhary (1985), as shown in equations 3.3 to 3.7. Since all the parents were fully inbred, *F* = 1, the additive genetic variance, Var (Additive) was equated to twice the average covariance of half-sibs, and the dominance variance, Var (Dominance) was equated to the specific combining ability variance.

$$Cov\ HS\ lines = \frac{M_l - M_{lxt}}{rt} \qquad \text{Equation 3.3}$$

$$Cov\ HS\ testers = \frac{M_t - M_{lxt}}{rl} \qquad \text{Equation 3.4}$$

$$Cov\ HS\ average = \frac{M_l + M_t - 2M_{lxt}}{r(1+t)} = Var(GCA) \qquad \text{Equation 3.5}$$

$$Cov\ FS = \frac{M_l + M_t + M_{lxt} - 3M_{error} + 6r\ Cov\ HS - r(1+t)Cov\ HS}{3r} \qquad \text{Equation 3.6}$$

$$Var(SCA) = Cov\ FS - 2Cov\ HS \qquad \text{Equation 3.7}$$

The estimates of general combining ability, *gca* effects of parents ($\hat{g}_i$) and specific combining ability, *sca* effects of crosses ($\hat{s}_{ij}$) were obtained as in equations 3.8 to 3.11 Singh and Chaudhary (1985).

$$\mu = \frac{X_{...}}{rlt} \qquad \text{Equation 3.8}$$

$$\hat{g}_i = \frac{X_{i..}}{rt} - \frac{X_{...}}{rlt}$$ Equation 3.9

$$\hat{g}_j = \frac{X_{.j.}}{rl} - \frac{X_{...}}{rlt}$$ Equation 3.10

$$\hat{s}_{ij} = \frac{X_{ij.}}{r} - \frac{X_{i..}}{rt} - \frac{X_{.j.}}{rl} - \frac{X_{...}}{rlt}$$ Equation 3.11

Where,

$X_{...}$ = total of all hybrid combinations, $X_{i..}$ = total of the *i*th line over *t* testers and *r* replications, $X_{.j.}$ = total of the *j*th tester over *l* lines and *r* replications, $X_{ij.}$ = total of the hybrids of *i*th line and *j*th tester over *r* replications.

3.2.8. Estimation of combining ability effects

The significance of combining ability effects was tested by performing a *t* – test, and the t value was obtained as in Equation 3.12, and the standard errors as in Equations 3.13 to 3.15. The critical difference values in each case were calculated by multiplying their corresponding SE values with Table '*t*' value at error degrees of freedom at 5% level of significance.

t statistic, $t = \frac{Effect}{SE}$ Equation 3.12

Standard error for *gca* effect of lines

$$\text{SE (gi)} = \sqrt{\left(\frac{\sigma_e^{\,2}}{\text{rt}}\right)}$$ Equation 3.13

Standard error for *gca* effect of testers

$$\mathrm{SE\ (gj)} = \sqrt{\left(\frac{\sigma_e^{\ 2}}{\mathrm{rl}}\right)} \qquad \text{Equation 3.14}$$

Standard error for *sca* effect of hybrids

$$\mathrm{SE\ (sij)} = \sqrt{\left(\frac{\sigma_e^{\ 2}}{\mathrm{r}}\right)} \qquad \text{Equation 3.15}$$

3.2.9. Contribution of parents and their interaction to hybrid progeny variation

The contribution of parents and their interaction to hybrid progeny variance was determined using Equations 3.16 to 3.18.

$$Contribution\ of\ lines = \frac{SS_l * 100}{SS_c} \qquad \text{Equation 3.16}$$

$$Contribution\ of\ testers = \frac{SS_t * 100}{SS_c} \qquad \text{Equation 3.17}$$

$$Contribution\ of\ lines\ x\ testers = \frac{SS_{lxt} * 100}{SS_c} \qquad \text{Equation 3.18}$$

Where

SS_l = line sum of squares

SS_t = tester sum of squares

SS_e =error sum of squares

SS_{lxt} = line x tester sum of squares

Across environments, the combining ability effects were calculated using the model presented in Equation 3.19. Combining ability variances, genetic variances and related formulae used are presented in Equations 3.20 to 3.28. The combining ability effects were calculated just as for the single environment except that the total of all replications over the environments was used for the lines x tester table and the denominator was *rltn* instead of *rlt.* The standard errors of the effects were also calculated as for the single environment except for the denominator which changed after multiplying by number of environments. The same formulae as for a single environment were used for the contribution of lines, testers, and their interaction, to hybrid progeny variance.

$$X_{ijkr} = \mu + l_i + t_j lt_{ij} + n_k + \ln_{ik} + tn_{jk} + ltn_{ijk} + e_{ijkr} \qquad \text{Equation 3.19}$$

Where, X_{ijkr} = the volume of the *ijkr*th observation, μ = the overall mean

l_i = the gca effect of the *i*th line parent, t_j = the gca effect of the *j*th tester parent

lt_{ij} = the sca effect of the hybrids, n_k = the effect of the kth environment, $\ln_{ik}$ = *i*th line and *k*th environment interaction effect, tn_{jk} = *j*th tester and *k*th environment interaction effect, ltn_{ijk} = hybrids x environment interaction effect, e_{ijkr} = the random error associated with *ijkr*th observation, l = number of line, t = number of testers

n = numbers of environments, r = number of replications

$$Cov\,HS\,lines = \frac{M_l - M_{lxt} - M_{lxe} + M_{lxtxe}}{rtn} \qquad \text{Equation 3.20}$$

$$Cov\,HS\,testers = \frac{M_t - M_{lxt} - M_{txe} + M_{lxtxe}}{rtn}$$ Equation 3.21

$$Cov\,HS\,average = \frac{M_l + M_t - 2M_{lxt} - M_{lxe} - M_{txe} + 2M_{lxtxe}}{rn(1+t)} = Var(GCA)$$ Equation 3.22

$$Cov\,FS = \frac{M_{lxt} - M_{lxtxe}}{rn} + \frac{2(M_l + M_t - 2M_{lxt} - M_{lxe} - M_{txe} + 2M_{lxtxe})}{rn(1+t)}$$ Equation 3.23

$$Var(SCA) = \frac{M_{lxt} - M_{lxtxe}}{rn}$$ Equation 3.24

$$Var(GCA)\,x\,n = \frac{M_{lxe} + M_{txe} - 2M_{lxtxe}}{rn(1+t)}$$ Equation 3.25

$$Var(SCA)\,x\,n = \frac{M_{lxtxe} - M_{error}}{r}$$ Equation 3.26

$$\text{GCA to SCA variance ratio} = \frac{Var(GCA)}{Var(SCA)}$$ Equation 3.27

$$Ba\,\text{ker's}\,ratio = \frac{2Var(GCA)}{2Var(GCA) + Var(SCA)}$$ Equation 3.28 (Baker, 1978)

3.3. Results

3.3.1. Combining ability mean squares

The analysis of variance for combining ability disclosed significant mean squares due to crosses, lines, testers, and line x tester for all considered traits (Table 3.3. and Table 3.4.). Significant mean squares were also observed for crosses, lines, testers, and line x tester

interaction, for SD, PH, EH, ASI, EPO, EPP, EA, GTX, GMH, EL, ED, KRN, KER, and GY (Table 3.3.) under intermediate drought stress. Across non-drought conditions, highly significant differences were observed between crosses, lines, testers, and line x tester interaction for all traits except EPO and GMH for which the line x tester interaction was not significant (Table 3.4.).

Table 3.3. Combining ability and genetic component variances of line*tester developed single-cross maize hybrids grown under intermediate drought stress

Source	DF	AD	SD	ASI	PH	EH	EPO	EPP	EA	GTX
REP	1	41.02	1.82	0.07	1898.85	866.72	0.00	0.01	0.11	0.29
CROSSES	109	23.02*	28.78***	0.02**	582.30***	289.25***	0.00***	0.15***	0.72***	0.91***
LINES	10	126.61	162.36**	0.04**	2303.02**	1112.76**	0.02***	0.80***	2.07***	3.60***
TESTERS	9	69.22*	65.40**	0.06	819.57**	573.15***	0.01***	0.22***	1.20*	3.98***
LINES*TESTE	90	6.89*	10.27	0.01	367.39	169.36**	0.00***	0.07**	0.52	0.31
Error	109	4.33	8.95	0.01	317.01	104.49	0.00	0.04	0.53	0.25
R^2		84.37	76.30	57.43	65.42	73.99	82.17	77.24	57.83	78.87
CV (%)		2.72	3.89	10.42	7.58	10.28	6.06	16.72	22.51	15.43
COVHSlines		5.99	7.60	0.00	96.78	47.17	0.00	0.04	0.08	0.16
COVHStesters		2.83	2.51	0.00	20.55	18.35	0.00	0.01	0.03	0.17
COVHS		4.33	4.93	0.00	56.85	32.08	0.00	0.02	0.05	0.17
COVFS		9.95	10.53	0.00	138.89	96.58	0.00	0.05	0.10	0.36
Var(GCA)		4.33	4.93	0.00	56.85	32.08	0.00	0.02	0.05	0.17
Var(SCA)		1.28	0.66	0.00	25.19	32.43	0.00	0.01	0.00	0.03
Var(GCA)/Var(		3.39	7.44	-1.75	2.26	0.99	1.24	1.57	-31.62	5.42
Baker's ratio		0.87	0.94	1.40	0.82	0.66	0.71	0.76	1.02	0.92
Var(Additive)		8.67	9.87	0.00	113.71	64.15	0.00	0.04	0.11	0.33
Var(Dominance		1.28	0.66	0.00	25.19	32.43	0.00	0.01	0.00	0.03

Cont.

Source	DF	GMH	EL	ED	KRN	KER	EW	SHL	HKW	GY
REP	1	0.89	160.85	0.04	0.06	155.20	36866.97	26.21	105.02	5.65
CROSSES	109	1.18**	3.77***	0.27*	1.35***	18.70***	10497.28	47.36*	29.40*	2.81**
LINES	10	1.79*	7.41***	0.53**	2.73***	17.22**	10865.60	33.51	43.52**	5.41***
TESTERS	9	3.50**	13.52***	0.25	6.66***	92.91***	7356.71**	153.76**	63.12***	3.11*
LINES*TESTE	90	0.88	2.39*	0.24	0.66	11.45**	10770.41	38.26	24.46**	2.49**
Error	109	0.74	1.56	0.18	0.49	6.91	9933.94	33.78	14.34	1.56
R^2		61.61	77.02	60.12	73.39	74.46	52.17	58.49	67.92	64.72
CV (%)		5.43	8.52	9.28	4.94	8.30	56.78	7.04	10.86	21.88
COVHSlines		0.05	0.25	0.01	0.10	0.29	4.76	-0.24	0.95	0.15
COVHStesters		0.12	0.51	0.00	0.27	3.70	-155.17	5.25	1.76	0.03
COVHS		0.08	0.38	0.01	0.19	2.08	-79.01	2.64	1.37	0.08
COVFS		0.24	1.18	0.05	0.47	6.42	260.21	7.52	7.81	0.63
Var(GCA)		0.08	0.38	0.01	0.19	2.08	-79.01	2.64	1.37	0.08
Var(SCA)		0.07	0.41	0.03	0.09	2.27	418.24	2.24	5.06	0.47
Var(GCA)/Var(		1.22	0.93	0.23	2.21	0.91	-0.19	1.18	0.27	0.18
Baker's ratio		0.71	0.65	0.32	0.82	0.65	-0.61	0.70	0.35	0.26
Var(Additive)		0.17	0.77	0.01	0.38	4.15	-158.02	5.27	2.75	0.17
Var(Dominance		0.07	0.41	0.03	0.09	2.27	418.24	2.24	5.06	0.47

Note: AD = Days to anthesis; SD = Days to silking; ASI = Anthesis-Silking interval; PH = Plant height; EH = Ear height; EPO = Ear position; EPP = Ears per plant; EA = Ear aspect; GTX = Grain texture; GMH = Grain moisture at harvesting; EL = Ear length; ED = Ear diameter; KRN = Kernel row number; KER = Kernels per ear row; EW = Ear weight; SHL = Shelling percentage; HKW = Hundred kernels weight; GY = Grain yield

Significant differences of GCA, SCA, and GCA: SCA variances existed for most traits considered, which reveals the presence of additive and non-additive gene action (Table 3.3 and Table 3.4). The ratio of GCA: SCA variances was very high and more one for AD, SD, PH, EPO, GTX, GMH, KRN, and SHL (Table 3.3) under intermediate drought stress. High significant difference of ratio of GCA: SCA variance across non-drought environments existed for AD, SD, EH, EPO, EPP, EA, GTX, GMH, EL KER, EW, and HKW. The estimate of components of variances were also expressed in Baker's ratio, where the ratio had range between (-0.61 to 1.40) in Table 3.3. Considered trait with the highest Baker's ratio that was above unity (one) was revealed to be ASI, which had a ratio of 1.40 (Table 3.3). across non-drought environments (Table 3.4), the Baker's ratio ranged between (-0.39 to 0.93), where 0.93 was the high ratio that was close to unity and it was observed for SD.

Table 3.4. Combining ability and genetic component variances of line*tester developed single-cross maize hybrids grown across optimal irrigated and rain-fed conditions

Source	DF	AD	SD	ASI	PH	EH	EPO	EPP	EA	GTX
ENV	3	3086.59	3128.98	1.56	159490.19	18373.62	0.10	5.68	0.96	3.30
REP(ENV)	4	181.12	27.92	0.00	3939.49	5458.04	0.08	0.87	2.03	0.30
CROSSES	109	76.50***	74.71***	0.00***	1515.45***	1143.81***	0.01***	0.33***	2.55***	2.07***
LINES	10	2454.78***	217.37***	0.01***	4406.32***	4917.01***	0.05***	1.59***	5.20***	7.91***
TESTERS	9	3863.29***	419.16***	0.01***	6150.43***	4477.73***	0.03***	0.79***	8.18***	11.60***
LINES*TESTERS	90	2020.28***	24.41***	0.00***	730.74***	391.17***	0.00	0.15***	1.69*	0.47***
ENV*CROSSES	327	26.10***	27.18***	0.00***	527.94***	266.07**	0.00	0.09**	1.28	0.16
ENV*LINES	30	2272.23***	72.09***	0.00**	1915.31***	889.36***	0.01*	0.13**	1.64	0.30***
ENV*TESTERS	27	1106.95***	42.03***	0.01***	596.29*	403.44**	0.00	0.13**	0.68	0.33***
ENV*LINES*TESTERS	270	5154.97***	20.70***	0.00*	366.95	183.08	0.00	0.09*	1.30	0.13
Error	436	9.62	9.48	0.00	357.89	198.26	0.00	0.07	1.27	0.14
R^2		82.76	86.52	90.05	84.21	76.95	61.78	73.61	56.09	82.84
CV (%)		4.03	3.97	3.25	6.98	11.26	14.13	23.50	35.14	11.61
COVHSlines		41.47	1.77	0.00	26.59	47.74	0.00	0.02	0.04	0.09
COVHStesters		66.94	4.24	0.00	58.98	43.93	0.00	0.01	0.08	0.12
COVHS		54.81	3.07	0.00	43.56	45.75	0.00	0.01	0.06	0.11
COVFS		359.74	7.88	0.00	132.18	115.79	0.00	0.03	0.17	0.26
Var(GCA)		54.81	3.07	0.00	43.56	45.75	0.00	0.01	0.06	0.11
Var(SCA)		-391.84	0.46	0.00	45.47	26.01	0.00	0.01	0.05	0.04
Var(GCA) x ENV		-165.02	1.73	0.00	42.33	22.06	0.00	0.00	-0.01	0.01
Var(SCA) x ENV		2572.67	5.61	0.00	4.53	-7.59	0.00	0.01	0.01	-0.01
Var(GCA)/Var(SCA)		-0.14	6.61	0.28	0.96	1.76	2.35	1.60	1.26	2.52
Baker's ratio		-0.39	0.93	0.36	0.66	0.78	0.82	0.76	0.72	0.83
Var(Additive)		109.62	6.13	0.00	87.11	91.50	0.00	0.02	0.12	0.22
Var(Dominance)		-391.84	0.46	0.00	45.47	26.01	0.00	0.01	0.05	0.04

Cont.

Source	DF	GMH	EL	ED	KRN	KER	EW	SHL	HKW	GY
ENV	3	489.63	401.62	45.64	1.60	384.92	140985.41	6178.00	3545.41	468.73
REP(ENV)	4	11.04	25.86	3.65	1.38	69.43	3833.10	126.94	47.91	69.84
CROSSES	109	4.62***	12.14***	0.86***	4.60***	55.66***	7089.38***	80.88***	77.64***	16.28***
LINES	10	19.16***	73.46***	1.90***	12.70***	325.01***	33759.90***	199.27***	342.65***	65.43***
TESTERS	9	14.21***	22.37***	1.37***	17.88***	146.31***	17101.19***	206.63***	194.87***	27.34***
LINES*TESTERS	90	2.05	4.30***	0.69***	2.38***	16.67***	3124.81***	55.15***	36.47***	9.71***
ENV*CROSSES	327	1.94	2.29**	0.18	1.14	9.22	2103.33*	48.59***	20.40	4.98***
ENV*LINES	30	2.69	4.59***	0.25	1.17	23.42***	2752.36*	82.34***	22.30	14.55***
ENV*TESTERS	27	2.91	3.42**	0.18	1.41	8.30	2529.92	69.95**	13.64	7.60***
ENV*LINES*TESTERS	270	1.75	1.92	0.17	1.11	7.74	1988.56	42.71**	20.87	3.66
Error	436	1.96	1.68	0.25	1.02	7.97	1707.02	33.10	18.05	3.54

R^2	75.60	82.18	73.67	66.52	75.16	71.84	75.20	76.74	76.72
CV (%)	8.02	7.65	10.87	7.07	7.49	18.37	7.24	11.70	24.69
COVHSlines	0.20	0.83	0.01	0.13	3.66	373.39	1.31	3.81	0.56
COVHStesters	0.13	0.19	0.01	0.17	1.47	152.67	1.41	1.88	0.16
COVHS	0.16	0.49	0.01	0.15	2.51	257.78	1.36	2.80	0.35
COVFS	0.34	1.35	0.08	0.47	6.21	703.29	5.47	7.99	1.49
Var(GCA)	0.16	0.49	0.01	0.15	2.51	257.78	1.36	2.80	0.35
Var(SCA)	0.04	0.30	0.07	0.16	1.12	142.03	1.56	1.95	0.76
Var(GCA) x ENV	0.05	0.10	0.00	0.01	0.39	31.08	1.59	-0.14	0.35
Var(SCA) x ENV	-0.10	0.12	-0.04	0.04	-0.11	140.77	4.80	1.41	0.06
Var(GCA)/Var(SCA)	4.42	1.66	0.16	0.95	2.25	1.81	0.88	1.44	0.46
Baker's ratio	0.90	0.77	0.25	0.66	0.82	0.78	0.64	0.74	0.48
Var(Additive)	0.32	0.99	0.02	0.30	5.02	515.55	2.72	5.60	0.70
Var(Dominance)	0.04	0.30	0.07	0.16	1.12	142.03	1.56	1.95	0.76

Note: AD = Days to anthesis; SD = Days to silking; ASI = Anthesis-Silking interval; PH = Plant height; EH = Ear height; EPO = Ear position; EPP = Ears per plant; EA = Ear aspect; GTX = Grain texture; GMH = Grain moisture at harvesting; EL = Ear length; ED = Ear diameter; KRN = Kernel row number; KER = Kernels per ear row; EW = Ear weight; SHL = Shelling percentage; HKW = Hundred kernels weight; GY = Grain yield

3.3.2. Contribution of lines, testers, and their interaction to total variance of crosses

The lines contribution, testers contribution, and line × tester contribution to total variances is presented in Table 3.5. The contribution of lines to total variance under drought and non-drought conditions was higher than of testers for approximately 50% of the considered traits (Table 3.5). The lines had higher contribution to the total hybrid variances than testers for SD, AD, PH, EH, EPP, EPO, EA, EW, and GY under drought, while under non-drought conditions lines showed higher contribution to the total hybrid variances than testers for ASI, EPP, EPO, GMH, EL, KER, EW, HKW, and GY. The line x tester interaction had a higher contribution to the total hybrid variance than both lines and testers under drought for ASI, PH, EH, EA, GMH, EL, ED, KER, EW, SHL, HKW, and GY. Across non-drought environment, the line x tester interaction had a higher contribution to the total variation than lines and testers for ASI, EA, ED, KRN, SHL, and GY.

Table 3.5. Contribution of lines, testers, and lines x tester interaction to variation among crosses under drought and across non-drought environments.

Trait	Drought			Across Non-drought		
	Lines	Testers	Lines x Testers	Lines	Testers	Lines x Testers
	%	%	%	%	%	%
Days to anthesis	50.46	24.83	24.71	29.44	46.33	24.23
Days to silking	51.76	18.76	29.48	26.69	46.33	26.98
Anthesis-Silking interval	19.90	25.51	54.59	19.93	15.63	64.44
Plant height	36.28	11.62	52.09	26.68	33.51	39.81
Ear height	35.29	16.36	48.34	39.44	32.32	28.24
Ear position	46.72	15.90	37.38	44.03	21.07	34.91
Ears per plant	48.75	12.22	39.03	44.03	19.60	36.37
Ear aspect	26.39	13.70	59.91	18.75	26.53	54.71
Grain texture	36.21	36.06	27.74	35.00	46.23	18.77

Trait	Drought			Across Non-drought		
	Lines	Testers	Lines x Testers	Lines	Testers	Lines x Testers
	%	%	%	%	%	%
Grain moisture at harvesting	13.93	24.59	61.47	38.03	25.39	36.58
Ear length	18.03	29.62	52.35	55.53	15.22	29.25
Ear diameter	18.20	7.82	73.98	20.23	13.14	66.62
Kernel row number	18.57	40.83	40.60	25.30	32.07	42.64
Kernels per ear row	8.45	41.02	50.53	53.57	21.70	24.73
Ear weight	9.50	5.79	84.72	43.69	19.92	36.39
Shelling percentage	6.49	26.80	66.71	22.60	21.10	56.30
Hundred kernels weight	13.58	17.73	68.69	40.49	20.72	38.79
Grain yield	17.65	9.12	73.23	36.88	13.87	49.25

3.3.3. Breeding Potential of Parents

General combining ability effect in drought conditions and non-drought environments

The GCA effects were estimated among lines and testers for 18 plant traits (Table 3.6). The positive or negative significant GCA effect was exhibited between various lines and testers for all traits under drought conditions. The line CML440 presented a highly significant negative GCA effect for AD, SD, PH, and EH, CML540 presented highly significant negative effect for AD, SD, and EH where CZL0919 exhibited significant negative GCA effect for AD and SD. The line CML571 exhibited significant negative GCA effect for AD only. The line CML550 showed significant negative GCA for PH only. Other lines like CKDHL0467 presented a highly significant positive GCA for AD, SD, and EH while CKDHL0378 presented significant positive GCA for AD and SD. Line CZL1380 and tester CML539 showed positive significant GCA effect for GY. Among testers CML539 is the potential male parent having negative and highly significant GCA for AD, SD, and EH while tester CML545 showed highly significant negative GCA for AD, SD, PH, and EH (Table 3.6).

Table 3.6. General combining ability (*gca*) effects of parental maize inbred lines for grain yield and associated traits under intermediate drought conditions

Parent	AD	SD	ASI	PH	EH	EPO	EPP	EA	GTX
Lines									
CML550	1.48**	0.60	-0.03	-7.97*	3.24	0.03***	0.11*	0.08	-0.56***
CML568	1.63***	2.85***	0.05*	-6.75	1.23	0.02**	0.28***	0.48**	0.04
CML571	-0.42	0.50	0.05	14.42***	10.04***	0.02**	-0.14**	0.08	0.49***
CKDHL0378	1.43**	2.65**	0.06*	-2.30	-3.17	-0.01	0.18***	-0.37*	-0.46***
CZL0919	-1.07*	-0.40	0.04	7.13	-3.32	-0.03***	-0.17***	-0.42*	-0.36**
CML440	-4.12***	-4.65***	-0.02	-22.68***	-15.69***	-0.03***	-0.33***	0.33*	-0.46***
CKDHL0467	4.68***	3.90***	-0.04	-6.65	8.83***	0.05***	0.28***	0.08	0.59***
CML540	-3.72***	-4.60***	-0.04	7.08	-8.62***	-0.05***	-0.14**	-0.32*	0.44***
CML572	-0.47	-0.30	-0.02	6.75	1.78	0.00	-0.12*	-0.07	0.34**
CZL1380	1.28**	1.60*	0.02	11.13**	4.36	0.00	-0.01	-0.27	0.09
MAK1-122	-0.72	-2.15**	-0.07*	-0.15	1.33	0.01	0.05	0.43**	-0.16
SE(gi)	0.47	0.67	0.03	3.98	2.29	0.01	0.05	0.16	0.11
Testers									
CML442	0.15	1.15*	0.04	1.25	-4.24	-0.02***	0.02	0.22	0.88***
CML312	1.24**	-0.13	-0.09***	4.96	1.32	0.00	0.02	0.10	-0.35**
CML537	-0.35	1.19	0.07*	2.98	-1.02	-0.01	0.17***	0.10	-0.53***
CML539	-1.26**	-1.81**	-0.03	-6.16	-6.02**	-0.01	-0.12**	0.23	-0.25*
CML545	-2.85***	-3.04***	0.00	-7.93*	-8.08***	-0.02***	-0.03	-0.13	0.06
CML444	0.97*	-0.04	-0.04	-5.42	-0.30	0.01	-0.15**	-0.09	0.15
CML566	3.24***	2.24***	-0.05	-1.69	1.29	0.01	0.04	-0.18	0.06
CML547	0.47	1.46*	0.06	11.20**	8.64***	0.02**	0.13**	-0.09	0.11
CML395	0.60	0.87	0.02	4.95	3.65	0.01	-0.04	-0.22	-0.48***
K64R	-2.21	-1.90	0.02	-4.14	4.76*	0.03	-0.05**	0.50**	0.34**
SE(gj)	0.44	0.64	0.03	3.80	2.18	0.01	0.04	0.15	0.11
Cont.									
Lines	GMH	EL	ED	KRN	KER	EW	SHL	HKW	GY

CML550	0.23	-1.12***	-0.02	0.32*	-1.61**	-10.40	-0.54	-0.27	0.18
CML568	0.30	-0.44	-0.24*	0.33*	-0.95	-25.10	0.13	-1.37	-0.52
CML571	-0.38	0.54	-0.14	-0.37*	1.57**	-8.28	0.46	-0.97	-0.16
CKDHL0378	0.15	0.00	-0.19*	-0.33*	-0.21	-24.37	-2.79*	-0.97	-0.57*
CZL0919	-0.16	0.80**	0.03	-0.30	0.11	4.16	0.30	2.23**	0.28
CML440	-0.67***	0.18	-0.11	0.35*	-0.73	-5.25	0.03	-0.07	-0.91**
CKDHL0467	0.23	0.20	0.01	0.25	0.68	-2.25	-1.89	-1.47	0.02
CML540	-0.05	0.63*	0.09	-0.01	0.05	2.18	1.15	3.13***	0.12
CML572	0.14	0.03	0.12	-0.41*	1.05	-1.51	1.04	-0.37	0.38
CZL1380	0.01	0.08	0.32**	0.56***	0.53	8.29	1.32	0.73	0.98***
MAK1-122	0.20	-0.91**	0.13	-0.39*	-0.48	62.50**	0.78	-0.57	0.20
SE(gi)	0.19	0.28	0.09	0.16	0.59	22.29	1.30	0.85	0.28
Testers									
CML442	0.15	0.87**	-0.02	-0.24	2.42***	4.29	-0.38	-0.15	0.43
CML312	0.00	-0.72**	-0.01	0.59***	-2.27***	-15.06	1.40	-0.42	-0.20
CML537	-0.12	0.70**	-0.07	0.43**	0.24	48.18	-3.62**	-1.42*	-0.58*
CML539	-0.20	0.81**	-0.06	-0.54***	1.79**	3.59	5.72***	1.40***	0.53*
CML545	-0.24	-0.84**	0.09	-0.50**	-1.54**	1.81	-0.88	3.95*	0.47
CML444	0.69***	-1.10***	0.04	0.76***	-1.78**	-9.61	-0.57	-1.60	-0.02
CML566	0.40*	-0.84**	0.26**	0.54***	-2.44***	-7.09	-3.51**	0.85	-0.31
CML547	0.19	0.57*	-0.02	-0.84***	2.26***	-2.56	0.15	-0.33	-0.11
CML395	-0.07	0.35	-0.10	-0.05	-1.11	-12.02	1.02	-0.96	0.09
K64R	-0.79***	0.20	-0.09	-0.15	2.43***	-11.55	0.67	-1.33	-0.31
SE(gj)	0.18	0.27	0.09	0.15	0.56	21.25	1.24	0.81	0.27

Note: AD = Days to anthesis; SD = Days to silking; ASI = Anthesis-Silking interval; PH = Plant height; EH = Ear height; EPO = Ear position; EPP = Ears per plant; EA = Ear aspect; GTX = Grain texture; GMH = Grain moisture at harvesting; EL = Ear length; ED = Ear diameter; KRN = Kernel row number; KER = Kernels per ear row; EW = Ear weight; SHL = Shelling percentage; HKW = Hundred kernels weight; GY = Grain yield

Under non-drought conditions, all traits had either positive or negative GCA effect between different lines and testers. The line CML540, CZL0919 and CML572 exhibited highly significant negative GCA effect for AD, SD, PH, and EH. The line CML571 presented a highly significant negative GCA effect for only AD and SD, while CML440 showed highly significant negative GCA effect for PH and EH, and MAK1-122 showed highly significant negative GCA effect for PH only (Table 3.7). The line CML440 and CKDHL0467 presented highly significant positive GCA effect for AD and SD. Various lines across non-drought conditions presented positive significant GCA effect for GY, namely CML568, CKDHL0378, CKDHL0467, CML572, and CZL1380. Out of the testers, tester CML442, CML539, CML545 and K64R presented highly significant negative GCA effect for AD, SD, PH, and EH, while line CML537 showed a significant negative GCA effect for EH only. The testers CML312 and CML547 presented positive significant GCA effect for GY.

Table 3.7. General combining ability (*gca*) effects of parental maize inbred lines for grain yield and associated traits across non-drought environments

Parent	AD	SD	ASI	PH	EH	EPO	EPP	EA	GTX
Lines									
CML550	0.06	-0.16	-0.01*	-1.50	4.01*	0.02*	0.11***	0.12	-0.32***
CML568	1.44***	1.44***	0.00	1.79	6.87***	0.03***	0.26***	0.18	0.04
CML571	-1.91***	-1.68***	0.01	6.57**	-0.88	-0.01	-0.15***	-0.28*	0.57***
CKDHL037	1.26***	1.24***	0.00	3.99	3.64*	0.01	0.03	0.21	-0.33***
CZL0919	-0.93**	-0.92**	0.00	6.14**	-4.81**	-0.03***	-0.15***	-0.40**	-0.18***
CML440	2.02***	2.23***	0.01*	-17.47***	-13.68***	-0.02**	-0.12***	0.32*	-0.47***
CKDHL046	3.01***	2.54***	-0.02***	1.21	11.34***	0.04***	0.16***	0.06	0.16***
CML540	-2.18***	-2.12***	0.00	0.13	-11.39***	-0.04***	-0.15***	-0.20	0.21***
CML572	-2.06***	-1.90***	0.01	1.98	-3.42*	-0.02*	-0.06	-0.20	0.34***
CZL1380	-0.07	-0.11	0.00	6.76**	7.50***	0.02*	0.01	-0.13	0.06
MAK1-122	-0.64	-0.56	0.00	-9.60***	0.81	0.02***	0.06*	0.33**	-0.07
SE(gi)	0.35	0.34	0.00	2.12	1.57	0.01	0.03	0.13	0.04
Testers									
CML442	-1.98***	-1.97***	0.00	-8.79***	-8.34***	-0.02*	-0.01	0.15	0.86***
CML312	1.14***	1.29***	0.01	7.31***	2.84	0.00	-0.01	-0.11	-0.50***
CML537	2.23***	2.32***	0.01	5.77**	-3.09*	-0.02**	0.16***	0.18	-0.32***
CML539	-1.42***	-1.34***	0.00	-5.19*	-8.64***	-0.02***	-0.07*	0.03	-0.18***
CML545	-2.75***	-2.83***	0.00	-6.82***	-7.30***	-0.01	-0.02	-0.15	0.15***
CML444	0.23	-0.10	-0.02***	-7.46***	2.09	0.02**	-0.11***	-0.22	-0.06
CML566	4.32***	4.14***	-0.01*	10.49***	12.22***	0.03***	0.05	-0.08	0.08*
CML547	-0.15	0.06	0.01**	10.95***	6.15***	0.00	0.16***	-0.41***	0.01
CML395	0.62	0.60	0.00	3.46	6.67***	0.01*	-0.07*	-0.11	-0.15***
K64R	-2.25***	-2.16***	0.00	-9.70***	-2.60	0.01	-0.08**	0.72***	0.11***
SE(gj)	0.33	0.33	0.00	2.02	1.50	0.01	0.03	0.12	0.04

Cont.

Lines	GMH	EL	ED	KRN	KER	EW	SHL	HKW	GY
CML550	-0.08	-0.95***	0.07	0.17	-0.80*	-17.89***	-0.28	-2.98***	0.03
CML568	0.59***	-0.21	-0.02	0.25*	-0.45	-16.45***	-0.63	-1.93***	0.52*
CML571	-0.65***	1.74***	0.01	-0.36**	4.86***	16.45***	0.35	-1.19*	-0.38
CKDHL037	0.95***	0.14	-0.09	-0.44***	0.02	-5.36	0.65	0.05	0.52*
CZL0919	-0.17	1.00***	0.20***	-0.37**	0.31	18.49***	-1.57*	2.45***	-0.21
CML440	-0.50**	-0.43**	-0.22***	0.03	-3.36***	-30.97***	0.40	0.77	-2.20***
CKDHL046	0.24	-0.24	-0.25***	0.43***	0.01	13.25**	-2.86***	-2.00***	0.78***
CML540	-0.11	0.44**	-0.01	-0.04	-0.21	10.42*	0.52	3.04***	-0.37
CML572	-0.31*	0.47**	0.20***	-0.31**	1.11***	24.72***	0.65	2.90***	0.80***
CZL1380	0.39*	-0.08	0.17**	0.85***	0.34	16.62***	-0.68	-0.51	1.00***
MAK1-122	-0.35*	-1.88***	-0.06	-0.21	-1.83***	-29.28***	3.45***	-0.60	-0.47*
SE(gi)	0.16	0.14	0.06	0.11	0.32	4.62	0.64	0.48	0.21
Testers									
CML442	-0.71***	0.88***	-0.08	-0.09	2.21***	-6.36	0.64	-1.91***	0.04
CML312	0.23	0.10	0.10	0.36***	-1.26***	7.54	0.80	1.44**	0.60**
CML537	-0.32*	0.01	-0.04	0.31**	-0.92**	-3.19	-0.76	-0.52	-0.16
CML539	-0.03	0.60***	-0.11*	-0.30**	1.20***	1.75	1.21*	0.62	-0.12
CML545	-0.11	-0.53***	0.12*	-0.13	-1.12***	-3.32	-0.17	1.10*	0.04
CML444	0.17	-0.50***	0.02	0.82***	-0.46	1.10	-1.36*	-1.05*	-0.56**
CML566	0.59***	-0.70***	0.23***	0.36***	-1.49***	22.42***	-2.86***	0.27	0.09
CML547	-0.04	0.13	0.01	-0.68***	0.90**	13.85**	0.68	2.24***	1.14***
CML395	0.54**	0.22	-0.06	-0.24*	-0.33	-3.46	-0.81	0.28	-0.20
K64R	-0.32*	-0.22	-0.20***	-0.41***	1.27***	-30.33***	2.62***	-2.46***	-0.86***
SE(gj)	0.15	0.14	0.05	0.11	0.30	4.40	0.61	0.45	0.20

Note: AD = Days to anthesis; SD = Days to silking; ASI = Anthesis-Silking interval; PH = Plant height; EH = Ear height; EPO = Ear position; EPP = Ears per plant; EA = Ear aspect; GTX = Grain texture; GMH = Grain moisture at harvesting; EL = Ear length; ED = Ear diameter; KRN = Kernel row number; KER = Kernels per ear row; EW = Ear weight; SHL = Shelling percentage; HKW = Hundred kernels weight; GY = Grain yield

3.3.4. Genetic potential of crosses

Specific combining ability effect

The estimated specific combining ability effect of the best ten maize single crosses for grain yield under drought and non-drought environment are presented in Table 3.8. On this study, significant negative SCA effect were estimated from the CKDHL0467 x CML395 for AD only under non-drought conditions (Table 3.8). A positive significant SCA is desired for grain yield. Under drought conditions the following crosses exhibited a significant positive SCA for grain yield, CZL0919 x CML539, CZL1380 x CML395, CML440 x CML312, CML540 x CML547, CKDHL0378 x CML442, and CML571 x CML442. The cross CML540 x CML547 is the only cross that had a significant positive SCA effect for grain yield and presented significant positive SCA effect for HKW which is one of the important yields contributing traits. The cross CKDHL0467 x CML312 disclosed significant positive SCA for HKW and a positive SCA for grain yield (Table 3.8). The following crosses disclosed a significant positive SCA effect across non-drought conditions, CKDHL0467 x CML312, CML540 x CML442, CML440 x CML566, CKDHL0378 x CML566, and CKDHL0467 x CML395. The cross CKDHL0467 x CML312 is the only cross that presented a significant positive SCA for grain yield and a significant positive SCA for HKW under non-drought conditions (Table 3.8).

Table 3.8. Specific combining ability (*SCA*) effects of best ten maize single crosses for grain yield and associated traits under intermediate drought and across non-drought environments.

Crosses	AD	SD	ASI	PH	EH	EPO	EPP	EA	GTX
Drought									
CZL0919 x CML539	1.16	0.31	-0.04	2.46	-0.43	-0.01	-0.08	-0.53	0.40
CZL1380 x CML395	-0.55	-2.37	-0.09	3.69	4.38	0.01	-0.14	-1.23*	0.68
CML440 x CML312	-1.29	-0.62	0.06	6.49	-4.23	-0.03	0.05	-0.15	-0.40
CML540 x CML547	-1.42	-1.26	0.01	-24.85	-1.29	0.04*	-0.06	-0.31	0.24

CKDHL0378 x	0.25	-2.70	-0.12	22.32	10.48	0.01	-0.30*	-0.63	-0.13
CML571 x CML442	-1.40	-2.05	-0.02	16.93	7.26	0.00	0.07	-0.08	0.42
CML550 x CML537	-0.80	-1.69	-0.03	6.76	5.50	0.01	-0.20	0.10	-0.12
CKDHL0467 x	-1.59	-1.67	0.02	8.95	15.08*	0.04*	-0.31*	-0.40	0.05
CKDHL0467 x	-0.50	2.51	0.12	7.94	4.92	0.00	0.20	-0.40	0.23
CML568 x CML566	-0.04	0.01	0.02	6.54	-6.12	-0.04*	0.07	-0.02	0.19
SE(ij)	1.47	2.12	0.08	12.59	7.23	0.02	0.15	0.51	0.35
Trait grand mean	76.68	76.90	1.15	234.91	99.44	0.42	1.26	3.22	3.21

Cont.	**GMH**	**EL**	**ED**	**KRN**	**KER**	**EW**	**SHL**	**HKW**	**GY**
CZL0919 x CML539	0.05	0.03	0.27	0.18	1.10	-22.30	25.18***	1.50	2.23*
CZL1380 x CML395	0.40	0.80	0.23	0.00	2.50	7.98	15.60***	1.36	2.06*
CML440 x CML312	-0.24	-0.07	0.39	0.57	0.41	8.76	9.46*	1.62	1.93*
CML540 x CML547	-1.15	-0.05	0.22	0.36	1.34	32.73	0.90	7.33**	1.92*
CKDHL0378 x	-0.26	1.02	0.35	0.25	2.20	40.03	-3.06	4.25	1.91*
CML571 x CML442	-0.53	3.06**	0.30	0.12	4.26	60.44	0.09	5.25	1.87*
CML550 x CML537	-0.22	0.65	0.40	0.43	0.52	-29.53	2.34	2.82	1.59
CKDHL0467 x	-0.39	0.08	0.37	-0.17	1.83	34.36	0.28	8.02**	1.43
CKDHL0467 x	0.08	0.32	0.04	0.33	1.57	-41.38	4.45	-0.98	1.41
CML568 x CML566	0.29	0.17	0.14	0.97	1.80	35.04	0.53	-0.35	1.36
SE(ij)	0.61	0.88	0.30	0.49	1.86	70.48	4.11	2.68	0.88
Trait grand mean	15.81	14.69	4.56	14.16	31.67	175.55	82.59	34.87	5.71

Non-drought	**AD**	**SD**	**ASTR**	**PH**	**EH**	**EPO**	**EPP**	**EA**	**GTX**
CKDHL0467 x	0.69	0.87	0.01	11.16	20.59***	0.05*	0.03	-0.40	0.15
CML540 x CML442	-0.39	-0.33	0.00	-5.89	-0.65	0.00	-0.02	0.23	-0.01
CML440 x CML566	3.24**	2.71*	-0.02	-4.04	4.28	0.03	0.08	-0.20	0.20
CKDHL0378 x	1.63	1.33	-0.02	13.29*	12.76*	0.02	0.10	-0.83*	0.06
CKDHL0467 x	-2.79*	-3.06**	-0.01	8.64	-2.52	-0.02	0.06	-0.27	-0.08
CML540 x CML547	-1.59	-1.36	0.01	-10.76	-6.49	0.00	-0.12	-0.34	0.09

CML568 x CML395	1.90	2.04	0.01	10.74	-0.20	-0.02	0.01	0.10	0.04
MAK1-122 x CML566	-1.85	-1.12	0.03*	10.71	-5.50	-0.04	0.06	0.29	0.05
CZL1380 x CML566	-1.30	-1.57	-0.01	8.78	11.34*	0.02	-0.03	-0.12	-0.08
CML540 x CML312	-0.38	-0.84	-0.02	5.95	2.44	0.00	0.15	0.36	0.10
SE(ij)	1.10	1.09	0.01	6.69	4.98	0.02	0.09	0.40	0.13
Trait grand mean	76.92	77.55	1.17	270.98	125.02	0.46	1.14	3.20	3.19

Cont.	**GMH**	**EL**	**ED**	**KRN**	**KER**	**EW**	**SHL**	**HKW**	**GY**
CKDHL0467 x	0.22	1.19**	0.61***	-0.07	2.66**	42.47**	3.78	4.01**	1.87**
CML540 x CML442	0.45	0.14	-0.44*	0.35	-0.80	0.58	-2.03	0.08	1.65*
CML440 x CML566	-0.77	0.39	0.12	0.44	0.44	1.10	2.74	-0.84	1.63*
CKDHL0378 x	0.74	-1.18*	0.11	0.33	-1.77	0.77	2.23	2.13	1.57*
CKDHL0467 x	0.99*	0.40	-0.16	-0.27	1.01	26.35	-1.08	1.67	1.41*
CML540 x CML547	-0.33	-0.03	0.25	-0.38	-0.63	20.85	2.19	2.92	1.29
CML568 x CML395	-0.10	0.50	0.23	0.19	-0.19	6.90	5.76**	1.10	1.27
MAK1-122 x CML566	0.82	0.79	-0.10	-0.06	-0.16	5.06	1.12	1.03	1.26
CZL1380 x CML566	-0.21	1.08*	0.11	-0.95**	1.59	13.89	-1.49	-0.30	1.25
CML540 x CML312	0.05	0.69	0.17	-0.13	2.31*	8.54	2.35	0.48	1.23
SE(ij)	0.50	0.46	0.18	0.36	1.00	14.61	2.03	1.50	0.67
Trait grand mean	17.46	16.95	4.59	14.28	37.69	224.90	79.45	36.30	7.62

Note: AD = Days to anthesis; SD = Days to silking; ASI = Anthesis-Silking interval; PH = Plant height; EH = Ear height; EPO = Ear position; EPP = Ears per plant; EA = Ear aspect; GTX = Grain texture; GMH = Grain moisture at harvesting; EL = Ear length; ED = Ear diameter; KRN = Kernel row number; KER = Kernels per ear row; EW = Ear weight; SHL = Shelling percentage; HKW = Hundred kernels weight; GY = Grain yield

3.4. Discussion

The significant crosses mean squares for majority of traits under drought, including GY and for all traits under non-drought conditions indicates that the parents involved in hybridization had a lot of variation, which could be exploited by breeders during selection for improvement of traits and identification of the most desirable hybrids. Similar result was reported by Anilkumar and Chandappa (2019) and Rahman et al. (2013), which revealed that the mean squares due to crosses exhibited highly significant differences for most of the traits apart from ASI indicating that these crosses were adequately different from each other for these traits and therefore, selection is possible to determine the most desirable crosses. Under drought the lines performed differently in respect of GCA for some traits, specifically for ASI, EPO, EPP, EA, GTX, GMH, EL, ED, KRN, KER, and GY. In the same environment, the testers differed in GCA for SD, PH, EPO, EPP, EA, GTX, GMH, EL, KRN, KER, and GY. This indicates that the lines and testers used were diverse resulting in expression of variation among progenies of these considered traits and hence, selection is possible to identify the most desirable crosses. These observations agree with the findings reported by Sundararajan and Senthil Kumar (2011**)** and Tesfaye et al. (2019)**.**

Specific combining ability (SCA) was significant for AD, EH, EPO, EPP, EL, KER, and GY. Tesfaye et al. (2019) observed similar result, mean squares from the combined analysis for line x tester (SCA) interaction showed highly significant differences for some traits which included AD, SD, EH, ED, TKW, and GY reflecting an overwhelming contribution of non-additive effect type gene action in these traits. Some traits showed significance of either the lines or testers and the line x tester interaction implying that both GCA effect (which reflect additive gene effect), these traits include EH, EPO, EPP, EL, and KER and SCA effect (which reflect non-additive gene effect) were important for the improvement of these traits. Across non-drought environments, lines and testers disclosed significant GCA for almost all traits in exclusion of

EW. This indicated the presence of additive type of gene actions in the expression of these traits (Adenike et al., 2017). The SCA was significant for all traits, excluding EPO and GMH across non-drought conditions, as reflected by line x tester mean squares showing a presence of non-additive gene action in the traits. Therefore, non-additive genetic variance can be exploited for improvement of these traits in cultivar development (Rahman et al., 2013).

The genotype x environment interaction significantly influenced the performance of the hybrid progenies. The significant environment x lines or environment x testers for traits AD, SD, ASI, PH, EH, EPO, EPP, GTX, EL, KER, EW, SHL, and GY across non-drought conditions imply that different lines or testers performed differently in respect of GCA in diverse environments and that various lines and testers are adapted and stable to a different specific environment. Thus testing inbred lines in various test environments will ensure selection of stable parents that can perform to the potential of that environment (Seyoum et al. 2016) or emphasizing the importance of the environment in phenotypic expression of agronomic characters (Bello and Olaoye, 2009; Murtadha et al., 2018). The GCA of the lines and testers was influenced by the environment indicating that lines and testers did not respond consistently across the environments and that using different line and testers at different environments for hybrid development and selection would be more effective and successful when based on performance across environments (Tesfaye et al., 2019). All traits indicated significance of line, testers and/or line x tester except for EW which indicated significance for line x tester interaction only across non-drought conditions, this implies that both GCA and SCA were considered important for improvement of all traits except EW. These observations agree with findings reported by Murtadha et al. (2018). The significant environment x lines x testers mean squares across non-drought conditions for AD, SD, ASI, EPP, and SHL imply that the SCA effect was influenced by the environment.

Under drought environment, estimates of GCA variance reflected were of higher magnitude than SCA variance for AD, SD, PH, KRN, EA, and GTX, while SCA variance were of higher magnitude than GCA variance for EL, EW, and HKW. Across non-drought environments GCA variance reflected to be of higher magnitude than SCA variance for AD, SD, EH, GTX, GMH, KER, EW, and HKW, whereas SCA variance was of higher magnitude than GCA variance for PH and GY. When the estimates of SCA variance is higher than GCA variance for considered traits that indicate that non-additive effect gene action is present and when the estimates of GCA variance is higher than SCA variance for considered traits it shows that additive effect gene action is present (Sundararajan and Senthil Kumar, 2011). Non-additive effect gene action was also reported by Seyoum et al. (2016) for AD and SD and Amin et al. (2015) for PH, EL, KER, and HGW. Additive effect gene action was also reported by Premlatha and Kalamani (2010) for GY. If the ratio of SCA: GCA variances is more than unity it reflects that additive type gene action is present and GCA variance is more important than SCA for considered traits (Sundararajan and Senthil Kumar, 2011). The high Baker's ratios, SD, ASI, PH, EPO, EPP, EA, GTX, GMH, KRN, and SHL, under drought, and for AD, SD, EH, EPO, EPP, EA, GTX, GMH, EL, KER, EW, and HKW, across non-drought imply that the performance of the crosses for these traits could be predicted based on the GCA effect of the parents involved (Baker, 1978). In the drought environment the lines had a greater contribution to total hybrid variance than testers for AD, SD, PH, EH, EPO, EPP, EA, and GY and across non-drought environments line had greater contribution to the total maize variance for ASI, EPP, EPO, GMH, EL, KER, EW, HKW, and GY. This specify that lines contributed to the maximum in the total hybrid variance than testers (Jahan et al., 2015). The combined contribution of lines and testers was more than the contribution of line x tester interaction for AD, SD, EPO, EPP, GTX, and KRN in drought condition whereas across non-drought conditions combined contribution of line and testers combined was more than contribution of their interaction for AD, SD, PH, EH, EPO, EPP, GTX, GMH, EL, KRN, KER, EW, and HKW implementing that lines and testers combined, contributed

to the maximum in the total hybrid variance than their interaction for these considered traits. The line x tester interaction contributed more than the combined contribution of lines and testers for ASI, EA, GMH, ED, EW, SHL, and HKW in drought stressed environment, while across non-drought stressed environments the SCA contribution was greater than the combined contribution of line and tester towards the total hybrid variance for ASI, EA, ED, and SHL. This signify that SCA effect, that is, non-additive gene action, was present and that SCA contributed to the variation in the total hybrid variance for considered traits (Ahmed et al., 2016; Jahan et al., 2015; Talukder et al., 2016).

Analysis of combining ability effect helps breeders in the choice of parents for improvement of traits in their breeding programs. Combining ability can be positive or negative for different traits, the direction desired by the breeder depends on the trait and how it was measured (Ahmed et al., 2017; Uddin et al., 2006; Ahmed et al., 2016). In drought and across non-drought environments, all traits were either positive or negative GCA effect between lines and testers for GY and related traits. The parents with high magnitude GCA effect in the desirable direction for more traits are desirable (Ahmad and Saleem, 2003; Pswarayi and Vivek, 2008, Kamara et al., 2014; Legesse et al., 2009; and Chiuta and Mutengwa, 2020). For example, in respect of grain yield, the most desirable parents were line CZL1380 and tester CML539 under drought environments, while across non-drought environment, line CML568, CKDHL0378, CKDHL0467, CML572 and CZL1380 and testers CML312 and CML547 were the most desirable parents. These lines and testers are more desirable since they presented significant positive GCA meaning they could be selected as good combiners for grain yield improvement (Ahmad and Saleem, 2003; Pswarayi and Vivek, 2008; ; Legesse et al., 2009; Kamara et al., 2014 ; Chiuta and Mutengwa, 2020).

The parents with desirable GCA for most traits including GY were line CML572 significant GCA for 13 traits followed by CKDHL0467 (13 traits), CML568 (9 traits), CZL1380 (8 traits), and lastly

CKDHL0378 (7 traits). The line CML440 presented a highly significant negative GCA effect for AD, SD, PH, and EH, CML540 presented highly significant negative GCA effect for AD, SD, and EH where CZL0919 exhibited significant negative GCA effect for AD and SD. The line CML571 exhibited significant negative GCA effect for AD only. The line CML550 showed significant negative GCA for PH only. In contrast to this line CZL1380 showed highly significance positive GCA effect for AD, SD, and PH, line CKDHL0467 presented highly significant positive GCA effect for AD, SD, and EH, line CML568 and CKDHL0378 exhibited significant positive GCA effect for AD and SD. Lines that presented significant negative effect for AD and SD are more desirable since they indicate a quick maturing habit therefore they could be selected for the improvement of early-medium maturing hybrid and narrow anthesis-silking interval in future breeding work (Seyoum et al., 2016; Bello and Olaoye, 2009).

Adenike et al. (2017) reported similar findings in respect of AD and SD, significant negative and positive GCA effect among tested lines for days to tasselling and silking. The lines that reflect negative and significant GCA effect for PH and EH suggests that they could contribute to shorter plant height with lower ear placement , hence genotypes with shorter plant height and lower ear placement can be good in the improvement of maize for lodging resistance (Seyoum et al., 2016; Ji et al., 2006). Across non-drought environments, the line CML540, CZL0919 and CML572 exhibited highly significant negative GCA effect for AD, SD, PH, and EH**.** Additionally, these lines could be selected for improvement of early to medium maturing hybrids and lodging resistance (Rahman et al., 2013; Anilkumar and Chandappa, 2019)**.**

Crosses with high SCA effect for majority of traits including GY were CZL0919 x CML539, CZL1380 x CML395, CML440 x CML312, CML540 x CML547, CKDHL0378 x CML442, and CML571 x CML442 under drought condition. Across non-drought conditions the following crosses showed a significant positive SCA effect *viz*, CKDHL0467 x CML312, CML540 x CML442, CML440 x CML566, CKDHL0378 x CML566, and CKDHL0467 x CML395. Presence

of positive SCA effect in crosses indicate that lines were from opposite heterotic group, while negative SCA effects refer lines were from the same heterotic group. For instance, under drought conditions cross CML440 x CML312 showed positive significant SCA effect but had lines with low GCA effect for GY. Under both environmental conditions, drought and non-drought conditions, positive SCA effect were manifested by crosses of low x low (CML440 x CML312) (drought condition), low x high (CZL0919 x CML539) (drought condition), and high x high (CKDHL0467 x CML312) (non-drought conditions), showing the presence of complementary gene action for GY. Inbred lines CML440, CML312, and CZL0919 had poor general combining ability, but resulted with hybrids with higher SCA effect for GY (Anilkumar and Chandappa, 2019).

Moreover, this implies that inbred line with poor GCA might produce desirable hybrids depending on the other parent with which it combines. Under drought none of the crosses were good specific combiners for AD, SD, PH, and EH, whereas across non-drought conditions only CKDHL0467 x CML395 was considered a good specific combiner for AD hence, showed negative significant SCA effect for this trait. These parents involved inbred lines with general combiners of poor x poor GCA effect. It is evident that high specific combinations in crosses involved inbred lines with high x high, high x low, and low x low, with respect to GCA effect of the parents and SCA effect of hybrids for GY, and other traits. Therefore, the best performance of these combinations may be due to additive x additive (high x high), additive x non-additive (high x low), or non-additive x non-additive (low x low) gene interaction (Dey et al., 2014; Talukder et al., 2016; and Zhang et al., 2015).

3.5. Conclusions

The parents used to develop crosses were not selected randomly from all possible white tropical maize inbred lines, therefore model I (fixed effects) analysis was followed which render the

inferences made only applicable to the inbred lines used. The genetic analysis of line x tester developed crosses was successfully executed and the conclusions drawn are as follows

- The parents differed in general combining ability (GCA) effects for GY and other traits under drought and non-drought conditions.
- Crosses varied in specific combining ability (SCA) effects for GY and other traits under the drought and non-drought regimes.
- Line CZL1380 and tester CML539 were good general combiners for GY under drought.
- Lines CML568, CKDHL0378, CKDHL0467, CML672, and CZL1380 and testers CML312 and CML547 had good GCA effect across non-drought regime.
- Two crosses, CML540 x CML547 and CKDHL0467 x CML312 had high SCA values for GY under drought and non-drought regimes.
- The additive type of gene action was predominant for days to anthesis (AD), days to silking (SD), anthesis-silking interval (ASI) plant height (PH), ear position (EPO), ears per plant (EPP), ear aspect (EA), grain texture (GTX), grain moisture (GMH), kernel row number (KRN), and shelling percentage (SHL) under drought, and for AD, SD, ear height (EH), EPO, EPP, EA, GTX, GMH, ear length (EL), kernels per ear row (KER), ear weight (EW), and hundred kernel weight (HKW) across non-drought conditions.
- Non-additive gene action prevailed for EH, EL, ED, KER, EW, HKW, and GY under drought and for ASI, PH, ED, KRN, SHL, and GY across non-drought conditions.
- Baker's predictability ratios were quite high for AD, SD, ASI, PH, EPO, EPP, EA, GTX, GMH, KRN, and SHL under drought condition. Under non-drought conditions, high predictability ratios were realized for AD, SD, EH, EPO, EPP, GMH, EL, KER, EW, and HKW. For these traits and in the respective environmental conditions, the performance of hybrids can be predicted using the GCA of the parents.

The identified hybrids could be targeted for release as cultivars, and the types of gene action are practically relevant for improvement of early to medium maturing maize germplasm for Southern Africa.

References

Adenike, D.I., A. Richard, O. Allen, and I.A. Idris. 2017. Combining ability of tropical early maize (Zea mays L.) inbred lines for grain yield and resistance to maize streak virus disease. J. Plant Breed. Crop Sci. 9(10): 186–194. doi: 10.5897/jpbcs2017.0649.

Ahmad, A., and M. Saleem. 2003. Combining Ability Analysis in Zea mays L. Int. J. Agric. Biol. 5(3)(Department of Plant Breeding and Genetics, University of Agriculture, Faisalabad–38040, Pakistan): 239–244.

Ahmed, S., S. Begum, M. Islam, M. Ratna, and M. Karim. 2017. Combining ability estimates in maize (Zea mays L.) Through line × tester analysis. Bangladesh J. Agric. Res. 42(3): 425–436. doi: 10.3329/bjar.v42i3.34501.

Ahmed, A., S. Begum, S. Omy, M. Rohman, and M. Amiruzzaman. 2016. Evaluation of inbred lines of baby corn through line × tester method. Bangladesh J. Agric. Res. 41(2): 311–321. doi: 10.3329/bjar.v41i2.28233.

Ahmed, M.., M.H. El-Sheikh, and S.A. Shamarka. 2000. Diallel analysis of yielding ability and earliness in maize. J. Agric. Sci. 25(7)(Mansoura Univ): 3717–3726.

Amin, M., M. Amiruzzaman, A. Ahmed, and M. Ali. 2015. Evaluation of inbred lines of maize (Zea mays L.) through line × tester method. Bangladesh J. Agric. Res. 39(4): 675–683. doi: 10.3329/bjar.v39i4.22547.

Anilkumar, C., and L. Chandappa. 2019. Combining ability analysis in newly developed inbred lines of maize. 7(2): 113–124. doi: 10.20448/journal.512.2020.72.113.124.

Baker, R.J., 1978. Issues in diallel analysis. Crop Sci 18: 533–536.

Bello, O.B., and G. Olaoye. 2009. Combining ability for maize grain yield and other agronomic characters in a typical southern guinea savanna ecology of Nigeria. African J. Biotechnol. 8(11): 2518–2522. doi: 10.5897/AJB09.296.

Chiuta, N.E., and C.S. Mutengwa. 2020. Combining ability of quality protein maize inbred lines for yield and morpho-agronomic traits under optimum as well as combined drought and heat-stressed conditions. Agronomy 10(2). doi: 10.3390/agronomy10020184.

Das, A., B. Paudel, and J.S. Rohila. (2016). Potentials of proteomics in crop breeding.

de Souza Jr. C.L., L.M.M. Ottoboni, M.L.C. Vieria, and A.P. de Souza. 1997. Genetic distance of inbred lines and prediction of maize single-cross performance using RAPD markers. 94: 1023–1030.

Dey, S.S., N. Singh, R. Bhatia, C. Parkash, and C. Chandel. 2014. Genetic combining ability and heterosis for important vitamins and antioxidant pigments in cauliflower (Brassica oleracea var. botrytis L.). Euphytica 195(2): 169–181. doi: 10.1007/s10681-013-0981-4.

El-Hosary, A.A.A., and A.A. Elgammaal. 2013. Utilization of Line X Tester Model for Evaluating the Combining Ability of Some New White Maize Inbred Lines. Egypt. J. Plant Breed. 17(1): 79–92. doi: 10.12816/0003979.

Fasahat, P. 2016. Principles and Utilization of Combining Ability in Plant Breeding. Biometrics Biostat. Int. J. 4(1): 1–22. doi: 10.15406/bbij.2016.04.00085.

Griffing, B. 1956. A Generalised Treatment of the use of Diallel Crosses in Quantitative Inheritance. (Department of Genetics, Cambridge): 31–50.

Hallauer, A.R., W.A. Russell, and K.R. Lamkey. 2015. Corn Breeding. : 463–564. doi: 10.2134/agronmonogr18.3ed.c8.

Hayes, H.K., and I.J. Johnson. 1939. The Breeding of Improved Selfed Lines of Corn 1 . Agron. J. 31(8): 710–724. doi: 10.2134/agronj1939.00021962003100080008x.

Istipliler, D., E. Ilker, F. Aykut Tonk, G. Civi, and M. Tosun. 2015. Line × tester analysis and estimating combining abilities for yield and some yield components in bread wheat. Turkish

J. F. Crop. 20(1): 72–77. doi: 10.17557/.57234.

Jahan, N., M. Uddin, M. Islam, S. Hasna, and A. Saifullah. 2015. Evaluation of Inbred Line Through Line X Tester Method. J. Environ. Sci. Nat. Resour. 7(2): 79–84. doi: 10.3329/jesnr.v7i2.22211.

Ji, H.C., J.W. Cho, and T. Yamakawa. 2006. Diallel analysis of plant and ear heights in tropical maize (Zea mays L.). J. Fac. Agric. Kyushu Univ. 51(2): 233–238.

Kamara, M.M., I.S. El-Degwy, and H. Koyama. 2014. Estimation combining ability of some maize inbred lines using line × tester mating design under two nitrogen levels. Aust. J. Crop Sci. 8(9): 1336–1342.

Kempthorne, O. (1957). An introduction to genetic statistics. Wiley.

Legesse, B.W., K. V. Pixley, and A.M. Botha. 2009. Combining ability and heterotic grouping of highland transition maize inbred lines. Maydica 54(1): 1–9.

Mosa, H.E., and A.A. Motawei. 2005. Combining ability of resistance to late wilt disease and grain yield and their relationships under artificial and natural infections in maize. J. Agric. Sci 30 (2)(Mansoura Univ): 731–742.

Murtadha, M.A., O.J. Ariyo, and S.S. Alghamdi. 2018. Analysis of combining ability over environments in diallel crosses of maize (Zea mays). J. Saudi Soc. Agric. Sci. 17(1): 69–78. doi: 10.1016/j.jssas.2016.01.004.

Premlatha, M., and A. Kalamani. 2010. Heterosis and combining ability studies in maize (Zea mays L.). 5(1): 369–372.

Pswarayi, A., and B.S. Vivek. 2008. Combining ability amongst CIMMYT's early maturing maize (Zea mays L.) germplasm under stress and non-stress conditions and identification of testers. Euphytica 162(3): 353–362. doi: 10.1007/s10681-007-9525-0.

Rahman, H., A. Ali, Z. Shah, M. Iqbal, M. Noor, et al. 2013. Line x tester analysis for grain yield and yield related traits in maize variety Sarhad-White. Pakistan J. Bot. 45(SPL.ISS): 383–387.

Rashid, M., A.A. Cheema, and M. Ashraf. 2007. Line X tester analysis in basmati rice. Pakistan J. Bot. 39(6): 2035–2042.

Rodriguez et al., 2015. The Astropy project: Building an open-science project and sta tus of the v5.0 core package. *The astronomical Journal,* 156(3), p. 123.

Seyoum, A., D. Wegary, and S. Alamerew. 2016. Test cross performance and combining ability of elite highland maize (Zea Mays L.) Inbred Lines at Jimma, South West Ethiopia. Int. J. Trend Res. Dev. 3(2): 13–26. internal-pdf://2.13.217.53/Amare_et_al_2016_Test cross performance and co.pdf.

Singh, R.., and B.D. Chaudhary. 1985. Biometrical Method in Quantitative Genetics Analysis. Am. J. Plant Sci. (Kalyani Publishers, New Delhi.).

Sprague, G.F., and L.A. Tatum. 1942. General vs. Specific Combining Ability in Single Crosses of Corn. Am. Soc. Agron.

Sundararajan, R., and P. Senthil Kumar. 2011. Studies on combining ability through Line X Tester analysis in maize (Zea mays L.). Plant Arch. 11(1): 75–77.

Talukder, M. Z. A., A.N.M.S. Karim, S. Ahmed, and M. Amiruzzaman. 2016. Combining ability and Heterosis in Yield and its Components Traits in Maize (Zea mays L.). J. Agril. Res 41(3): 565–577.

Tesfaye, D., D. Abakemal, and E. Habte. 2019. Combining Ability of Highland Adapted Double Haploid Maize Inbred Lines using Line X Tester Mating Design. East African J. Sci. 13(2): 121–134. doi: 10.4314/eajsci.v13i2.

Uddin, M.S., F. Khatun, S. Ahmed, M.R. Ali, and S.A. Bagum. 2006. Heterosis and combining ability in corn (Zea mays L.). Bangladesh J. Bot. 35(2): 109–116.

Zhang, X., L. Lv, C. Lv, B. Guo, and R. Xu. 2015. Combining ability of different agronomic traits and yield components in hybrid barley. PLoS One 10(6): 1–9. doi: 10.1371/journal.pone.0126828.

CHAPTER 4: Genotype by Environment Interaction Analysis of Grain Yield of Early to Medium Maturing Maize Hybrids across Non-Drought and Drought Environments

Abstract

The grain yield of 122 testcross maize hybrids and eight check hybrids were evaluated under five different environments over three locations and three seasons in KwaZulu-Natal province of South Africa to explore genotype-by-environment interaction (GEI) patterns and identify broadly and specifically adapted hybrids. Analysis of variance of the yield data across all the environments showed huge environmental, genotypic and GEI effects, with the environment contributing the largest proportion of the variation followed by genotype and lastly GEI. The additive main and multiplicative interaction effects (AMMI), and the genotype and genotype-by-environment interaction (GGE) methods were employed on selected 62 entries to visualize the GEI patterns. Among the selected hybrids, the AMMI revealed two significant interaction principal component axes (IPCA1 and IPCA2) contributed 50.32% and 20.84%, respectively, to the total GEI variation. The AMMI1 revealed that hybrid n62 (MAK1-122 x CML545) was specifically adapted to drought conditions whereas hybrids n21 (CKDHL0467 x CML312) and n98 (CZL1380 x CML547) were broadly adapted. The identified two high yielding and broadly adapted experimental hybrids were superior to the best check CK3 (WE3127) across all environments. Hybrids n80 (CML569 x CML566) and n95 (CKDHL0467 x CML547) were specifically adapted to irrigated conditions. The GGE-biplots had two principal components, PC1 and PC2, which together explained 69.87% of variation due to genotype and GEI. The GGE-biplots showed similar GEI patterns as AMMI, with the same hybrids identified as broadly and specifically adapted. The identified hybrids could be assessed further in multi-environmental and multiple stress trials to confirm their suitability under high and low input production systems in South Africa and the sub-region before release to farmers.

Key words: Adaptability, AMMI biplot, GGE biplot, GEI signal, interaction principal component axis, Stability, which-won-where

4.1 Introduction

Genotype by environment interaction (GEI) is important to plant breeders in development of crop cultivars. GEI is defined as variability among the phenotypic value and the value expected from corresponding genotypic and environmental values (Kang et al., 2004). GEI is the difference due to the interaction effects of the genotypes and the environments (Dickerson, 1962). Breeders/agronomists normally test various groups of genotypes in various environments, which implies that GEI is to be expected. Singh et al., (1999a) reported that GEI is vital only if genotypes switch ranks from one environment to a different. The genotype by environment interaction can often be grouped into two broad classifications: crossover and non-crossover interactions. The crossover interaction being the one where variety ranks change from one environment to a different. Cross-over interaction, is shown graphically by lines for genotypes intersecting (Kang, 1988).

In-plant breeding, the crossover interaction is more significant than the non-crossover interaction (Kang, 1997). When new genotypes are developed and tested across locations and/or seasons, the relative ranking of the entries for quantitative traits is rarely the same in each environment. Since the presence of crossover interaction has strong inference for breeding for specific adaptation, it is vital to access the frequency of crossover interactions (Singh et al., 1999a). Non-crossover, on the other hand, refers to genotypes that are genetically heterogeneous but the test environments are more or less homogeneous, or genotypes are genetically homogeneous but environments are heterogeneous (Kang, 1998).

The importance of GEI can be seen from the respective contributions of new varieties and enhanced management to yield increases from direct comparison of yields of old and new varieties in a single trial (Cargnin et al., 2009; Hongyu et al., 2014). A large GEI could mean that the establishment of two or more full-fledged breeding stations in a region, instead of one, is

necessary, consequently requiring increased input of resources (Kang, 1998). The useful information that determines the adaptation and stability of maize hybrids is obtained by the evaluation of the genotypic performance of these hybrid candidates in various environments. Kang (1988) revealed that selection for enhancing yield only may not always be appropriate when genotype by the environment is significant (Kang, 1998).

Most significant agronomic and economical attributes like grain yield, are quantitative and regularly show GEI (Fan et al., 2007). There have been countless attempts to investigate GEI for registered cultivars of hybrids maize under various environments. Epinat-Le Signor et al. (2001) reported 132 maize hybrids that were early maturing and were evaluated for grain yield in 229 environments over 12 years. They revealed that; on the tested environments, the early maturity of hybrids, water balance around flowering and average temperature from the stage of 12 leaves to the end of grain filling section were the factors of GEI respectively. Moreover, Kang et al., (1991) suggest that diverse maize hybrid varieties evaluated in their study were most likely influenced by distinctive fertility or cultural practices than by weather determinants. Additionally, they concluded that none of the hybrids were influenced by GEI for grain yield (Tonk et al., 2011). Oliveira et al. (2003) showed that the majority of maize hybrids they evaluated displayed minimal GEI, while the single cross presented considerable average yield and therefore the double cross hybrids demonstrated essential yield stability in ten environments in central Brazil (Tonk et al., 2011).

One of the foremost critical problems in plant breeding is to completely analyse GEI since it depends on knowledge from experiments evaluated in many locations and different seasons (Rodríguez et al., 1989) In most trials, GEI is observed only after statistically analysis. Genotype by environment interaction that changes the grain yield ranking of genotypes in different locations makes it difficult to choose superior genotypes (Miah et al., 2016). The classification of GEI is after statistical modelling. Models are often linear formulations and multiplication

formulations, for instance, additive main effect and multiplication interaction (AMMI), or nonparametric methods (Mohammed, 2020). The objective of the study was to explore genotype-by-environment interaction (GEI) patterns of the developed crosses and identify broadly and specifically adapted entries, with the intention of developing early to medium maturing hybrids for South Africa and the sub region.

4.2. Material and Methods

4.2.1. Generation of hybrids

Using early and medium maturity germplasm lines, most of which were sourced from the International Maize and Wheat Improvement Center, Southern Africa station (CIMMYT-Harare), a line by tester mating design was used for hybridizations to develop testcross hybrids. Hybridization yielded 122 testcross hybrids. The testcross hybrids, together with eight standard check hybrids, seven from the Water Efficient Maize for Africa (WEMA) project, and one from Capstone Seeds Ltd (Table 4.1), were evaluated in multi-environmental trials.

Table 4. 1. Testcross hybrids and hybrid checks evaluated across three warm South African environments over two seasons.

Hybrid code	Pedigree	Hybrid code	Pedigree	Hybrid code	Pedigree
n1	A1220-4CYL x CML442	n45	CML440 x CML539	n89	CML550 x CML547
n2	CML550 x CML442	n46	CKDHL0467 x CML539	n90	CML568 x CML547
n3	CML568 x CML442	n47	CML540 x CML539	n91	CML571 x CML547
n4	CML571 x CML442	n48	CML572x CML539	n92	CKDHL0378 x CML547
n5	CKDHL0378 x CML442	n49	CZL1380 x CML539	n93	CZL0919 x CML547
n6	CML569 x CML442	n50	MAK1-122 x CML539	n94	CML440 x CML547
n7	CZL0919 x CML442	n51	A1220-4CYL x CML545	n95	CKDHL0467 x CML547
n8	CML440 x CML442	n52	CML550 x CML545	n96	CML540 x CML547
n9	CKDHL0467 x CML442	n53	CML568 x CML545	n97	CML572 x CML547
n10	CML540 x CML442	n54	CML571 x CML545	n98	CZL1380 x CML547
n11	CML572x CML442	n55	CKDHL0378 x CML545	n99	MAK1-122 x CML547
n12	CZL1380 x CML442	n56	CZL0919 x CML545	n100	CML550 x CML395
n13	MAK1-122 x CML442	n57	CML440 x CML545	n101	CML568 x CML395
n14	CML550 x CML312	n58	CKDHL0467 x CML545	n102	CML571 x CML395

Hybrid code	Pedigree	Hybrid code	Pedigree	Hybrid code	Pedigree
n15	CML568 x CML312	n59	CML540 x CML545	n103	CKDHL0378 x CML395
n16	CML571 x CML312	n60	CML572x CML545	n104	CML569 x CML395
n17	CKDHL0378 x CML312	n61	CZL1380 x CML545	n105	CZL0919 x CML395
n18	CML569 x CML312	n62	MAK1-122 x CML545	n106	CML440 x CML395
n19	CZL0919 x CML312	n63	A1220-4CYL x CML444	n107	CKDHL0467 x CML395
n20	CML440 x CML312	n64	CML550 x CML444	n108	CML540 x CML395
n21	CKDHL0467 x CML312	n65	CML568 x CML444	n109	CML572x CML395
n22	CML540 x CML312	n66	CML571 x CML444	n110	CZL1380 x CML395
n23	CML572x CML312	n67	CKDHL0378 x CML444	n111	MAK1-122 x CML395
n24	CZL1380 x CML312	n68	CZL0919 x CML444	n112	CML550 x K64R
n25	MAK1-122 x CML312	n69	CML440 x CML444	n113	CML568 x K64R
n26	CML550 x CML537	n70	CKDHL0467 x CML444	n114	CML571 x K64R
n27	CML568 x CML537	n71	CML540 x CML444	n115	CKDHL0378 x K64R
n28	CML571 x CML537	n72	CML572x CML444	n116	CZL0919 x K64R
n29	CKDHL0378 x CML537	n73	CZL1380 x CML444	n117	CML440 x K64R
n30	CML569 x CML537	n74	MAK1-122 x CML444	n118	CKDHL0467 x K64R
n31	CZL0919 x CML537	n75	A1220-4CYL x CML566	n119	CML540 x K64R
n32	CML440 x CML537	n76	CML550 x CML566	n120	CML572x K64R
n33	CKDHL0467 x CML537	n77	CML568 x CML566	n121	CZL1380 x K64R
n34	CML540 x CML537	n78	CML571 x CML566	n122	MAK1-122 x K64R
n35	CML572x CML537	n79	CKDHL0378 x CML566		
n36	CZL1380 x CML537	n80	CML569 x CML566	*Checks*	
n37	MAK1-122 x CML537	n81	CZL0919 x CML566	CK1	WE5323A
n38	A1220-4CYL x CML539	n82	CML440 x CML566	CK2	WE3128
n39	CML550 x CML539	n83	CKDHL0467 x CML566	CK3	WE3127
n40	CML568 x CML539	n84	CML540 x CML566	CK4	WE4145
n41	CML571 x CML539	n85	CML572x CML566	CK5	WE4308
n42	CKDHL0378 x CML539	n86	CZL1380 x CML566	CK6	WE5321
n43	CML569 x CML539	n87	MAK1-122 x CML566	CK7	WE5323B
n44	CZL0919 x CML539	n88	A1220-4CYL x CML547	CK8	CAP9001

4.2.2. Trial environments, design, and cultural practices

The evaluation sites were in warm non-drought rain-fed, warm non-drought well-watered, and drought zones of KwaZulu-Natal (KZN). Ukulinga research farm and Cedara Research station are both located in uMgungundlovu district and Makhathini research station is in Mkhanyakude district in KZN. All location sites are among the primary maize testing sites in KZN. Evaluation environment was considered as site and season combination. The information about location coordinates, planting date, and management are presented in Table 4.2. The fields at

the trial sites were disc ploughed and harrowed to a fine seedbed. The experimental units were marked and were 1 row, 5 m long plots. The distance separating the rows was 0.8 m. Within each row, planting stations (holes) were holed, and these were spaced 0.3 m apart. The 122 testcross hybrids and eight standard check hybrids were planted in a 13x10 alpha lattice design with two replications in each evaluation environment.

Fertilizers were applied at a rate of 70 kg/ha N, 40 kg/ha P_2O_5 and 30 kg/ha K_2O. One-fifth of the required N and all of P_2O_5 and K_2O were applied at planting, and the remaining N was applied in three splits during the growing period at 28, 42 and 55 days after planting. A herbicide, Dual® was applied pre-emergence, and post emergence weed control was by application of Gramoxone® supplemented by hand weeding. Insecticide Karate® was applied pre-emergence to control cutworms. Stalk borer control was by application of Coragen®.

Table 4. 2. Description of the test environments used in the study

Environment code	Location	Latitude	Longitude	Altitude (m ASL)	Mean Seasonal rainfall (mm)	Agroecological zone	Planting date	Management
Ukul2018-19	Ukulinga Research Farm	29º 40'S	30º 24'E	800	750	Warm non-drought watered	6 December 2018	irrigated
Ukul2019-20	Ukulinga Research Farm	29º 40'S	30º 24'E	800	750	Warm non-drought watered	18 December 2019	irrigated
Ced2018-19	Cedara Research Station	29º 32'S	30º 16'E	1400	900	Warm non-drought rainfed	13 December 2018	Rain-fed
Ced2019-20	Cedara Research Station	29º 32'S	30º 16'E	1400	900	Warm non-drought rainfed	20 December 2019	Rain-fed
Mak2019	Makhathini Research Farm	27º 23'S	32º 10'E	450	635	Drought region	12 April 2019	Managed drought

4.2.3. Performance analysis across environments

The following traits were recorded to assess the performance of each hybrid in each of the evaluation environments.

i. **Field weight (FW):** This refers to the total weight of ears harvested per plot in kilograms (kg).

ii. **Grain moisture content (MC):** This is grain moisture expressed as a percentage (%). This was recorded just after harvesting using a Dole® E.T.N model 500 grain moisture tester.

iii. **Ten ears weight**: Ten ears were randomly selected, and their weight was recorded in kg.

iv. **Ten ears grain weight**: The ten randomly selected ears in (iii) were shelled and grain weight was recorded in kg.

v. **Shelling % (SHL):** this was determined as in Equation 4.1.

$$Shelling\ \ (\%) = \frac{Ten\ ears\ grain\ weight\ (kg) * 100}{Ten\ ears\ \ weight\ (kg)} \qquad \text{Equation 4.1}$$

vi. Grain yield (t ha^{-1}): The components FW, MC, and SHL were input into Equation 4.2 to get an estimate of grain yield in tons per hectare. The grain yield was adjusted to 12.5% moisture content.

$$Grain\ \ yield\ (t / ha) = \frac{FW\ (kg) * 10000\ (m^2) * (100 - MC) * SHL}{1000\ (kg) *\ plot\ area\ (m^2) * (100 - 12.5) * 100} \qquad \text{Equation 4.2}$$

The performance analysis included analysis of variance, and means for individual environments and ANOVA and means across environments. The GLM procedure in SAS version 9.4 **(**SAS Institute Inc., 2018**)** was employed, and a Tukey post hoc test was used for mean separation at 5% probability level. For individual environment data, the linear model (Equation 4.3) was used for the analysis of variance.

$$Y_{ijk} = \mu + G_i + R_j + B_k(R_j) + \varepsilon_{ijk} \qquad \text{Equation 4.3}$$

Where,

Y_{ijk} =*ijk*th observation

μ = the mean effect

G_i = the *i*th genotype effect

R_j = the effect of replication *j*

$B_k(R_j)$ = the effect of the *k*th incomplete block within replication *j*

ε_{ijk} = random error term

The linear model that was employed for analysis of variance across environments is presented in Equation 4.4. A bar plot was made to graphically present the mean grain yields for each environment using R statistical programming software (R Core Team, 2019). This analysis detected the significance of genotype-by-environment interaction (GEI) before proceeding with the additive main effects and multiplicative interaction (AMMI) and genotype plus genotype-by-environment interaction (GGE) biplot analyses. Only 62 hybrids, inclusive of all the checks, were identified based on grain yield under drought and across non-drought environments, for further analyses. Some parameters were estimated from results of ANOVA

of the selected 62 entries as suggested by Gauch (2013) and these include GEI noise (GEI_n), which is the product of error mean square and the degrees of freedom for GEI, GEI signal (GEI_s) which is obtained by subtracting GEI_n from the GEI sum of squares. A positive value of GEIs which is comparable to the sum of squares due to genotype would mean that GEI is not concealed in noise and that results of AMMI analysis would be reliable.

$$Y_{ijkl} = \mu + G_i + E_j + (GE)_{ij} + R_k E_j + B_l(R_k E_j) + \varepsilon_{ijkl} \qquad \text{Equation 4.4}$$

Where,

Y_{ijkl} = *ijkl*th observation

μ = the mean effect

G_i = the *i*th genotype effect

E_j = the *j*th environment effect

$(GE)_{ij}$ = the interaction effect of genotype *i* and environment *j*

$R_k E_j$ = the effect of replication *k* within environment *j*

$B_l(R_k E_j)$ = the effect of the *l*th in a complete block within replication *k* and environment *j*

ε_{ijkl} = random error term

4.2.4. Genotype by environment interaction analysis by AMMI method

The examination of GEI using AMMI followed the model that was given by Gauch (1988) and Gauch (1992) and is as presented (Equation 4.5), using GENSTAT statistical software, and procedures described by Payne et al. (2015). AMMI1 biplot was constructed by plotting

IPCA1 scores of genotypes and environments against their main effects. AMMI2 biplot was constructed by plotting IPCA2 scores of genotypes and environments against IPCA1 scores of the same. Only 62 hybrids inclusive of checks, most of which performed the best under drought, and few which were top performers across non-drought environments were assessed, to avoid cluttering in biplots.

$$Y_{ijk} = \mu + \alpha_i + \beta_j + \sum_n \lambda_n \gamma_{in} \delta_{jn} + \rho_{ij} + \varepsilon_{ijk}$$

$$Y_{ijk} = \mu + \alpha_i + \beta_j + \sum_n \lambda_n \gamma_{in} \delta_{jn} + \rho_{ij} + \varepsilon_{ijk} \qquad \text{Equation 4.5}$$

Y_{ijk} = the of i^{th} genotype in j^{th} environment and k^{th} replication

μ = the general mean

α_i = the effect of i^{th} genotype

β_j = the effect of j^{th} environment

λ_n = the eigenvalue of the n^{th} interaction principal component (IPCA)

γ_{in} = the IPCA score of genotypes *i* for the n^{th} IPCA

δ_{jn} = the IPCA score of environment *j* for the n^{th} IPCA

ρ_{ij} = the residual

n = the number of IPCAs retained in the model

ε_{ijk} = random error effect

4.2.5. Exploration of genotype by environment interaction by GGE-biplot method

The GGE-biplot model used was as given by Yan (2002), and is as presented (Equation 4.6). The procedures given by Payne et al. (2015) were followed in constructing biplots in GENSTAT. The “which won where” graph was plotted to visualize the performance of genotypes across the environments and identify the best genotypes in particular environments. Also, a comparison plot was made to visualize the comparative performance of the genotypes concerning grain yield and stability across the environments. Again, to avoid over-crowding in biplots, only 62 hybrids inclusive of checks, most of which performed the best under drought, and few which were top performers across non-drought environments were assessed.

$$Y_{ij} = \mu + \beta_j + \sum_{l=1}^{k} \lambda_l \xi_{il} \eta_{lj} + \varepsilon_{ij} \qquad \text{Equation 4.6}$$

Y_{ij} = the mean of i^{th} genotype in j^{th} environment

μ = the general mean

β_j = the effect of j^{th} environment

λ_l = the singular value of the l^{th} principal component (PC)

ξ_{il} = the eigenvalue of genotype i for the n^{th} PC

η_{lj} = the eigenvalue of environment j for the n^{th} PC

ε_{ij} = random error effect

4.3 Results

4.3.1. Performance analysis across environments

The analysis of variance across all environments and across non-drought environments (Table 4.3) partitioned variation among the hybrids into main effects of environments ($p < 0.001$) and genotypes ($p < 0.001$), and genotype-by-environment interaction (GEI) ($p < 0.001$), all which were highly significant. Across all the environments, the contributions to total hybrid variation were 27.67% due to the environment effect, 19.20% due to the genotype effect, and 22.06 due to the GEI effect. Across non-drought environments, the genotype effect contributed to the highest proportion of variability (23.50%), followed by the environment effect (21.93%), and GEI effect (16.75%). Under drought, there were significant differences among genotypes (Table 4.3). The grain yield means in each of the environments are shown in Figure 4.1. The hybrid means under drought, across non-drought, and across all environments are presented in Table 4.4.

Table 4. 3. Analysis of variance of grain yield of testcross hybrids and checks across all environments, non-drought and under drought environments.

Across All Environments						
Source of variation	DF	SS	MS	F value	Pr > F	% of Total
Environments (E)	4	2578.42	644.61	240.74	<.0001	27.67
Replications (R) in E in	5	254.07	50.81	18.98	<.0001	
Blocks (B) in (E x R)	120	635.86	5.30	1.98	<.0001	
Genotypes (G)	129	1789.64	13.87	5.18	<.0001	19.20
G x E	516	2055.96	3.98	1.49	<.0001	22.06
Error	525	1405.74	2.68			15.08
Total	1299	9319.21				
Mean	7.33					
CV	22.34					
R^2	84.92%					
Across Non-Drought Environments						
Source of variation	DF	SS	MS	F value	Pr > F	% of Total
E	3	1732.98	577.66	183.21	<.0001	21.93
R in E	4	251.64	62.91	19.95	<.0001	
B in (E x R)	96	525.35	5.47	1.74	0.0001	

G	129	1857.10	14.40	4.57	<.0001	23.50
G x E	387	1697.39	4.39	1.39	0.0005	16.75
Error	420	1324.30	3.15			16.76
Total	1039	7903.98				
Mean	7.73					
CV	22.97					
R^2	83.25%					
Drought						
Source of variation	DF	SS	MS	F value	Pr > F	% of Total
R	1	2.43	2.42831	3.13	0.0797	
B in (R)	24	110.52	4.60478	5.94	<.0001	
G	129	290.67	2.25324	2.9	<.0001	50.86
Error	105	81.45	0.7756713			14.25
Total	259	571.45				
Mean	5.71					
CV	15.41					
R^2	85.75 %					

Note: DF = Degrees of Freedom, SS = Sum of Squares, MS = Mean Squares, *CV* = Coefficient of Variation (%), R^2 = R-Square

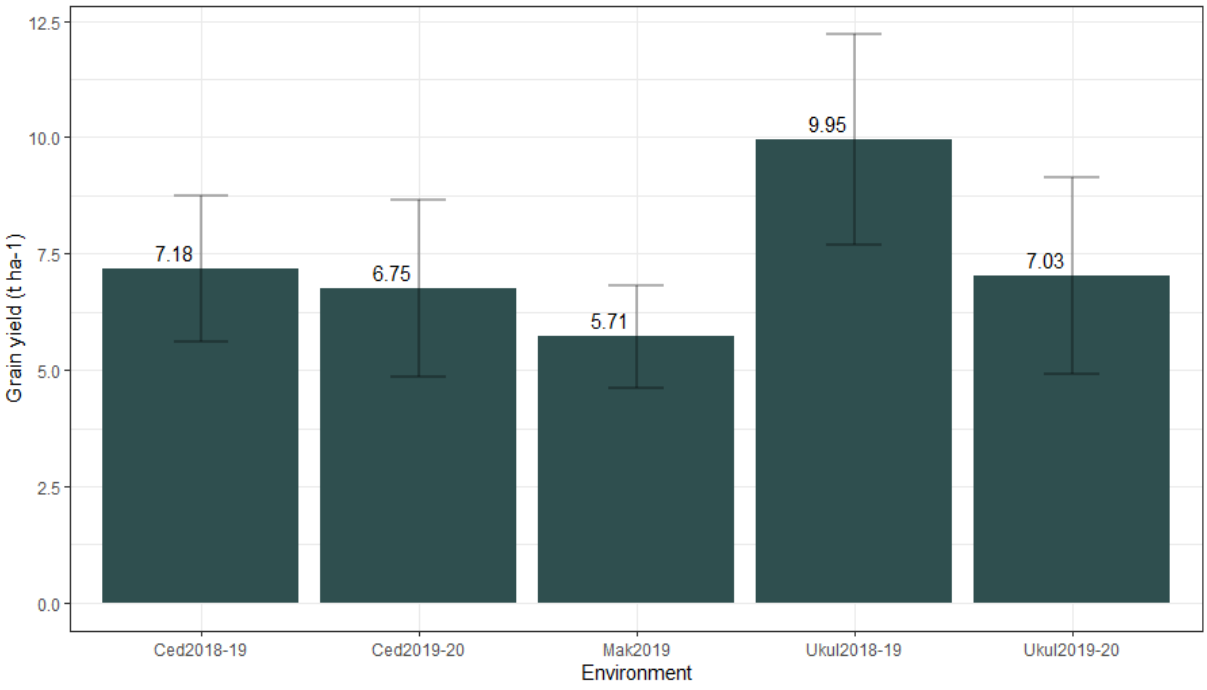

Figure 4. 1. Mean grain yield in each evaluation environment

Table 4. 4. Mean grain yield (t ha^{-1}) of selected testcross hybrids and checks under drought, non-drought, and across all environments

Genotype	Mean/ rank under drought		Mean across non-drought	Mean across all environments	Genotype	Mean/ rank under drought		Mean across non-drought	Mean across all environments
n110	8.86a	1	8.64a-g	8.69a-f	**CK4**	6.47a-	32	8.56a-	8.14a-h
n12	8.15ab	2	7.54a-k	7.66a-k	**n90**	6.42a-	33	9.70a-	9.04a-e
n44	7.80a-c	3	7.26a-l	7.37a-k	**n71**	6.41a-	34	7.40a-l	7.20a-k
n96	7.67a-	4	9.00a-f	8.74a-f	**n31**	6.35a-	35	6.82a-l	6.73a-m
n107	7.41a-	5	9.82a-d	9.33a-d	**n41**	6.33a-	36	7.28a-l	7.09a-k
n76	7.39a-	6	8.22a-j	8.05a-i	**n7**	6.32a-	37	7.44a-l	7.21a-k
n4	7.38a-	7	7.47a-l	7.45a-k	**n63**	6.31a-	38	8.38a-i	7.96a-i
CK1	7.21a-e	8	7.89a-k	7.75a-j	**n58**	6.30a-	39	8.80a-i	8.30a-h
n61	6.94a-e	9	8.37a-j	8.09a-i	**n120**	6.30a-	40	6.55b-l	6.50a-m
n50	6.89a-f	10	7.52a-k	7.39a-k	**n64**	6.28a-	41	6.24b-l	6.25a-m
n59	6.85a-	11	7.40a-l	7.29a-k	**n18**	6.21a-	42	9.17a-	8.58a-f
n21	6.84a-	12	10.11a-c	9.45a-c	**CK5**	6.21a-	43	7.82a-	7.50a-k
n55	6.82a-	13	8.89a-g	8.48a-g	**n49**	6.18a-	44	8.52a-i	8.05a-i
n72	6.82a-	14	8.96a-g	8.53a-f	**n115**	6.18a-	45	7.84a-	7.51a-k
n33	6.79a-	15	8.24a-h	7.95a-i	**n75**	6.16a-	46	8.78a-i	8.25a-h
n20	6.79a-	16	6.43a-l	6.50a-m	**n56**	6.13a-	47	7.16a-l	6.95a-m
CK6	6.75a-	17	8.26a-h	7.96a-i	**n19**	6.11a-	48	8.76a-	8.23a-h
n48	6.75a-	18	7.72a-k	7.53a-l	**n6**	6.08a-	49	9.57a-	8.87a-f
n54	6.72a-	19	6.40a-l	6.46a-m	**n37**	6.06a-	50	7.18a-l	6.95a-m
CK3	6.70a-	20	9.80a-d	9.18a-i	**n68**	6.05a-	51	7.89a-	7.52a-k
n81	6.60a-	21	8.34a-j	7.99a-i	**n86**	6.05a-	52	9.88a-	9.12a-e
n98	6.60a-	22	11.04a	10.15a	**n95**	5.93a-	59	11.10a	10.07a
n47	6.60a-	23	7.38a-l	7.23a-k	**n27**	5.90a-	62	9.36a-f	8.67a-f
n26	6.59a-	24	6.90a-l	6.84a-m	**n79**	5.79a-	68	9.88a-	9.06a-e
n40	6.59a-	25	8.01a-k	7.72a-k	**n92**	5.64a-	79	9.25a-f	8.53a-f
n43	6.58a-	26	7.96a-k	7.68a-h	**CK7**	5.59a-	80	7.46a-l	7.09a-k
n5	6.57a-	27	8.68a-h	8.26a-m	**n97**	5.50a-	83	9.95a-	9.06a-e
n62	6.56a-	28	6.99a-l	6.91a-m	**n80**	5.48a-	84	10.12a	9.19a-d
n1	6.55a-	28	6.10b-l	6.19a-h	**n23**	5.32a-	88	9.36a-f	8.55a-f
n109	6.52a-	30	8.70a-h	8.26a-h	**CK8**	3.48c-	125	8.75a-i	7.70a-k
n11	6.52a-	31	7.46a-l	7.27a-k	**CK2**	2.37h	130	7.53a-	6.50a-m
Mean						5.71		7.73	7.31
CV						15.41		22.97	22.34
SEm						0.08		0.16	0.14

Note: Means followed by the same letter(s) are not significantly different

4.3.2. Genotype by environment interaction analysis by AMMI method

The GEI sum of squares among the selected 62 hybrids was markedly noticeable (1387) relative to the GEI_n (817.4). Apart from the significant effects of environment ($p < 0.001$), genotype ($p < 0.001$) and GEI effect ($p < 0.001$), AMMI analysis of variance further partitioned the GEI sum of squares into two significant interaction principal component axes, IPCA1 ($p < 0.001$) and IPCA2 ($p < 0.05$) (Table 4.5). The IPCA1 and IPCA2 contributed 50.32 and 20.84, respectively, to the GEI variation.

Table 4. 5. AMMI analysis of variance for grain yield of 54 testcross hybrids and 8 checks across five environments

Source of variation	DF	SS	MS	F value	F pr	% of Treatment SS	% of GEI SS
Total	619	4434	7.16				
Treatments	309	3309	10.71	3.2	<0.001		
Genotypes	61	599	9.83	2.94	<0.001	18.10	
Environments	4	1323	330.68	16	<0.001	39.98	
Replications in Environments	5	103	20.66	6.17	<0.001		
Genotype x Environment	244	1387	5.68	1.7	<0.001	41.92	
IPCA 1	64	698	10.91	3.26	<0.001		50.32
IPCA 2	62	289	4.66	1.39	0.0373		20.84
Residuals	118	400	3.39	1.01	0.4588		28.84
Error	305	1021	3.35				

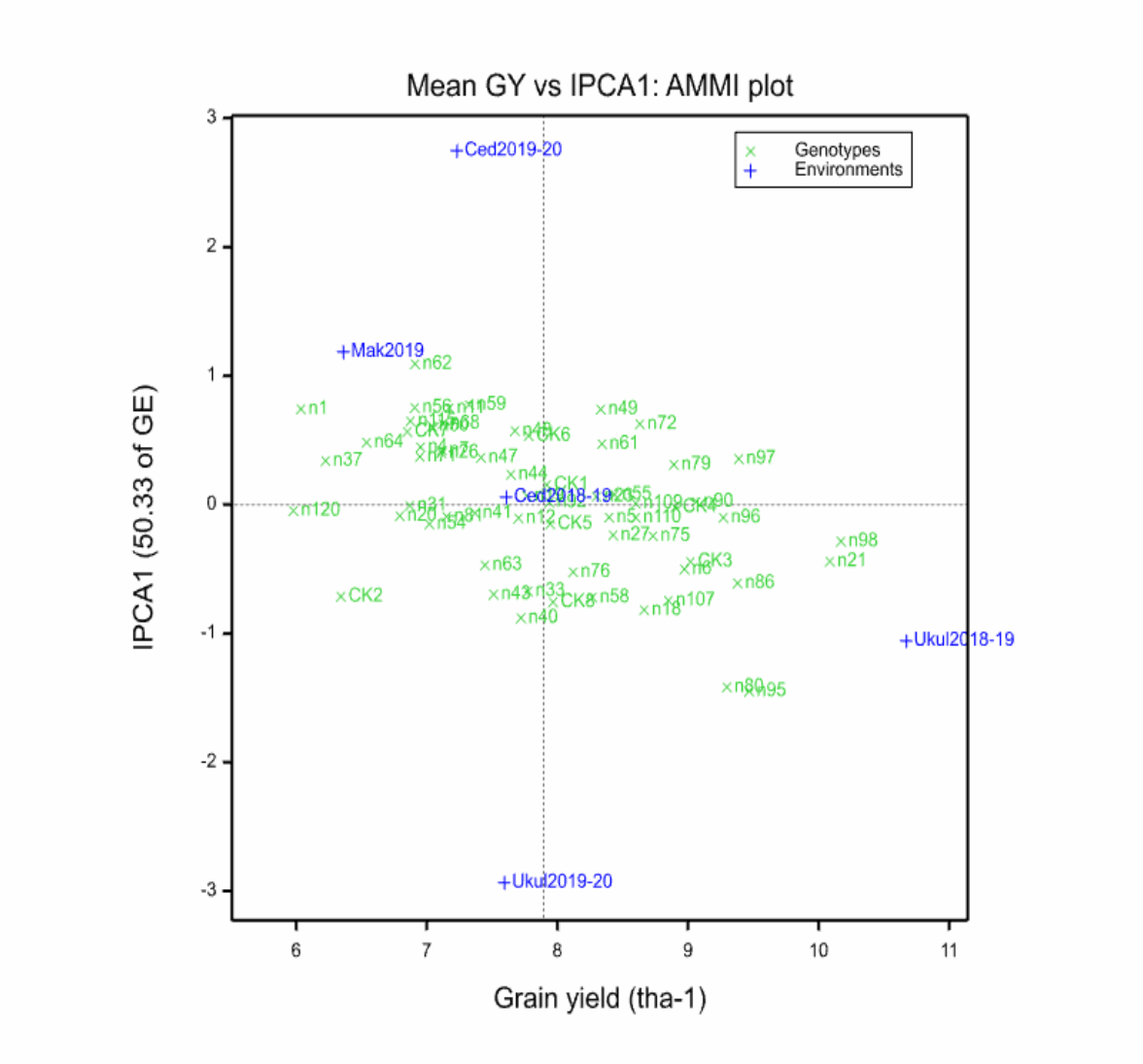

Figure 4. 2. The AMMI1 biplot for grain yield of the best 54 testcross hybrids and eight check hybrids of early-medium maturing high yielding maize hybrids indicating genotypes and five environments plotted against IPCA1 scores.

In the AMMI1 biplot (Figure 4.2), the dotted horizontal line represents an IPCA1 score of zero. Genotypes with a shorter perpendicular projection to this line are stable and those with longer perpendicular projections are relatively unstable. Likewise, environments with shorter perpendicular projections to the dotted horizontal line contributed less to GEI, whereas those with longer projections were relatively more interactive. Thus, the IPCA1 score that is closer to zero, indicates that the genotype is more stable across environments. The higher the IPCA1 score, positive nor negative, the more certainly adapted a genotype is to a particular

environment with a similar sign IPCA1 score. The vertical dotted line represents the grand mean grain yield performance of genotypes across all environments, thus, genotypes and environments to the left side of this line had mean performance below the grand mean, and those to the right side of the line had mean yields above the grand mean. The highest yields were obtained at Ukulinga in the 2018-2019 season (Ukul2018-19) and the lowest yields were realized at Makathini Research Station under drought (Mak2019).

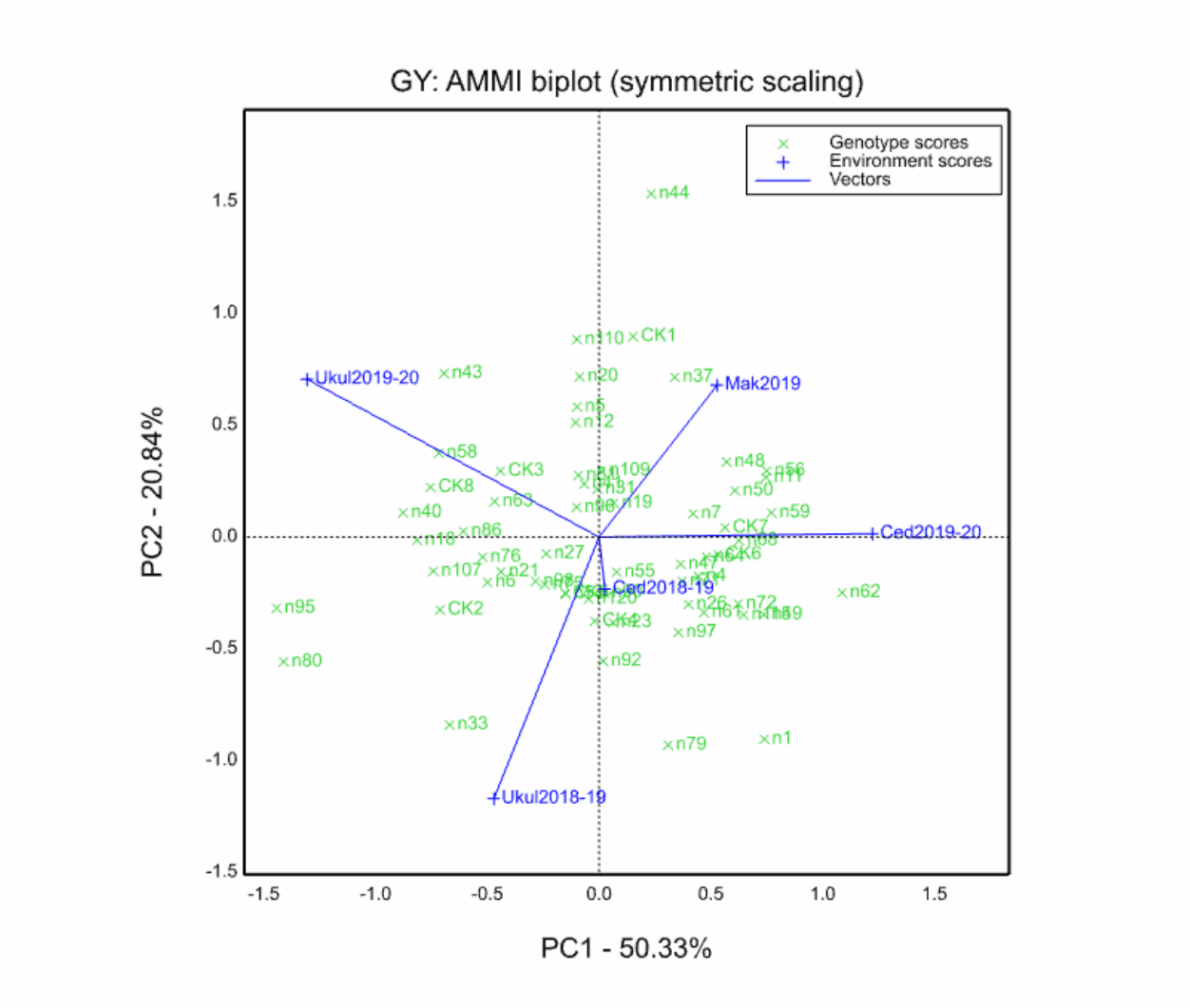

Figure 4. 3. The AMMI2 biplot indicating the relationship between five experimental environments.

The angle between each environment vector provides information on the relationship between the presented environments. There was an obtuse angle between Ukul2019-202

and Ced2019-20, and Ukul2018-19 and Ced2019-20 vectors (Figure 4.3). There was an acute angle between Mak2019 and Ced2019-20; Ced2018-19 and Ced2019-20, and Ukul2018-19 and Ced2018-19 vectors, indicating a positive relationship between these pairs of environments. Environments Ukul2019-20 and Mak2019 were located at right angles in the biplot. Genotypes that are crowded close to the origin are insensitive to environmental changes, whereas those located far away from the origin are sensitive to changes in the environment.

4.3.3. Exploration of Genotype by Environment Interaction by GGE-Biplot method

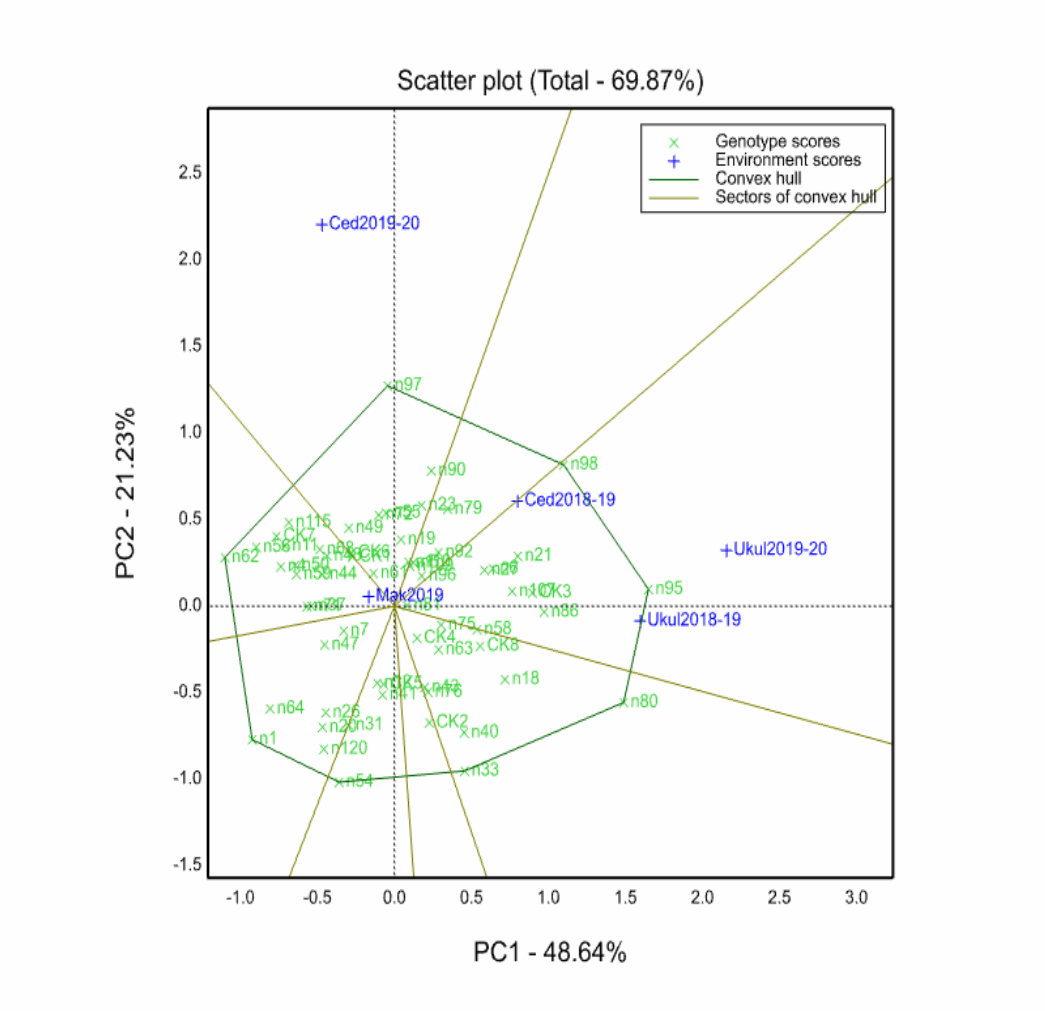

Figure 4. 4. The "which won where" of GGE biplot of the best 54 hybrids and eight hybrid checks of early-medium high yielding maize hybrids evaluated in five environments.

The GGE biplot for 62 genotypes is represented in Figure 4.4, in a polygon view of the genotypes. Respectively, primary (PC1) and secondary (PC2) scores explained 48.64 and 21.23 of the variation and they were significant. Together they explained 69.87% of the primary impact of genotype and GEI effect for grain yield of the selected early-medium maturing maize hybrids assessed in 5 environments in KZN. The “which-won-where” design was shown by the polygon view of the GGE biplot in Figure 4.4. The convex hull of the polygon connects genotype markers found furthest away from the biplot origin in different ways, to such an extent that all genotype markers were contained within the subsequent polygon. Lines that are perpendicular to each side of the polygon and connected to the origin of the biplot divided the biplot into eight sectors. Environments are found in only three sectors together with associated genotypes. There are vertex genotypes in each of the sectors, however, some of these genotypes are in sectors that are not associated with any of the environments.

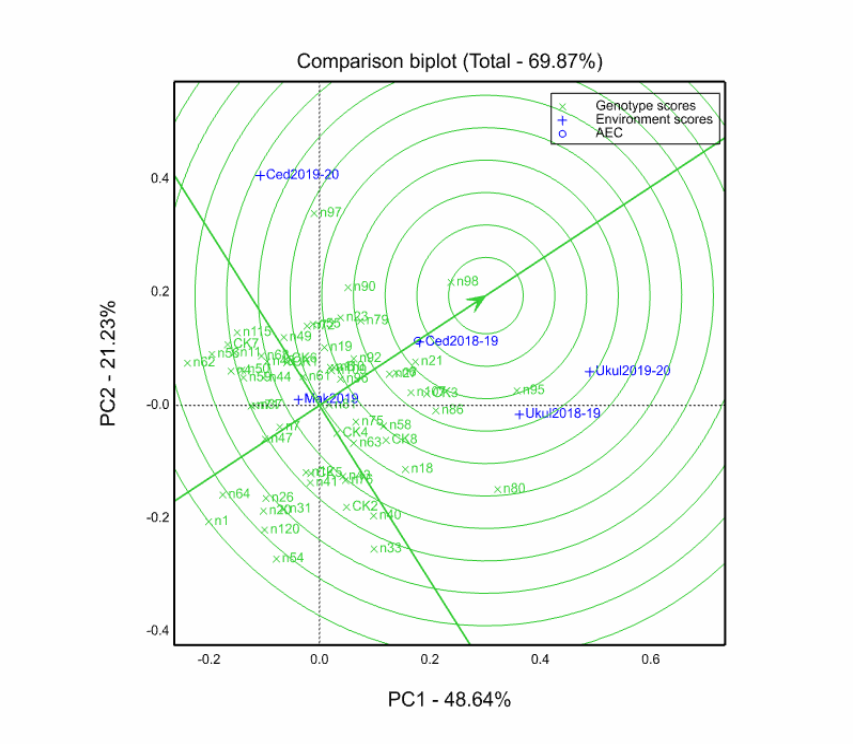

Figure 4. 5. The comparison view of GGE biplot of 62 maize hybrids with the highest yielding and stable genotype based on grain yield and stability for grain yield among five environments.

The comparison biplot (Figure 4.5) represents the ranking of genotypes dependent on both mean grain yield and stability performances of evaluated hybrids and hybrid checks to identify the higher-yielding and most stable genotypes. The line that passes through the biplot origin and the average environment coordinate (AEC, a small circle in the biplot) is called the AEC abscissa. The AEC abscissa is unidirectional and is pointing toward the direction of increasing grain yield. The line that passes through the biplot origin and is perpendicular to the AEC abscissa is the AEC ordinate which indicates the effect of GEI, the stability of genotypes, and separate genotypes that are superior and inferior to the overall mean. Genotypes that perform below the mean are found below the AEC ordinate, and those with mean yields above the grand mean are found above the AEC ordinate.

4.4 Discussion

The highly significant environmental and genotypic effects meant that the environments were diverse and influenced the genotypes differently and that the hybrids differed at the genotypic level. The results of this study showed that the environment contributed the largest proportion to total hybrid variation, followed by GEI and genotypes in that order. The significant genotype effect under drought implies that the hybrids were differentially endowed with genes for drought tolerance, with varying levels of tolerance from highly susceptible to highly tolerant. The hybrids exhibited differential performance across the environments as revealed by the significant GEI effect. Some genotypes were broadly adapted, for example, n98 which was among the best 25 both under drought and across non-drought environments, whereas some genotypes were specifically adapted, and this included n110 (the best under drought) and n80 (one of the best under non-drought). A broadly adapted hybrid that has a good performance both under drought and across non-drought conditions is highly desirable because it would be advantageous to farmers in a drought season and a good season. Seven experimental hybrids performed better than the best check hybrid (CK1) under drought and these could be recommended for release especially if they also perform

well under non-drought conditions, after further evaluations in multi-environmental trials across locations and over seasons in South Africa. The check CK3 performed fairly well both under drought and across non-drought conditions, thus exhibiting broad adaptation. The GEI exhibited by the hybrids in this study was the qualitative or cross-over type since there was a change in ranking (Singh, Ceccarelli and Grando, 1999) in respect of grain yield performance from drought conditions to the non-drought conditions. The GEI signal (GEI_s) among the selected 62 hybrids that were assessed using AMMI and GGE-biplots was significant (569.6) and was not masked by GEI noise (GEI_n) meaning that the data was dependable (Gauch, 2013), and this justified proceeding with further analysis using AMMI.

AMMI1 analysis revealed that n90 had an IPCA1 score of zero and n96 close to zero and therefore both can be considered to have a small interaction with the environments and the most stable hybrids (Apala Mafouasson et al., 2018). Hybrids n90 and n96 had grain yield over the grand mean; however, n96 was higher yielding compared to n90, and the differences are not huge. Several hybrids among the 62 represented and selected had grain yield response beneath the grand mean, which include, n12, n44, n41, n47, and n84. The other various hybrids among the selected and represented hybrids had their grain yield response above the grand mean, and to mention a few, n109, n55, n110 n97, and n98. Among the various hybrids that had their grain yield response above the grand mean, n98 had the highest grain yield, followed by n21, n95, n97, and n86. N49, n72, and n61 had grain yield over grand mean with high positive IPCA1 scores. Conversely, n80, and n95 had a negative interaction with the IPCA1. Hybrids n98 and n21 were the highest yielding genotypes; nevertheless, n98 was higher-yielding and more stable than n21. Hybrids n80, n95, n86, and n6 had grain yield over the grand mean and had negative IPCA1 scores, consequently a negative interaction with the environments. The lowest yielding genotypes were n1 and n120, and n1 was less stable compared to n120.

Considering AMMI2 analysis, the obtuse angle between Ukul2019-202 and Ced2019-20, and Ukul2018-19 and Ced2019-20 vectors indicate that there is a negative association between these environments (Kroonenberg, 1995) and their influence on the performance of genotypes was very different. The acute angle between Mak2019 and Ced2019-20; Ced2018-10 and Ced2019-20, and Ukul2018-19 and Ced2018-19 imply a positive association between these pairs of environments, and they had a similar influence on the performance of the hybrids (Kroonenberg, 1995). There is no association between Ukul2019-20 and Mak2019 since there is a right angle between these environment vectors (Kroonenberg, 1995). Hybrid n44, n95, n80 are located far from the original meaning they are sensitive to environmental changes; hence they are specifically adapted. In contrast, hybrids n19, n27, and n98 are located near the origin, which means they are insensitive to environmental interactions; hence they are broadly adopted. Ced2018-19 is located near the origin indicated by a short vector from the origin which implies lowest grain yield variability in this environment, followed by Mak2019, whereas Ukul2018-19 is the furthest environment from the origin which means it exhibited highest grain yield variability and this offered the best chance to select for yield improvement (Yan and Tinker, 2006; Kroonenberg, 1995). The further the environment is from the origin also shows that it had a large interaction effect with at least one genotype (Kroonenberg, 1995).

The GEI patterns displayed by the GGE biplots are dependable since PC1 and PC2 together explained a high proportion (67.87%) of the primary impact of genotype and GEI effect (Kroonenberg, 1995; Yang et al., 2009) for grain yield of the selected early-medium maturing maize hybrids assessed in 5 environments in KZN. The polygon indicated eight vertex hybrids, n1, n54, n33, n80, n95, n98, n97, and n62, and it was divided into eight sectors. Hybrids n98 and n95 appeared in the same sector as, Ukul2018-19, Ukul2019-20, and Ced2018-19, which means these two genotypes are the best performers and high yielding within these environments. Hybrids n97, n90, and n23 appeared in the same sector as Ced2019-20; hence they are the best performers and high yielding within this environment

(Yan and Tinker, 2006; Yang et al., 2009). Hybrids n44, n48, and n68 appeared in the same sector as the Mak2019 environment, which indicates that they are the best performers within this environment (under drought). Hybrids n1 and n54 were vertex genotypes but were not associated with any environment, and according to Yan and Tinker (2006).

Genotypes that are situated closest to the “ideal genotype” (the focal point of the concentric circles) are the highest yielding and most stable, therefore most desirable (Kang et al., 2005). The comparison biplot of GGE recognized n98 as the most superior hybrid genotype since it was found close to the centre or focal point of the concentric circles. This hybrid was followed by n21, n95, and ck3 (Figure 4). Hybrid check (ck3) was the highest yielding hybrid check compared to the other seven hybrid checks. Hybrid n1 was situated far from the vertical axis at the left, and far from the focal point of the concentric circle, consequently, it is the most inferior hybrid in both grain yield and stability.

4.5 Conclusions and Recommendations

Maize farmers in South Africa and the sub-region are located in different agro-ecologies. Additionally, these farmers practice different systems of maize production with some having irrigation facilities and some practicing low input and rain-fed crop agriculture. Not all maize hybrids developed and released for commercialization would be suitable for production in every maize production zone in the country. Even in a province like KwaZulu-Natal the maize production areas significantly differ in terms of growing conditions. Thus, the need for maize breeders to explore the patterns of genotype by environment interaction (GEI) among their candidate hybrids before they commercialize them. This study aimed to explore GEI patterns among early to medium maturity experimental maize hybrids and checks, in the KwaZulu-Natal maize producing province of South Africa. The following conclusions and recommendations were made.

- The evaluation environments under which the maize hybrids were grown were diverse and influenced the performance of hybrids differently.

- The maize hybrids tested differed significantly at the genotypic level as revealed by analysis of variance.
- The genotype by environment interaction signal was high, and this justified further explorations by AMMI and GGE-biplot methods, and results from these analyses were deemed reliable.
- The GEI was of a non-crossover type, and different maize hybrids could be recommended for different environments.
- Hybrid n62 (MAK1-122 x CML545) was identified as adapted to drought conditions; however, this hybrid may not be recommended for release since it was not among the top performers under non-drought environments
- Hybrids n98 (CZL1380 x CML547) and n21 (CKDHL0467 x CML312) were broadly adapted, that is, they performed well across the evaluation environments. These hybrids are advantageous to farmers, especially those practicing low input and rain-fed crop agriculture since these hybrids would yield well in a droughty season and a good season.
- Hybrids n95 (CKDHL0467 x CML547) and n80 (CML569 x CML566) were specifically adapted to irrigated conditions
- Four hybrids could be released for broad and specific environments after further evaluation in multi-environment and multiple stress trials and these are n98 (CZL1380 x CML547) and n21 (CKDHL0467 x CML312) for multiple environments areas, and n80 (CML569 x CML566) and n95 (CKDHL0467 x CML547) for irrigated production.

References

Apala Mafouasson, H.N. Ile, V. Gracen, M.A. Yeboah, G. Ntsomboh-Ntsefong, L.N. Tandzi, et al. 2018. Genotype-by-environment interaction and yield stability of maize single

cross hybrids developed from tropical inbred lines. Agronomy 8(5): 1–17. doi: 10.3390/agronomy8050062.

Cargnin, A., M.A. de Souza, V. Fronza, and C.M. Fogaça. 2009. Genetic and environmental contributions to increased wheat yield in minas gerais, Brazil. Sci. Agric. 66(3): 317–322. doi: 10.1590/s0103-90162009000300006.

Dickerson, G.E. 1962. Implications of genetic-environmental interaction in animal breeding. Anim. Prod. 4(1): 47–63. doi: 10.1017/S0003356100034395.

Epinat-Le Signor, C., S. Dousse, J. Lorgeou, J.B. Denis, R. Bonhomme, et al. 2001. Interpretation of genotype X environment interactions for early maize hybrids over 12 years. Crop Sci. 41(3): 663–669. doi: 10.2135/cropsci2001.413663x.

Fan, X.M., M.S. Kang, H. Chen, Y. Zhang, J. Tan, et al. 2007. Yield stability of maize hybrids evaluated in multi-environment trials in Yunnan, China. Agron. J. 99(1): 220–228. doi: 10.2134/agronj2006.0144.

Gauch, H.G. 1988. Model selection and validation for yield trials with interaction. Crop Sci. 44(3): 705–715.

Gauch, H.G. 1992. Statistical analysis of regional yield trials: AMMI analysis of factorial designs. Crop Sci. pp221

Gauch, H.G. 2013. A simple protocol for AMMI analysis of yield trials. Crop Sci. 53(5): 1860–1869. doi: 10.2135/cropsci2013.04.0241.

Hongyu, K., M. García-Peña, L.B. de Araújo, and C.T. dos Santos Dias. 2014. Statistical analysis of yield trials by AMMI analysis of genotype × environment interaction. Biometrical Lett. 51(2): 89–102. doi: 10.2478/bile-2014-0007.

Kang, M.S. 1997. Using Genotype-by-Environment Interaction for Crop Cultivar Development. Adv. Agron. 62(C): 199–252. doi: 10.1016/S0065-2113(08)60569-6.

Kang, M.S. 1998. Genotype-Environment Interaction : Progress and Prospects 15 Genotype – Environment Interaction : Progress and Prospects.. https://www.researchgate.net/publication/313607384.

Kang, M. 1988. A rank-sum method for selecting high-yielding, stable corn genotypes. Cereal Res. Commun 16(1): 113–115.

Kang, M.S., V.D. Aggarwal, and R.M. Chirwa. 2005. Adaptability and stability of bean cultivars as determined via yield-stability statistic and GGE biplot analysis. J. Crop Improv. 15(1): 97–120. doi: 10.1300/J411v15n01_08.

Kang, M.S., D.P. Gorman, and H.N. Pham. 1991. Application of a stability statistic to international maize yield trials. Theor. Appl. Genet. 81(2): 162–165. doi: 10.1007/BF00215718.

Kang, M.S., V.T. Prabhakaran, and R.B. Mehra. 2004. Genotype-by-Environment Interaction in Crop Improvement. Plant Breed.: 535–572. doi: 10.1007/978-94-007-1040-5_23.

Kroonenberg, P.M. 1995. Introduction to Biplots for G x E Tables Centre for Statistics. : 1–22. http://www.fsw.leidenuniv.nl/~kroonenb/document/biplot.zip.

Miah, M., S. Ahmed, and M. Uddin. 2016. Assessment of yield stability of maize inbred lines in multi-environment trials. Bangladesh J. Sci. Ind. Res. 51(1): 61–68. doi: 10.3329/bjsir.v51i1.27064.

Mohammed, A. 2020. Genotype by environment interaction and yield stability analysis of open pollinated maize varieties using AMMI model in Afar Regional State, Ethiopia. J. Plant Breed. Crop Sci. 12(1): 8–15. doi: 10.5897/jpbcs2019.0839.

Oliveira, J.P., W.N. Moreira Jr., J.B. Duarte, L.J. Chaves, and J.B. Pinheiro. 2003. Genotype-environment interaction in maize hybrids: an application of the AMMI model. Crop. Breed. Appl. Biotechnol. 3(3): 185–192. doi: 10.12702/1984-7033.v03n03a02.

Payne, R.W., P. Brain, R.C. Butler, P.W. Goedhart, P.W. Goedhart, et al. 2015. Genstat Reference Manual (Release 18), Part 3 Procedures.

Rodríguez, E., J.L. Arqués, R. Rodríguez, M. Nuñez, M. Medina, et al. 1989. We are IntechOpen , the world ’ s leading publisher of Open Access books Built by scientists , for scientists TOP 1 %. Intech 32(tourism): 137–144. https://www.intechopen.com/books/advanced-biometric-technologies/liveness-detection-

in-biometrics.

Singh, M., S. Ceccarelli, and S. Grando. 1999a. Genotype x environment interaction of crossover type: Detecting its presence and estimating the crossover point. Theor. Appl. Genet. 99(6): 988–995. doi: 10.1007/s001220051406.

Singh, M., S. Ceccarelli, and S. Grando. 1999b. Genotype x environment interaction of crossover type: Detecting its presence and estimating the crossover point. Theor. Appl. Genet. 99(6): 988–995. doi: 10.1007/s001220051406.

Tonk, F.A., E. Ilker, and M. Tosun. 2011. Evaluation of genotype x environment interactions in maize hybrids using GGE biplot analysis. Crop. Breed. Appl. Biotechnol. 11: 1–9.

Yan, W. 2002. Singular-value partitioning in biplot analysis of multienvironment trial data. Agron. J. 94(5): 990–996. doi: 10.2134/agronj2002.0990.

Yan, W., and N.A. Tinker. 2006. Biplot analysis of multi-environment trial data: Principles and applications. Can. J. Plant Sci. 86(3): 623–645. doi: 10.4141/P05-169.

Yang, R.C., J. Crossa, P.L. Cornelius, and J. Burgueño. 2009. Biplot analysis of genotype × environment interaction: Proceed with caution. Crop Sci. 49(5): 1564–1576. doi: 10.2135/cropsci2008.11.0665.

CHAPTER 5: General overview

5.1 Introduction

Climate change is predicted to cause more variable rainfall and an increase in temperature to above normal in South Africa and sub-region and this is expected to negatively affect maize production (Turpie and Visser, 2015). The development of early to medium maturing maize germplasm, especially those that are high yielding and drought tolerant is one of the sustainable ways to mitigate against climate change and variabilities. Smallholder farmers are expected to be more affected since they are often subjected to moisture stress during periodic drought in the growing season and they cannot afford irrigation (Niang et al., 2015).

Therefore, this research was undertaken with an overall goal to improve the livelihoods of smallholder farmers in the country and the region through breeding of early to medium hybrids of high yielding potential. The germplasm used were inbred lines of early to medium maturity group, and most of them (20 lines) were sourced from the International Maize and Wheat Improvement Center (CIMMYT); in addition, three South African inbred lines were included. Hybridizations were made following the line x tester format among the 23 inbred lines (13 lines and 10 testers) and 122 hybrids were successfully developed. The specific objectives of this study were: (i) to determine combining ability and gene action among germplasm lines for grain yield (GY) and other traits under drought and non-drought conditions using the line x tester mating design (ii) to explore genotype-by-environment interaction (GEI) patterns of the developed crosses and identify broadly and specifically adapted entries, with the intention of developing early to medium maturing hybrids for South Africa and the sub region.

The newly developed testcross hybrids together with eight standard check hybrids were grown in a 13 x 10 alpha lattice design with two replications in five different environments in the Kwa-Zulu Natal maize production region (region 4) of South Africa. The environments

could be grouped into drought (one environment) and non-drought. Data were collected for grain yield and related traits and analyses were made.

5.2. Line x tester of Early to Medium Maize Germplasm across Warm Optimal and Drought Location in South Africa

The line x tester design was used to identify best parents through combining ability analysis for use in developing early to medium maturing hybrids. However, inferences about the prevailing gene action can also be made based on model I (fixed effects) analysis, which means the interpretations apply only for the fixed set of lines used. The inferences drawn from the genetic analyses are as follows:

- The parents differed in general combining ability (GCA) effects for GY and other traits under drought and non-drought conditions.
- Likewise, the crosses varied in specific combining ability (SCA) effects for GY and other traits under the drought and non-drought regimes.
- Line CZL1380 and tester CML539 were good general combiners for GY under drought.
- Lines CML568, CKDHL0378, CKDHL0467, CML672, and CZL1380 and testers CML312 and CML547 had good GCA effect across non-drought regime.
- Two crosses, CML540 x CML547 and CKDHL0467 x CML312 had high SCA values for GY under drought and non-drought regimes.
- The additive type of gene action was predominant for days to anthesis (AD), days to silking (SD), anthesis-silking interval (ASI) plant height (PH), ear position (EPO), ears per plant (EPP), ear aspect (EA), grain texture (GTX), grain moisture (GMH), kernel row number (KRN), and shelling percentage (SHL) under drought, and for AD, SD, ear height (EH), EPO, EPP, EA, GTX, GMH, ear length (EL), kernels per ear row (KER), ear weight (EW), and hundred kernel weight (HKW) across non-drought conditions.
- Non-additive gene action prevailed for EH, EL, ED, KER, EW, HKW, and GY under drought and for ASI, PH, ED, KRN, SHL, and GY across non-drought conditions.

- Baker's predictability ratios were quite high for AD, SD, ASI, PH, EPO, EPP, EA, GTX, GMH, KRN, and SHL under drought condition. Under non-drought conditions, high predictability ratios were realized for AD, SD, EH, EPO, EPP, GMH, EL, KER, EW, and HKW. For these traits and in the respective environmental conditions, the performance of hybrids can be predicted using the GCA of the parents.
- The identified hybrids could be targeted for release as cultivars, and the types of gene action are practically relevant for improvement of early to medium maturing maize germplasm for Southern Africa.

5.3. Genotype by Environment Interaction Analysis of Grain Yield of Early to Medium Maturing Single-Cross Maize Hybrids Across Warm Non-Drought and Drought Environments of KwaZulu-Natal in South Africa

A desirable maize hybrid should be of high yield potential and it can be broadly or specifically adapted. The response of newly developed hybrids to different environments can be used to identify those that are broadly adapted and the specifically adapted ones. This will help the breeder in making recommendations regarding the best hybrid for a particular environment. The following conclusions and recommendations were made from the genotype – by – environment interaction (GEI) analysis of the early to medium maturing Line x Tester developed single-cross hybrids and check hybrids.

- The evaluation environments under which the maize hybrids were grown were diverse and influenced the performance of hybrids differently.
- The maize hybrids tested differed significantly at the genotypic level as revealed by analysis of variance.
- The genotype by environment interaction signal was high, and this justified further explorations by AMMI and GGE-biplot methods, and results from these analyses were deemed reliable.
- The GEI was of a non-crossover type, and different maize hybrids could be recommended for different environments.

- Hybrid n62 (MAK1-122 x CML545) was identified as adapted to drought conditions; however, this hybrid may not be recommended for release since it was not among the top performers under non-drought environments
- Hybrids n98 (CZL1380 x CML547) and n21 (CKDHL0467 x CML312) were broadly adapted, that is, they performed well across the evaluation environments. These hybrids are advantageous to farmers, especially those practicing low input and rain-fed crop agriculture since these hybrids would yield well in a droughty season and a good season. In addition, these hybrids were superior to the best check, WE3127
- Hybrids n95 (CKDHL0467 x CML547) and n80 (CML569 x CML566) were specifically adapted to irrigated conditions. They performed better than the best check, WE3127 across non-drought conditions
- Four hybrids could be released for broad and specific environments after further evaluation in multi-environment and multiple stress trials and these are n98 (CZL1380 x CML547) and n21 (CKDHL0467 x CML312) for multiple environments areas, and n80 (CML569 x CML566) and n95 (CKDHL0467 x CML547) for irrigated production.

References

Niang, I., O.C. Ruppel, M.A. Abdrabo, A. Essel, C. Lennard, et al. 2015. Africa. Clim. Chang. 2014 Impacts, Adapt. Vulnerability Part B Reg. Asp. Work. Gr. II Contrib. to Fifth Assess. Rep. Intergov. Panel Clim. Chang.: 1199–1266. doi: 10.1017/CBO9781107415386.002.

Turpie, J., and M. Visser. 2015. The Impact of Climate Change on South Africa's Rural Areas. Finacial Fisc. Commision: 100–162.