BREEDING FOR DROUGHT TOLERANCE AND HIGH GRAIN YIELD OF SORGHUM (*Sorghum bicolor* [L.] Moench) IN TANZANIA

By

ANDEKELILE SALONI MWAMAHONJE

(10600529)

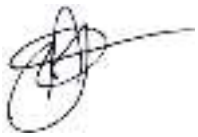
THIS THESIS IS SUBMITTED TO THE UNIVERSITY OF GHANA, LEGON IN PARTIAL FULFILMENT OF THE REQUIREMENT FOR THE AWARD OF DOCTOR OF PHILOSOPHY DEGREE IN PLANT BREEDING

WEST AFRICA CENTRE FOR CROP IMPROVEMENT
COLLEGE OF BASIC AND APPLIED SCIENCES
UNIVERSITY OF GHANA
LEGON

DECEMBER, 2020

DECLARATION

I hereby declare that except for references to works of other researchers, which have been duly cited, this work is my original research and that neither part nor whole has been presented elsewhere for the award of a degree.

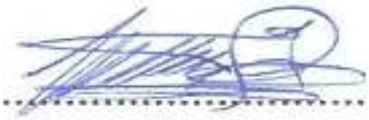


.....

ANDEKELILE SALONI MWAMAHONJE
(Student)

.....
PROF. KWADWO OFORI
(Supervisor)

.....
DR. JOHN ELEBLU
(Supervisor)



.....
PROF. TILEYE FEYISSA
(Supervisor)

.....
DR. SANTOSH DESHPANDE
(Supervisor)

ABSTRACT

Sorghum is an important staple food crop for semi-arid areas in Tanzania. However, the productivity is very low due to several constraints, particularly drought. The present study was conducted to develop sorghum genotypes with improved drought tolerance and grain yield. The specific objectives were to: i) identify farmers' production constraints and traits preferences of sorghum in central Tanzania. ii) Introgress drought tolerant quantitative trait loci (QTL) from donor parents into farmers preferred sorghum varieties and iii) identify traits contributing to drought tolerance of sorghum genotypes. A participatory rural appraisal (PRA) was conducted at Kongwa district in Dodoma region and Ikungi and Iramba district in Singida region to identify constraints facing sorghum production, farmers' trait preferences and coping strategies to address drought. Two donor parents B35 and S35 with stay green (STG) QTL 1, 2 and 3 were introgressed to the farmers preferred sorghum varieties to develop F_1, BC_1F_1 and BC_2F_1 populations. Five BC_2F_1 (NA307, W82, NA316, NA241 and SE438) populations and their parents were genotyped using 30 Single Nucleotide Polymorphism (SNP) markers. Results of the PRA showed that bird damage, poor soil fertility, drought stress, pests and diseases and lack of improved varieties were the major constraints of sorghum production. Early maturity, drought tolerance, high yield were ranked the highest preference when selecting new sorghum varieties. The coping strategies to address drought stress in sorghum involved early planting and use of drought tolerant varieties. Three (W82, NA241 and NA307) genotypes with heterozygous alleles and two genotypes (NA316 and SE438) with homozygous alleles were selected for selfing to generate BC_2F_3 populations. Moreover, plants with favourable alleles for either STG 1, 2, or 3 and good agronomic performance in field condition were selected. Eight genotypes namely NA241A, NA241B, NA307, NA316A, NA316B, NA316C, SE408 and SE438 from BC_2F_3

populations alongside with three parents and one check were selected after phenotyping BC_2F_2 population. The genotypes were planted in a split plot design under well watered and water stressed environments to determine the performance, correlation, heritability and genotypes by environments interaction. The results from the genotyping study revealed that, 7 out of 30 markers were for STG 1, 2 and 3 after genotyping of BC_2F_1 population; the remaining 23 markers were for traits contributing to STG in plants such as heat shock domain, programmed cell death triggering, aspartic proteases and chloroplast precursor. Genotyping of BC_2F_3 population indicated that, 7 SNP markers out of 10 had favourable alleles for STG in sorghum. Seventy one out of 728 BC_2F_1 samples genotyped were heterozygous. Of these, SNP markers snpSB00075, snSB00102 and snSB00103 were scored as heterozygous allele in seven samples of BC_2F1 with B35*Wahi background. Markers snSB00049, snSB00077, snSB00102 and snSB00103 indicated heterozygous allele in 37 samples of BC_2F_1 with S35*Pato background. The rest (19) SNP markers showed homozygous alleles for BC_2F_1 population. Eighteen SNP markers indicated favourable alleles among 728 of BC_2F_1 samples genotyped including nsSB00049 and nsSB00054 for STG 1, snpSB00089 FOR STG 2 and nsSB00102 and nsSB00103 for STG 3. The rest of alleles were favourable for other roles related to STG in sorghum during post flowering drought condition. Similar trend were observed in the genotyping of BC_2F_3 population which seven SNP markers indicating favourable alleles for STG 1, STG 2 and STG 3. The SNP marker snpSB00089 indicated the highest (729) total number of favourable alleles for STG followed by snpSB00101 (728) and snpSB00102 with 727 after genotyping of BC_2F_3 population. However, the SNP marker snpSB00103 failed to identiy at least one favourable alleles for STG in this study.The genotype W82 was associated with snSB00102 marker, NA241 (snSB00102 and snSB00103) and NA307 was linked with snSB00101 and snSB00102 markers for STG 3 in BC_2F_1 population. Grain yield per hectare varied from 1770 kg/ha of

donor parent B35 to 3415 kg/ha of BC_2F_3 genotype NA316C under well watered trial and from 1711 kg/ha of the donor parent B35 to 2652 kg/ha of genotype SE438 under water stressed trial. Significant differences were recorded in plant height, chlorophyll content, panicle length, panicle width, inflorescence exsertion, leaf rolling, grain weight per plant, panicle weight per plant and STG in both environments at $P < 0.01$. The mean performance of plant height was the highest (142.2 cm) in NA316C under water stressed condition and 143.3 cm under water irrigation. Interaction between genotypes by the environments were directed by majority of traits namely panicle length, panicle width, chlorophyll content, inflorescences exsertion, total number of leaves at physiological plant maturity, total number of green leaves at plant maturity, panicle weight, grain weight per panicle and root biomass. However, the interactions were not differed to plant height, leaf length and grain weight per plant. Panicle weight, panicle width and panicle length were significantly correlated with grain yield. STG and inflorescences exsertion were negatively correlated with grain yield. Chlorophyll content was correlated with total number of green leaves. A negative correlation was detected between traits STG and total number of green leaves. Above 50% heritability estimates were recorded in well watered and water stressed conditions. However, the interaction between genotypes by environments lowered the heritability of traits evaluated. Geometric mean productivity (GMP) and mean productivity (MP) were significantly correlated with yield under well watered environment (YP) and yield under water stressed environment (YS) and each other. There was low correlation between tolerance index and grain yield (0.12) in water stress environment. Stress sustainability index (SSI) showed low correlation with all indices compared thus; SSI is suggested as the best index for screening low yield under stressed environments. The genotypes NA307, NA316C and SE408 produced the highest grain yield per hectare across the environments. Therefore this study provided the bases of new genotypes which are promising for drought tolerance and yield.

The genotypes should be advanced to lines and recommended for release after further evaluations in different geographical locations cultivating sorghum in Tanzania.

DEDICATION

This thesis is dedicated to my parents Saloni Wenela Mwamahonje, Ane Adamu Mwasile and Lutiness Mwampashi, my lovely wife Elizabeth Nduna Mwampamba and my children Kelvin Andekelile Mwamahonje, Christopher Andekelile Mwamahonje and Kilian Andekelile Mwamahonje.

ACKNOWLEDGEMENTS

I extend my acknowledgement to those who contributed the achievements of this study. I am grateful to the DAAD and ACE II Project for sponsoring my PhD studies. I thank the Management and Staff of West Africa Centre for Crop Improvement (WACCI), University of Ghana, for considering me as a potential doctoral candidate in 2017-2021 and for support and the administration of my studies and scholarship. My gratitude goes to my supervisors, Prof. Kwadwo Ofori and Dr. John Eleblu for their guidance and valuable inputs in this thesis. My sincere thanks go to my in-country supervisors Prof. Tileye Feyissa and Dr. Santosh Deshpande for their guidance on this thesis. I am thankful to Dr. Justine Ringo of seed production at Agricultural Seed Agency (ASA) in Morogoro, Tanzania, Dr. Emmanuel Mrema Centre Director at Tanzania Agricultural Research Institute (TARI)- Tumbi Centre, Mr. Elias Letayo a retired Centre Coordinator at TARI- Makutupora Centre for supporting sorghum lines, selfing bags and constructive advice. Professor Vernon Gracen is acknowledged for guiding the appropriate method of crossing to generate backcrosses. My special thanks to Dr. Rajaguru Bohar, HTPG Project coordinator, Dr. Ana Luísa Garcia-Oliveira and Milcah Kigoni for training and co-funding genotyping cost at Intertek lab Sweden. I thank my colleagues Ashura Dulazi, Davis, Faraji Matata, Frank Mbilinyi, Isack Teya, Mary Stephen, Sara Mawele, Stella Libent and Zacharia Maseta at TARI- Makutupora and TARI-Hombolo Centres for assistance during research work. Dr. Cornel Massawe, Centre Director at TARI- Makutupora is acknowledged for providing me favourable environments for handling my research and write-up. I thank Mr. Matapa Ngungu, Atanansio Lampard, Joyce Mwaigaga and Leward Mwamahonje for participating in one way or another toward completion of this study. Mr. Leon Mrosso and the Director General TARI are acknowledged for offering me study leave for four years. I extend my thanks to WACCI, all my friends for their encouragements and technical support, and everyone who may have

contributed in one way or another to accomplish this work. I am also grateful to my late father Saloni Wenela Mwamahonje who passed away when I was a third year (2019); he made the foundation of my education. My gratitude goes to my mother and siblings, for their love, encouragements and prayers. My sincere thanks go to my wife, Elizabeth Nduna Mwampamba and our children, Kelvin, Christopher and Kilian Simon Mwamahonje, for their endurance, love and encouragement throughout my PhD studies.

TABLE OF CONTENTS

LIST OF TABLES

LIST OF FIGURES

ABBREVIATIONS

ACE: African Centre for Excellence

AFLP: Amplified Fragment Length Polymorphism

DAAD: The German Academic Exchange Service

DNA: Deoxyribo Nucleic Acid

FAOSTAT: Statistical Database of the Food and Agriculture of the United Nations

F1s: First Generation after Fertilization

GMP: Geometric Mean Productivity

HTGP: High Throughput Genotyping Project

ICRISAT: International Crops Research Institute for Semi-Arid Tropics

LSD: Least Significance Difference

MABC: Marker Assisted Backcrossing

MP: Mean Productivity

NGO: Non-governmental Organization

NIL: Near Isogenic Line

PCA: Principal Component Analysis

PRA: Participatory Rural Appraisal

QTL: Quantitative Trait Loci

RAPD: Randomly Amplified Polymorphic

RFLP: Restriction Fragment Length Polymorphism

SI: Stress Intensity

SNP: Single Nucleotide Polymorphism

SPSS: Statistical Product and Service Solutions

SSI: Stress Susceptibility Index

SSR: Simple Sequence Repeats

STG: Stay Green

STI: Stress Tolerance Index

TARI: Tanzania Agricultural Research Institute

TOL: Tolerance Index

URT: United Republic of Tanzania

USA: United States of America

WACCI: West Africa Centre for Crop Improvement

YP: Yield potential

YS: Yield in the water stress condition

YSI- Yield Stability Index

CHAPTER ONE

1.0. GENERAL INTRODUCTION

Globally, sorghum (*Sorghum bicolor* (L) Moench) ranks the fifth staple food crop after maize, rice, wheat and barley (FAOSTAT, 2017). It is the second priority staple crop after maize in Sub-saharan Africa and major food and nutritional security crop to over 100 million people in Eastern Africa. The centre of genetic diversity of sorghum is in Ethiopia (Teshome *et al.*, 1997). It was spread to Arabia, India and reached China in 3rd century; sorghum entered in the United States of America (USA) through slave trade (Ayana and Bekele, 2000). Seven sorghum races are commonly cultivated in different geographical regions globally. Dura is common in the Horn of Africa, Middle East and India, Sballu in India, Koaliang in China (Olsen, 2012). Dura is the oldest *Sorghum bicolor* race which was consumed for the first time by Egyptian over 4000 years ago. The genetic centre of Kafir race covers Tanzania and Southern parts of Africa (National Research Council, 1996). Kafir has thick, juicy stems, large leaves, and awnless cylindrical-shaped panicles. Milo originated from East Africa; it has stems that are less juicy than Kafir. The centre of origin for caudatum race ranges from Eastern Nigeria, Chad and Western Sudan while guinea race covers Western Nigeria and Senegal in West Africa (National Research Council, 1996). Hegari sorghum originated from Sudan, the group is part similar to Kafir but panicles are oval in shape with chalky white grain seed. Sorghum, which is very common in erratic rainfall areas, has not been exploited enough for utilization (Muui *et al*., 2013).

The United States of America (USA) is the top of sorghum producer globally, producing 9.2 million tons per year with productivity of 4.5 t/ha followed by Nigeria (6.8 million tons per

year with productivity of 1.2 t/ha), Sudan and Ethiopia (4.9 million tons per year with productivity of 2.7 and 0.7 t/ha respectively) (FAOSTAT 2018).

Tanzania produces a total of 796,570 tonnes of sorghum annually with the productivity of 1 tonne/ha (FAOSTAT, 2018). Three quarter of sorghum production in Tanzania is produced from Central, Lake and Eastern zones (Wortmann *et al*., 2006). Sorghum is mainly grown as the major food crop in the central zone (Dodoma and Singida regions), followed by Western zone (Tabora region) and Lake zone (Shinyanga, Mwanza, Simiyu and Mara) (Brown, 2013). Semi-arid lands comprise four zones including central zone with Dodoma and Singida regions, Shinyanga, Mara and Mwanza region in lake zone, some parts of Manyara region and the low land of Arusha and Kilimanjaro regions in the northern zone, part of Iringa region in the southern-highland zone; part of Tabora region in western zone and Lindi and Mtwara regions in the southern zone of Tanzania (Lyimo and Kangalawe, 2010; Mongi *et al*., 2010; Schechambo *et al*., 1999; Swai *et al*., 2012; URT, 2007).

In Tanzania sorghum is used as alternative source of food in areas where maize does not do well. It is used as the raw materials for making processed and local beers which is consumed by many people in the country. The crop is used as livestock feed for maximizing milk, eggs and broilers production. Sorghum is used for production of fuel, oil and wax in countries with advanced technology like United States of America (USA), India, Brazil and South Africa (Agrama and Tuinstra, 2003). In addition, sorghum is used to make porridge, ugali and can be prepared by mixing with beans (Brown, 2013). The grains contain over 70 ppm of iron with more than 50 ppm of zinc, which is an important nutrient for human health. The need for sorghum grains is high due to increasing demand by the industry sectors for processing by-products (Ringo *et al*., 2015).

Despite its ability to adapt drought stress, pests and diseases in the marginal lands where other cereals usually perform poorly, its productivity in Tanzania is low compared to most

production regions elsewhere. Semi-arid areas (Dodoma and Singida regions) grow sorghum as one of the main food crops, and are most vulnerable to food shortage due to inconsistency of rainfall (Brown, 2013). Low yield of sorghum in central zone may be because of different factors including rain-fed agriculture, pests and diseases and dry spell during post-flowering (Barron, 2003). Early planting close to start of rainfall is among the strategies used by farmers in central Tanzania to escape drought, with assumption that, seed will germinate upon start of rainfall (URT, 1997). Further planting is done as soon as it rains during planting period in December/January to cater for moisture for seed germination and physiological growth of the plant. Nonetheless, rains may commence late and sometimes does not fall for some weeks which may cause wilting of the seedlings. Farmers may re-plant two to three times per season due to unpredictable rainfall. Farmers plant sorghum in different dates with probability that one of the planting date may end up with good yield (Mwanga, 2002). They plant both early and late maturing varieties. Early maturing sorghum varieties do not resist much dry spell compared to late maturing varieties which performs better if a dry-spell occurs during the middle of rainfall season (Mwanga, 2002). The central zone is considered as food insecure every year among regions in Tanzania due to drought constraint in semi-arid areas in Dodoma and Singida regions (Ministry of Agriculture, 2019). Drought causes leaf rolling and loss of STG in sorghum which reduces yield up to 100% majoring with terminal drought which is more important to yield reduction than other types. Terminal drought affects the crop at flowering and grain filling stages. At grain filling plants need sufficient water for high yield. In addition, *Striga hermothica and S. asiatica* causes 30 - 90% yield loss (Mrema *et al*., 2017). Farmers who prefer to grow maize in semi-arid areas are advised to grow sorghum as an alternative crop to avoid risk of maize harvest loss.

A number of approaches have been recommended to address the drought stress in sorghum. For instance, a use of participatory rural appraisal which involves farmers provides the

current information on traits lacking in their cultivated varieties for improvement. The information helps plant breeders to develop new varieties which contain farmers' traits preferences (Mamoudou *et al*., 2006). A number of improved sorghum varieties were released in Tanzania for drought tolerance and yield over two decades ago. For instance *Lulu* and *Serena* were released in 1970s (Gierend *et al*., 2014). Varieties *Tegemeo* and *Pato* were released in 1978 and 1995 respectively. *Pato* variety is early maturing, drought resistant, has good taste, resistant to lodging and with yield of 2.5-4 t/ha. The variety is also preferred by birds and field insects such as stem borers and storage pests. The grain is basically used for food and brewing. *Macia* was realised in 1999 with the attributes of good taste, high yielding, drought resistant, high tillering, and white grain colour. The crop is affected by field insects, storage pests and birds; it is used as a source of food (Mgonja *et al*., 2005). A private seed company released *Sila* as a new sorghum variety in 2008 (Monyo *et al*., 2004). *Wahi* was released in 2002 as tolerant to *Striga*, early maturing, drought resistant, high yield and high tillering. The variety is preferred by birds and also severely attached by stem borer, aphids and storage pests. It is purposely grown for food and brewing. *Hakika* variety was released for the first time in 2002 with attributes of *Striga* tolerance, early maturing and drought resistant. *Hakika* is preferred by birds, insects and storage pests. It is used as food and for alcohol production. *NACO- Mtama 1* was released in 2013 with attributes including early maturing, large grain size, and high yielding but is preferred by birds and pests (aphids and stem borer). The variety has characteristics of shattering after maturity which needs early harvesting. The variety was released for food and brewing. *Seguifa* was developed in West Africa (ICRISAT- Mali), it was released in 1995. It has attributes including; early maturing from 95-100 days from planting, drought tolerant, high yielding, high tillering but is highly susceptible to stem borer and aphids; this variety was released for food.

The above varieties were released as introductions for drought tolerance and high yielding traits. However, no pedigree information is available in Tanzania. Besides, the varieties were released a long time ago and efficiency of performance of traits has been reduced because of several factors including drought. Although sorghum tolerates drought, flooding and heat compared to other cereal crops, under extremely drought it fails to adapt (Dicko *et al*., 2006). Drought is a complex phenomenon which is controlled by different genes and exploitation of these genes needs advanced technology (Ashraf *et al*., 2008). Among the technologies, the application of molecular markers have the potential which allows for manipulation of genetic information and combine with appropriate agronomic traits associated with drought tolerance before final selection of lines to advance (Ejeta and Knoll, 2007). Molecular markers are used for detection of QTLs that help to express the phenotypes in plants such as, pre-flowering and post-flowering drought tolerance in sorghum (Ejeta and Knoll, 2007). Molecular markers associated with high yield and drought tolerance under well watered and water stressed conditions are screened for the improvement of drought tolerance in sorghum (Tuinstra *et al*., 1996). Marker alleles expressing high yield under well irrigation treatment are associated with phenotypic trait data under limited irrigation water condition. Selection of new sorghum lines should be for those with positive correlation under well watered and water stressed conditions (Tuinstra *et al*., 1996). Stay green (STG) is conditioned by many genes which need diversity approaches to identify the position of those genes or QTLs which are introgressed to non-stay-green sorghum (Kiranmayee *et al*., 2020). QTLs for STG 1, STG 2, STG 3, and STG 4 were mapped from B35 sorghum lines using molecular markers (Tuinstra *et al*., 1997; Tao *et al*., 2000; Xu *et al*., 2000; Kebede *et al*., 2001; Sanchez *et al*., 2002). These lines originated in Ethiopia and are used as donor parent (B35 and E-36) for introgressing to non- STG sorghum (Kassahun *et al*., 2010). Markers identify QTLs and chromosome location associated with genes expressing for drought tolerance in sorghum

(Ashraf *et al*., 2008). Other donor parents (K359w, K260, S35SG06008 and S35SG06040) for drought tolerance were developed at ICRISAT. Uses of donor parent lines with drought tolerance in origin have contribution in sorghum improvement for drought adaptability (Ringo *et al*., 2015).The use of marker assisted backcrossing (MABC) is a common method for introgression of already mapped STG QTLs associated with drought tolerance genes to drought sensitive varieties. MABC approach allows transfer of traits of interest from the donor parents to recurrent parents by successive backcrossing (Walulu *et al*., 1994). MABC supplements phenotypic data for the efficiency screening of traits in sorghum (Ashikari and Matsuoka, 2006; Takeda and Matsuoka, 2008). During phenotypic selection of the suitable sorghum lines based on the data, early maturity is one of the important traits considered for sorghum improvement as it can escape from drought effect (Ansarifard *et al*., 2020; Kamal *et al*., 2017; Mwanga, 2002). Physiological parameters contributing to drought tolerance are important for evaluation of sorghum lines performance. Unfortunately no studies have been conducted to address drought stress in sorghum by MABC in Tanzania. Therefore, there is a need to explore the feasibility of developing new sorghum lines for drought stress tolerance by marker assisted selection. Marker assisted selection simplifies selection of elite lines and shortens breeding cycles.

The general objective of this study was to develop drought tolerant and high yielding sorghum genotypes through maker assisted backcrossing using SNP markers

The specific objectives were to:

i. identify farmers' production constraints and traits preference of sorghum in central Tanzania;
ii. introgress QTLs for drought tolerance into farmers' preferred sorghum varieties; and
iii. determine traits' performance contributing to drought tolerance in sorghum.

CHAPTER TWO

2.0. LITERATURE REVIEW

2.1. Farmer involvement in sorghum improvement

Participatory rural appraisal (PRA) is a concept which indicates farmers and plant breeders have different and highly complementary skills which together achieve better outputs of crop yield through collaboration than each working alone (Mc Guire, 2008). Farmers and plant breeders stand a better chance of developing new crop varieties which address constraints of production (Mc Guire, 2008). PRA increases options for addressing sorghum production constraints such as diseases and pests, salinity, drought and poor soil fertility in sorghum crop (Mwamahonje *et al*., 2021). It is important for sorghum breeders to involve farmers who have diverse knowledge of traits to their local areas for successful breeding demand driven. Sorghum stakeholders for instance farmers, brewers and agro dealers have different prefferences of sorghum traits, therefore, should be consulted to identify their preferences for incorporation in the new varieties (Kaliba *et al*., 2018). New sorghum varieties which meet stakeholders' traits preferences are considered for release to increase the adoption rate ((Monyo *et al*., 2004; Nuijten *et al*., 2013). Trait preferences are also important to socio-economical, cultural, and individual factors, and these can be used for creating awareness of improved sorghum seed for increasing the adoption rate (Monyo *et al*., 2002). The approach is useful for crop improvement as it advocates interactive science where stakeholders are part of it (Roling *et al*., 2004).

2.2. Constraints facing sorghum production

2.2.1 Drought

Drought affects nutrients and water uptakes from the soil due to limited moisture which hinder root development. It affects the process of photosynthesis, respiration and dry matter partitioning in the plants depending on the type of drought (Pang *et al*., 2017). Drought affects sorghum plants differently at physiological growth stages; for instance, at seedling, pre-flowering and post-flowering stages which contribute to the loss of final yield. Pre-flowering and post-flowering drought reduce grain yield (Borrell *et al*., 2014; Hammer *et al*., 2014). Water stress at post-flowering growth stage causes yield losses of 87-100%, at this stage plants need plenty of water for grain filling (Craufurd and Peacock, 1993). The degree of drought impact in sorghum changes from one environment to another. The ability to maintain the stay-green trait differs from one variety to another; nonetheless, stay-green is positively correlated with grain yield in sorghum (Borrell *et al*., 2014). During drought condition, the plant utilises water present in the root zone for survival. The efficiency of a plant to absorb water from the soil depends on the ability of the root to penetrate into deep soil (Xiong *et al*., 2006). Studies show that, drought tolerant crop species have higher roots with more root density (Wasaya et al., 2018). Drought affects morphological features of a crop for instance, leaf rolling, yield, number of leaves, leaf area index, leaf shape, number of seeds per plant, reduced growth of roots and reduced different growth stages (Saddam *et al*., 2014). Moreover, 70-75% loss is caused by drought because of irregular rainfall which is associated with climate change. Farmers in these areas practice mixed farming, avoid the use of improved varieties which are susceptible to bird attack and abandoning of some local landraces especially late maturing with poor yield under drought condition (Bucheyeki *et al*., 2010).

Rainfall in semi-arid areas is erratic; it falls in a single rainy season from December to April per year depending on geographical, seasonal and annual variability (Msongaleli *et al*., 2015). Usually rainfall is characterized by heavy storms with floods. This causes run-off instead of penetrating the soil to support plant growth (Msongaleli *et al*., 2015). Sorghum is a drought tolerant crop that can survive and improve yield in low rainfall and soil fertility. It grows from 400-600 mm annual rainfalls. Under good agronomic management, for instance, timely planting, weeding, fertilizer application, pest and disease control, the crop produces high yield. However rain is unreliable and inconsistent ranging from 300- 400 mm per year which does not satisfy the crop requirement like sorghum in semi-arid areas. The rainfall may start early or is delayed which makes it difficult for farmers and agricultural experts to know the trends of the rainfall. Therefore, the sorghum crop is commonly adapted to semi-arid areas where most cereal crops fail to cope. The ability of sorghum to survive under drought stress helps farmers to produce sorghum for food security (Bibi *et al*., 2012). Sorghum has capacity to cope with different types of stresses, such as water and salt-stresses, high temperature, wind and flooding (Ejeta and Knoll, 2007). Nonetheless, sorghum is usually affected by drought stress at pre- flowering and post-flowering growth stages which have critical effect during seed setting after anthesis stage (Kapanigowda *et al*. 2013). Drought lowers sorghum grain yield causing food insecurity especially in Sub-saharan Africa where sorghum is used as the main food crop (Saddam *et al*., 2014). The erratic nature of rainfall constraint sorghum production in semi-arid areas where agriculture is typically dependent on rain fall (Mkonda and He, 2017). More efforts to research demand driven outputs are needed to improve the farmers' agronomic practices on sorghum production especially use of improved seed, fertilizer use and agronomic crop management (Ochieng *et al*., 2011). Sorghum varieties which were introduced from the research institutions have numerous problems including lodging, poor drought tolerance and poor grain quality, attacked by birds and difficult to store

because of poor storage infrastructure (Mwamahonje et al., 2021). Farmers need to develop management strategies for dealing with these problems (Kudadjie *et al*., 2004). Plant breeders should develop new varieties which are demand driven by stakeholders. The diagnostic study suggests that because farmers produce their own seed, improving the quality of their seed is a benefit for them (Kudadjie *et al*., 2004). Plant breeders have responsibility to work closely with farmers to identify the problems and solutions of sorghum production to increase the adoption rate.

2.2.2. *Striga* infestation

Striga asiatica and *S. hermonthica* infestation is among the constraints which hinders sorghum productivity worldwide (Zhu-Salzman *et al*., 2004). *Striga* infestation causes sorghum grain losses about 22-27% in Sub-saharan Africa (AATF, 2011). In Tanzania, it covers the major parts producing sorghum and maize. The infestation is concentrated in different zones like lake zone (Mwanza, Mara, Shinyanga and Simiyu regions), Western zone (Igunga in Tabora region), Central zone (Dodoma and Singida regions) (Mrema *et al*., 2017). This weed is associated with limited soil nutrients (Ejeta, 2007) and it causes a severe loss of yield resulting into food insecurity in sorghum and maize producing areas (Gebretsadik *et al*., 2014).

2.2.3. Lack of technology adoption

Farmers fail to apply technologies in their field which cause poor soil fertility, low moisture conservation and low yield (Marenya and Barrett, 2009). High compact soil is cultivated by magoe ripper to increase soil porosity. Uses of magoe ripper technology have been reported to increase sorghum grain yield by 100% compared to zero tillage. Zero tillage causes high run-off of water and poor water resources utilization due to low technology or inaccessibility

to extension officers who advise farmers to adopt improved agronomic management (Msongaleli *et al*., 2015). Deforestation reduces soil fertility and poor distribution of rainfall which affect sorghum production. Moreover, poor crop protection has facilitated the diseases and pests hindering crop in the field and storage room (Msongaleli *et al*., 2015).

Most of improved sorghum varieties produce higher yield than local varieties with short period to maturity (90-110 days) (Mwamahonje and Maseta, 2018) however, farmers face difficulties of storage facilities. Improved sorghum varieties are affected more by storage pests compared to the local ones same as in maize crop (Midega et al., 2016). Palatability is one of the traits which are considered by consumers in sorghum, in areas where improved palatable sorghum varieties are lacking farmers opt to adopt local varieties which have low yielding ability (Timu *et al*., 2014). Farmers produce improved sorghum varieties purposely for market while local is produced for food. It is recommended that breeders should consider palatability and taste in sorghum improvement (Timu *et al*., 2014). Improved varieties have low palatability and hence palatable traits need to be incorporated in during breeding new varieties to cut-out this constraint (ECARSAM, 2005). The farmers who use improved varieties produce about 1 t/ha which is very small compared to the standard yield proposed by world and Africa with an average of 5 t/ha (ECARSAM, 2005). More effort is needed to educate farmers on the use of improved sorghum varieties which are drought tolerant, high yielding and demand driven to increase production.

2.3. Drought coping mechanisms in sorghum

Drought coping mechanism is the ability of plants to withstand drought stress while maintaining appropriate physiological activities to stabilize and protect cellular and metabolic integrity at tissue and cellular level (Tuinstra *et al*., 1997; Xiong *et al*., 2006). Drought tolerance is a complex trait associated with QTL caused by the interaction of environmental

and genetic factors. It allows cell survival with less metabolic activity leading to low yield of plants (Izanloo *et al*., 2008). Sorghum is affected by drought in early growth stages and pre flowering and post flowering growth stages (Badigannavar *et al*., 2018). Drought occurring during early growth stage; weakens growth of sorghum plants because of limited water and nutrients absorption from the soil. Pre-flowering drought is common when plants are at vegetative growth stage to pre-flowering; it affects physiological and morphological plant growth (Wani *et al*., 2012). However, plants have ability to resume normal physiological growth on drought recovery. Post-flowering drought occurs during post-flowering growth stage. It affects grain filling in cereal crops as at this stage plants need sufficient water; therefore it causes the significance yield loss (Verma *et al*., 2018). Notwithstanding, plants have mechanisms to cope with the drought stress including physiological, morphological and biochemical mechanisms.

2.3.1 Physiological mechanisms

Physiological mechanism involves the adjustment in photosynthesis, chlorophyll content, stomatal conductance and transpiration rate. The plants reduce leaf water potential; regulate osmotic potential while keeping high chlorophyll content for photosynthesis to support plant growth (Baccari *et al*., 2020). Sorghum copes with drought attributable to regulated physiological processes which occur during growth. The physiological traits that account for the adaptation include stomata conductance which act as the regulator of photosynthetic process. It closes stomata during drought to minimize water losses during transpiration and photosynthesis (Tuinstra *et al*., 1997); (Xiong *et al*., 2006). Sorghum allows accumulation of high chlorophyll content and maintains STG which is important for grain yield (Blum *et al*., 1989). Below is the detailed discussion of physiological mechanism of sorghum to drought.

2.3.1.1 STG sorghum

STG refers to the retention of green colour in plant leaves which delays leaf senescence for enhancing photosynthesis during water stress (Thomas and Ougham, 2014). The role of STG in plant is to maintain chlorophyll while reducing the leaf senescence in plants for survival. Plants with sufficient STG have advantages to partition nitrogen in plant leaves which offer efficient transpiration and radiation which reduces sink carbon tank (Borrell and Hammer, 2000). This enhances yield under limited water conditions during post-flowering. STG trait is categorized into five types. The first and second type is functional STG, which triggers and regulates onset of leaf senescence. The third and fourth type plays a major role in maintaining plant greenness which undergoes photosynthesis with the STG (Thomas and Howarth, 2000). The fifth is type E of which its green leaves is associated with the initial chlorophyll content. The study of drought tolerance in sorghum targets the first and second type of STG which is functional STG (Thomas and Howarth, 2000). Functional STG has significant contribution to carbon income of the plant during drought stress (Meru, 2010). STG in sorghum is an important trait that contributes to drought tolerance. It has been exploited through marker assisted selection by backcrossing (Edema and Amoding, 2015). Sorghum varieties with low levels of STG at post-flowering stages lack drought tolerance mechanism. Water deficit, during post flowering, lowers chlorophyll content; this increases leaf senescence in the plant (Borrell *et al*., 2014). Genotypes with drought tolerance show low level of leaf senescence maintaining green colour on leaves which account for photosynthesis in sorghum plants. Sorghum has ability to grain fill under water shortage due to high level of STG (Borrell *et al*., 2000). Deployment of STG traits through delaying leaf senescence is one of the strategies to improve sorghum production in semi-arid areas. This improves grain production, fodder and quality of sorghum products with positive correlation to STG (Reddy *et al*., 2014). The critical stage of quantifying the ability of sorghum variety to maintain STG is during pre-flowering and post-flowering stages when optimum water is needed for vegetative growth

and production, respectively. The lines with high STG are screened in breeding programmes to develop drought tolerant varieties (Borrell *et al*., 2014). Large size of canopy, number of tillers, leaves, and leaf size at pre-flowering may contribute to the delay of pre-anthesis (Ahlawat *et al*., 2008). STG is associated with proline production in plants. During drought stress plants tend to secret high amount of proline as a strategy of coping with the drought condition (Hayat *et al*., 2012). This is enhanced by a combination of activities achieved by diversity of proteins in plants. Two drought tolerant sorghum varieties have been developed in Ethiopia B35 (BTx642) and E-36-1). Sorghum lines with QTL related with drought tolerance in addition to B35 and E-36-1 include K359w, K260, S35SG06008 and S35SG06040. The line B35 contains most of the QTLs STG1, STG 2, STG 3 and STG 4 for STG traits shown during post flowering and E-36-1 contains QTLs for STG shown during pre-flowering stage (Edema and Amoding, 2015).These sorghum lines are good sources of drought tolerance for improvement of sorghum. STG QTLs can be introgressed to drought sensitive recurrent parents by MABC method. In addition, STG has been reported to increase resistance to lodging (Sanchez *et al*., 2002). Delaying loss of green tissues is the trait of interest for addressing the challenge of drought stresses in plants. Sorghum varieties with delayed STG have high chlorophyll content, chlorophyll content index and the leaves remain green for a long period of time during post-flowering water stress (Luche *et al*., 2015). Drought tolerant sorghum produces high grain yield and relative high biomass compared to non- STG varieties under limited water conditions. The major genetic variations among STG sorghum have been reported in Ethiopia (Borrell *et al*., 2000). The delayed leaf senescence which accounts for STG in sorghum is increased by specific leaf nitrogen (Vadez *et al*., 2013). This occurs because during young stages, most hybrid sorghum varieties have high levels of nitrogen for vegetative growth that increases high specific leaf nitrogen during post-flowering water stress (Borrell *et al*., 2000b). Three regions in the sorghum genome are

associated with 30% variation in the STG (Thomas and Howarth, 2000; Tao *et al*., 2000). About 47% biomass of sorghum is produced at post flowering stage during water stress (Borrell *et al*., 2000). Phenotyping of drought tolerant sorghum accessions is tedious and time consuming. Biomass in sorghum shows good heritability in the yield performance in response to harvest index. In this approach, QTLs which are tightly linked with molecular markers and genes encoding drought tolerance in sorghum are selected for exploiting STG traits to potential lines (Walulu *et al*., 1994; Tuinstra *et al*., 1997). During introgression of drought tolerance QTLs both foreground selection and background selection of desired traits are exploited from targeted regions of the sorghum genome. Most of the QTLs introgressed to the potential recurrent parents are intermediate, about 40%, depending on the environmental factors which have influence to QTLs introgression (Walulu *et al*., 1994). The effect of post flowering under water stress starts 15 days from withholding irrigation (Kassahun *et al*., 2010). Sorghum plants use water stored in the deep and high density roots although the production of carbohydrate automatically is reduced. The reduction of water in the soil has effect on the coleoptile length, higher root shoot ratio and longer roots. Drought stresses reduces the fresh weight and the biomass of root, leaf growth and enhances roots growth (Ali *et al*., 2011). The root structure and anatomy account for water uptake by plants. Drought stress causes reduction of leaf area which is enhanced by decrease of cell division with small leaf size that reduces eva-transpiration. Leaf senescence is affected during overexpression and under expression of the hormones if enzymes do not work efficient, this reduces photosynthesis efficiency in plants (Jordi *et al*., 2000; Peleg *et al*., 2011). Rivero *et al*. (2007) reported the successful introgression of STG in sorghum by transgenic technology by using an enzyme isopentenyltransferase (*IPT*) which catalyses the rate of cytokinin expression in sorghum. However, the yield of new sorghum varieties developed by this technology is low compared to other methods of sorghum improvement (Rivero *et al*., 2007). Even though,

there are different techniques/mechanisms under which plants maintain STG during limited moisture, STG QTLs developed by mapping using marker assisted selection seems to be the one which to a large extend is expressing STG phenotype in sorghum.

2.3.1.2. Chlorophyll content

Chlorophyll is an important part of the plant which governs photosynthesis process for the production of food. Chlorophyll is classified into five; type a found in higher plants, b in plants and green algae and chlorophylls c, d and e, in some algae. Chlorophyll, a photosynthetic pigment, is involved in light absorption and plays an important role in plant photosynthesis. Chlorophyll converts sunlight energy to chemical energy in the in the presence of sunlight and water. Limited sunlight and water reduces chlorophyll production as well as yield in crops (Fu and Huang, 2001). Drought reduces the production of chlorophyll as it reduces surface area of greenleaf and causes high transpiration rate. Drought destroys photosynthetic parts thus reduces chlorophyll content in plants (Fu and Huang, 2001). Crops which grow in semi-arid areas have characteristics to cope with the situation, for instance sorghum has ability to resume photosynthesis and physiological growth after phase of drought stress. It develops roots which assist to transport water and nutrients from the soil to the plant but it reduces the final yield. Chlorophyll content decreases with increases of drought stress, however, in some cereals such as black gram [Vigna mungo (L.) Hepper] which contain high level of a chlorophyll content, chlorophyll content increases under moisture stress condition (Ashraf and Karim,1991). High level of type *a* chlorophyll can facilitate chlorophyll content recovery compared to type *b* which confers drought tolerance in in sorghum (Kapanigowda *et al*., 2013). Sorghum varieties which are drought tolerant might contain higher ratio of type *a* chlorophyll to type b which is useful for introgression into the recipient parents for improvement of drought tolerance (Kapanigowda *et al*., 2013).

2.3.2 Morphological mechanism

Morphological mechanism for drought tolerance in sorghum involves morphological traits which are associated with drought tolerance in sorghum for instance plant height, leaf arrangement, root system, root to shoot ratio and biomass accumulation. Plants with large weight of below to above ground biomass production have a high chance to tolerate water stress (Getnet *et al*., 2015). The biomass production depends on roots and shoots growth in plants. Drought affects more shoot growth than root growth in sorghum because of high level of temperature which increases rate of transpiration in plants (Bibi *et al*., 2010).

2.3.2.1. Leaf rolling

Leaf rolling is a mechanism used by plants during drought stress to prevent water loss by reducing leaf surface area (Kadioglu and Terzi, 2007). Leaf rolling is common in cereal crops for instance; previous studies by Kusaka *et al*. (2005) reported that, folding of pearl millet plant leaves during water deficit as the mechanism of survival. Leaf rolling and stomatal conductance play a major role as the physiological indices accounting for drought tolerance in plants. Leaf rolling is regulated by leaf water potential while stomatal conductance governs soil moisture (Assefa *et al*., 2010). Sorghum maintains stomata opening due to low level of leaf water potential. The positive correlation of leaf rolling against leaf water potential have gained advantage for breeders who use leaf rolling as one of the parameters for drought tolerance estimation (Baret *et al*., 2018). Leaf rolling caused by reduction of leaf water potential differs from one plant species to another. This is due to failure of plants to adjust to low leaf of osmotic potential at low leaf water potential (Amelework *et al*., 2015). In addition, leaf rolling is used as desiccation avoidance as it has negative influence to transpiration rate caused by changes in leaf stomatal conductance and reduction of leaf area so as to lower the water loss (Kadioglu and Terzi, 2007). Leaf rolling helps to rescue plant

survival by stomatal closure. While there is lack of sophisticated instruments for transpiration rate measurements and stomatal closure measurements for drought tolerance in sorghum, leaf rolling is a good alternative for screening drought tolerance. Plants with high drought tolerance show low stomatal conductance compared to susceptible plants (Kadioglu and Terzi, 2007). Low stomatal conductance tends to raise leaf temperature which causes high transpiration in drought tolerant plants (Kadioglu and Terzi, 2007). Stomatal closure is caused by high leaf temperature and transpiration rate. Plants with high susceptibility to drought are enhanced by high stomatal conductance with low leaf temperature (Kadioglu and Terzi, 2007). The difference between stress and non-stress sorghum genotypes could probably be due to poor correlation between drought tolerance and yield potential (Amelework *et al*., 2015). Plants with drought tolerance donot necessarily produce high yield. Therefore, plant breeders screen accessions with combined traits across the environments (Simova-Stoilova *et al*., 2016; Siebers *et al*., 2015).

2.3.2.2. Root system

The root systems influence drought tolerance in sorghum as they help to enhance water absorption from the soil to plant parts during water stress (Grieder *et al*., 2014). Roots grow about 2 to 3 cm per day and are the first parts of plant affected by drought stress (Routley *et al*., 2003). Sorghum roots grow to about 1 to 2 m depth in the soil at booting stage; this enables uptake of water at a distance of 1.6 m lateral from the plant (Routley *et al*., 2003). The root system is characterized by high level of osmotic adjustment all together contributing to drought tolerance in sorghum (Bibi *et al*., 2012). Root growth reaches stationary phase during the flowering stage, especially for non- STG sorghum genotypes (Robertson *et al*., 1993). Sorghum genotypes with seminal roots and large vessel diameters have high tolerance in limited water environments (Blum, 2004). Root distribution and root system structure in

sorghum depends on the carbon partitioning to the roots, which increases plant survival during drought stress (Blum, 2004). Plants with high ability to establish high root growth in water stress have high ability to tolerate water deficit attributable to increased root growth, which increases contact with soil thereby enhancing water uptake (Yambao *et al*., 1992). During drought situation, the plant absorbs water stored in the root zone however; the efficiency of absorption differs from one crop to another or from one variety to another. The crops with tolerance to drought absorb water from deep soil for survival (Xiong *et al*., 2006). Studies have revealed that drought tolerant varieties have deep roots and long lateral root systems which support the uptake of water and nutrients for plant use (Lynch, 2013; Yu *et al*., 2008). In addition, thick roots and large branches of roots penetrate the compact soil which facilitate high uptake of water for plant use (Simova-Stoilova *et al*., 2016). Therefore, the association of root architecture and root branching should be exploited to improve drought tolerance in sorghum as it helps to retain STG during water stress.

Root to shoot ratio is the proportion of root to shoot development during physiological plant growth. The production of root and shoot depends on availability of water for plant intake. Limited water affects root and shoot development in plants. In sorghum, the root to shoot ratio increases with decrease of soil moisture as the mechanism of survival (Sher *et al*., 2013). However, there is a variation in efficiency for maintaining root to shoot ratio among sorghum varieties. Sorghum with high level of STG accumulates large weight of biomass to support plant (Borrel *et al*., 2014). The primary roots growth of sorghum is slower than secondary roots growing from root crown. The strength of root system and biomass production therefore depend mainly in secondary growth root system (Tari *et al*., 2012). Root morphology, root biomass and structures of root systems enhance tolerance of sorghum in limited soil moisture (Assefa and Staggenborg, 2011). Thus, further studies to understand the

mechanism that confer increase of root to shoot ratio during drought tress should be given priority in sorghum improvement.

2.3.2.3. Biomass accumulation

Biomass refers to the dry matter after extraction of water by drying in the oven. Biomass production differs among plant species and varieties. It is among the traits which are used as an indicator of drought tolerance in plants including sorghum. Drought tolerant lines are characterized by high production of biomass as the way of adaptation for survival. About 47% biomass of sorghum is produced at post-flowering during water stress (Borrell *et al*., 2000b). Biomass is grouped into two types; the first type is above ground biomass which is composed of dry matter of plant parts above the ground for instance the shoot. The second type is below ground biomass composed dry matter below the ground for instance roots. Biomass accumulation in sorghum is contributed by high efficiency of photosynthesis which is driven by high chlorophyll content in leaves, STG and high concentration of carbon-dioxide (CO2) in the atmosphere (Ogbaga *et al*., 2019). Biomass accumulation is used as an indicator of drought tolerance in sorghum during post-flowering drought in the context that, it enables physiological plant growth and produce reasonable grain yield under low moisture though, the yield is low. Borrell *et al*. (2014) confirmed that biomass accumulation in sorghum is correlated with STG, sorghum genotypes with high STG have high biomass accumulation compared to non- STG genotypes. High production of biomass at the post-flowering growth stage increases yield potential than partitioning (Habyarimana *et al*., 2002). Increase of biomass accumulation in sorghum could also be attributable to enzymes which are active during moisture stress, to rescue plants from wilting. Therefore, it is recommended to take consideration of the biomass production when testing lines for drought tolerance in sorghum.

2.3.3. Biochemical mechanism

Biochemical mechanism involves various biochemical products accumulation by plants during drought (Ogbaga *et al*., 2016). Biochemical mechanism involves the biochemical traits such as proline and photosynthetic pigments which assist the plant to adapt to drought condition (Getnet *et al*., 2015). Drought in sorghum accumulates reactive oxygen species and breakdown of the cellular membrane which hinder the metabolic reactions in plants (Ahmed *et al*., 2016). Studies have shown that drought-sensitive crops like, maize, barley and tobacco accumulate higher H_2O_2 and lipid peroxidation than that in drought-tolerant ones (Ahmed *et al*., 2016). Drought stress enhances the reaction of enzyme anti-oxidant, it has been reported that plants which produce more anti-oxidants have more chance of tolerating drought than those with less anti-oxidants (Su *et al*., 2017). These findings indicate that, enzymes anti-oxidant activities increase in drought stress tolerant sorghum than drought stress sensitive sorghum (Amoah and Antwi-Berko, 2020). Such findings point out the need to further study of the mechanism of enzymes anti-oxidant activities to drought stress in sorghum. Sorghum accumulates high content of proline as the means of minimizing drought effects (Getnet *et al*., 2015). In addition, nitrogen also has significant contribution to adapt to drought in sorghum. Drought induces biochemical reactions in plants which persuade the secretion of compatible solutes, dehydrins and drought induced proteins (Badigannavar *et al*., 2018). The compatible solutes osmolytes are responsible for protecting plants from high levels of osmotic stress during water stress. The accumulation of compatible solutes in plants is the indicator of drought stress. The production of solutes plays the mechanism for adaptation of drought condition. Biochemical compounds are accumulated in herbaceous plants as the means of plants protection, such solutes should be exploited for improvement of crops (Ofgbaga *et al*., 2014). Heat Shock Proteins (HSPs) and dehydrins (DHNs) are classifications of proteins which increase with increase of drought stress in sorghum and maize (Ogbaga *et*

al., 2014). Thus, traits related to biochemical mechanism for drought tolerance in sorghum need to be studied at molecular level to widen the chance of detecting QTL related with drought tolerance in the biochemical compounds. Many studies have been conducted to understand the mechanisms which trigger drought tolerance in sorghum (Kapanigowda *et al*., 2014; Ngugi *et al*., 2013). Most studies have been undertaken in the greenhouse and/or screen-house which may not reflect the actual field conditions. Further tests are important under field conditions to determine the adaptability of plants not only against drought stress but also other biotic (pests and diseases) and abiotic stresses (soil nutrients). This will help to screen the genotypes with combination of traits associated with drought tolerance.

2.4. Water Use Efficiency

Water use efficiency is the biomass production per unit water use. It is imperative in physiology studies (Monclus *et al*., 2006). However it is difficult to screen for in a breeding programme as it differs depending on the accessions and environmental factors (Condon *et al*., 2002). There is inconsistent relationship between water use efficiency and dry matter accumulation in the experimental trials treated with different water regimes (Blum, 2005). Climatic and genotypic factors have effects on water use efficiency. High temperature and wind increase transpiration rate which is negatively influencing water use efficiency in crops (Schymanski and Or, 2016). Water use efficiency has contribution to drought tolerance and non-drought tolerance in crop genotypes depending on genetic differences (Condon *et al*., 2002). High water use efficiency increases biomass and leaf area per plant (Assefa *et al*., 2010). Yield performance under drought condition is controlled by genetic variations controlling yield potential, drought tolerance and water use efficiency (Hasan *et al*., 2016).

2.5. Methods of emasculation in sorghum

Sorghum is a cereal crop with outcrossing ranging from 5-30% close to anthesis and after flowering. Outcrossing in sorghum is attributed to wind and insects movement from one flowered plant to another (Barnaud *et al*., 2008). Different methods are used for anthers emasculation sorghum. Hand emasculation method involves the removal of anthers of flower close to flowering by use of forceps and needle. Emasculation is done a day before flowering to avoid outcrossing. Hand emasculation method has high risk of outcrossing if contamination is not properly controlled. The emasculated panicle of sorghum plant is covered by paper bag. Hand emasculation method need skilled person to avoid damage of floret during emasculation. It should ensure all pieces of anthers are removed to avoid self-pollination. The second method is hot water emasculation which involves hot water to kill anthers before producing pollen. The pollen has higher sensitive to temperature and humidity than stigma a female part of the flower. The head of sorghum plant prior to flowering is immersed in the hot water at 42-48°C for 10 minutes which kills pollen leaving viable female organs. However, there is possibility of self-pollination in few crossed plants. The self pollinated plants are identified by assessing segregation proportion of crossed plants (House, 1985). In addition, hot water is a method of emasculation which generates cytoplasmic male sterility for production of hybrid seed in sorghum (Hodnett and Rooney, 2018). Plastic bag is the method of anthers emasculation which uses polythene bag to cover the trimmed panicle of sorghum plant where the top part that initiates flowering and the bottom part of panicle are trimmed and covered to create high humidity and temperature which prevents dehiscence (Laxman, 1997). It is followed by the removal of polythene bag and allows crossing with pollen from the donor parent within three to four days. This depends on the efficiency of pollen death of the itended female plants before crossing (Laxman, 1997). This method is

useful when dealing with crossing of many plants within the limited time. However, the possibility of self-pollination is high if some pollen fail to die.

2.6. Molecular techniques for sorghum improvement

2.6.1. Genetic diversity of sorghum

The study of genetic diversity in sorghum is the basis of breeding materiels for development of new lines. The study of genetic diversity compares different genotypes that can generate elite lines and new varieties for food production and breeding programs in research institutes (Sinha and Kumaravadivel, 2016). Traits namely early maturity, disease resistance, grain yield and drought tolerance are considered when producing new genotypes to widen germplasm of the sorghum crop (Arunkumar *et al*., 2004). Landraces of sorghum exhibit moderate genetic diversity which is useful in the identification of diverse accessions which serve as parental lines for efficient utilization and application of germplasm into sorghum breeding programs (Burow *et al*., 2012). Assessment of genetic diversity of sorghum considers contrasting phenotypic and genotypic characters which can produce new seeds with high vigour and heterosis (Burow *et al*., 2012). One of the challenges facing genetic diversity of sorghum on the conserved germplasm is due to large number of accessions which need to be screened to obtain the elite genotypes (Casa *et al*., 2008). It is a challenge to screen for specific traits of interest which can bring impacts in the society. The proposed easy way is to group germplasm according to regional origin which could have significant correlation with adaptation to the trait of interest (Uphadyaya *et al*., 2009). Nevertheless, further studies to understand the genetic diversity of sorghum is imperative to increase possibility of obtaining new outstanding performing sorghum that can withstand harsh environment.

2.6.2. Molecular markers used in sorghum genotyping

Molecular markers have become the important tool in supplementing plant breeders to develop new varieties (Jiang, 2013). Markers show variation within and among genotypes and trace the QTLs/genes associated with the expression of certain traits of consumer preference (Platten *et al*., 2019). A number of studies have used molecular markers to study genetic diversity and traits related to crops improvement (Govindaraj *et al*., 2015; da Silva *et al*., 2017). For sorghum, a number of molecular markers have been utilized to identify the QTLs associated with stay-green, like delayed leaf senescence, leaf rolling, chlorophyll content, water use efficiency and yield. QTLs are mapped across the environments while others are specific to some environments (Hash *et al*., 2003).

2.6.2.1. Random Amplified Polymorphic DNA (RAPD)

RAPD is a type of molecular marker which is developed from PCR amplification of specific genomic DNA sequences recognized by random primers of arbitrary nucleotide sequence (Williams *et al*., 1990). RAPD markers are dominant and medium throughput thus; no needs to know the DNA sequence. They are simple, time saving due to its rapidity and requiring small amount of DNA (Welsh and McClelled, 1990). For this reason, RAPD markers are used to identify unknown species in crops and animals. Several studies on genetic diversity of wild and domesticated sorghum have revealed high genetic variation which may play as the bases of sorghum improvement (Agrama and Tuinstra, 2003; Akhare *et al*., 2003; Ayana *et al*. 2000). However, RAPD markers have low reproducibility which needs many primers that may not be good for genotyping of DNA materials in crops including sorghum (Van Haeringen, 2001).

2.6.2.2. Amplified Fragment Length Polymorphisms (AFLP)

AFLP markers refer to the markers that are generated from PCR and Restriction Fragment Length Polymorphism (RFLP) for amplification of DNA materials. AFLP amplification is selective to the subset of genomic restriction fragments. AFLP markers have good reproducibility compared to Inter-Simple Sequence Repeat, RAPD and RFLP markers (Costa *et al*., 2016; Paul *et al*., 1997). AFLP (Vos *et al*., 1995) and RFLP markers have polymorphism with high polymorphic information content (PIC) and Shannon diversity index. However, the cost is high for large-scale and locus-specific application which needs skilled person to run sophisticated modern equipment (Bradeen and Simon, 1998).The markers cannot differentiate homozygous and heterozygous individuals which make it cumbersome to achieve genotyping (Costa *et al*., 2016).

2.6.2.3. Simple Sequence repeats or Microsatellites (SSR)

SSR markers are DNA markers with short, tandem repeated di-, tri-, tetra-or penta-nucleotide motifs. The markers tandem arranged repeat units 1–6 bp long Di-, tri-and tetra-nucleotide repeats –(CA)n, (AAT)n and (GATA)n in a genome. It is characterized by high mutation rate between 10^{-2}-10^{-6} which makes them polymorphic markers and useful in genotyping compared to RAPD and RFLP and AFLP (Jenhan and Lakhanpaul, 2006). SSR primer design requires unique sub-set of flanking DNA to enable amplification of DNA fragments (Vieira *et al*., 2016). The amplicons show polymorphism which is attributed by allelic variation based on the number of repeat motifs in the microsatellite (Vieira *et al*., 2016). SSR markers are randomly distributed almost in the whole genome, co-dominant, high reproducibility, and high information content. This makes it a more important marker than ALFP, RAPD and RFLP markers. The marker is used for high throughput genotyping in plants. SSR markers require small amount of DNA about 100ng/sample for genotyping work. Microsatellites or Single Sequence Repeats (SSRs) are extensively employed in plant genetics studies, using

both low and high throughput genotyping approaches. On the other hand markers need experienced labour and high cost for automated work (Vieira *et al*., 2016). SSR markers have high polymorphic information content (PIC) and Shannon diversity index. SSR markers are useful for genetic studies in plants including mapping QTLs associated with traits of interest, genotyping of plants, estimation of relationship of genotypes, designing linkage group and marker assisted selection studies (Kalia *et al*., 2011). SSRs have been used in the genetic diversity of sorghumare. The SSR markers which indicate high polymorphism for STG are used as candidate markers for mapping STG QTLs which are closely linked to genes associated to drought tolerance (Edema *et al*., 2015). SSRs are used by plant breeders to find distant related STG trait sorghum for crossing to develop new sorghum lines (Mwamahonje et al., 2021)

2.6.2.4. Inter-simple sequence repeats (ISSR) markers

ISSR markers are multilocust markers which are generated by the amplification of DNA segments using microsatellite sequence primers (Reddy *et al*., 2002). ISSR markers have high efficiency compared to RAPD which has low reproducibility and AFLP that has higher cost of genotyping (Meyer *et al*., 1993, Gupta *et al*., 1994, *Wu et al*., 1994, Zietkiewicz *et al*., 1994). ISSR markers are designed by the amplification of ISSR sequence of DNA that is flanked by microsatellite sequences in two sides. The PCR amplification of the ISSR regions uses single primer to multiply the amplification products which are useful for the studies on genetic diversity, phylogeny and gene tagging on different crop species including sorghum (Ng and Tan, 2015; Satish *et al*., 2016). The ISSR markers are highly polymorphic, thus are used for genome mapping and evolutionary biology in different living organisms (Yang *et al*., 1996; Reddy *et al*., 2002). The markers are dominant, powerful, rapid, affordable and methodologically not sophisticated that can be handled by normal skilled person compared to

other dominant markers Gupta *et al*., 1994; Wang *et al*., 1998). Nonetheless, the markers segregate as co-dominant in some cases which can distinguish between homozygous and heterozygous alleles thus make it useful for crop improvement (Wu *et al*., 1994; Wang *et al*., 1998; Sankar and Moore, 2001). The markers are important for the study of genetic marker to new learners and for organisms which lack genetic information (Ng and Tan, 2015). ISSR markers have been used to determine genetic distance estimates of maize genotypes for introgression to develop new germplasm. These apply in other cereal crops like pearl millet, barley and finger millet for generating new lines (Idris *et al*., 2012). Therefore, ISSR markers can be used for the mapping of stay green QTLs in sorghum.

2.6.2.5. SNPs Markers

SNP markers show high level of polymorphism due to the high content in the genome, is not time consuming compared to other markers however, need skilled person and high cost at beginning for automated operation (Jenhan and Lakhanpaul, 2006). Molecular marker tools are growing; recently, SNPs markers have become the important marker because they are simple for automation data production and they are available almost in the whole genome. SNPs have ability to detect traits which other markers cannot because of abundance across the genome, ubiquitous and high throughput automation (Mammadov *et al*., 2012). SNP markers group the allele- specific molecular reaction products; separate and detect allele specific products for easy identification which SSR markers cannot do (Vignal *et al*., 2002). SNP markers are used in mapping QTLs associated with the traits of interest to improve the traits. This is due to availability of SNPs in the entire genome such that generates clear map resolution which clearly show polymorphism (Mammadov *et al*., 2012).With high variation present between and within hybrid and local accessions, characterization is important to

identify suitable genotypes for use in agriculture and preserving in the gene bank for future use (Akhare *et al*., 2008).

Regarding the difficulties to differentiate among sorghum accessions, SNPs do well, using these sweet sorghum accessions for instance, have been grouped into three major groups including; historical and modern genotypes for syrup production, modern genotypes mainly for sugar supply and amber genotypes (Murray *et al*., 2009). SNP has current at the peak due to high abundance and can accommodate the whole genome of sorghum with high throughput and high resolution compared to others. This marker can identify the diversity to single base level (Disasa *et al*., 2016). SNP markers can indicate the variability on a single nucleotide in the DNA present in the genome. Due to high efficiency, the number of SNPs and variations available in all regions of plant genome are detected (Sakiyama *et al*., 2014). SNPs markers can be exploited from genomic or from the expressed sequence tag sequences by high throughput sequencing technology, they can be obtained in the PCR products (Calviño *et al*., 2009).

2.6.2.6. Diversity Arrays Technology

Diversity Arrays Technology (DArT) refers to a technology which applies in the study of molecular genetics for the development of sequence markers for genotyping and genetic analysis. It is a hybridization based approach which can run at least 1000 of genomic loci in parallel (Jing *et al*., 2009). The technology offers a high multiplexing level with reasonable cost (Mace *et al*., 2008). DArT has become more applicable in several plant species than other technologies because of high throughput, genomic coverage and transferability (Boczkowska *et al*., 2020). Despite the fact that the different molecular markers have been useful in the study of genetic diversity, genome mapping, and marker assisted selection, they differ in the effectiveness. Moreover, the cost of genotyping using SSR and SNP markers are

based on the data point run. Therefore, the cost of investment using these markers is high. This have affected plant breeding programs due to large number of data set which most plant breeders cannot afford to pay. DArT involves the whole genome sequencing which identify different coding regions of the genome in various crops. The technology has developed DArT markers which are used for mapping QTLs of farmers preferred traits in sorghum for improvement. The QTLs are located in the specific position of chromosomes. DArT assays generate whole genome fingerprints by scoring the presence versus absence of DNA fragments in genomic representations generated from genomic DNA samples through the process of complexity reduction. DAarT markers are used for genotyping in sorghum, barley, wheat, plant height, drought tolerance and yield (Fiust *et al*., 2015; Mace *et al*., 2008; Sabadin *et al*., 2012). Gy9 is an examples of QTLs for yield that have been mapped in sorghum using DAarT markers while QTLs for drought tolerance was mapped in the chromosome number 4A in wheat (Ballesta *et al*., 2018).

2.6.3. Maker-assisted backcrossing of sorghum

Most traits of interest in the sorghum crop have been difficult to exploit through conventional breeding (Prohens, 2011). MABC can transfer alleles at one or more loci from the donor parent to the recurrent parent using molecular markers (Hasan *et al*., 2015; Saxena *et al*., 2002). Backcrossing for drought tolerance in sorghum is tedious and time consuming. SSR and SNP markers have been mapped for STG QTLs in sorghum. These markers are used for marker assisted backcrossing for the improvement of STG in sorghum. MABC simplifies the breeding programs by reducing the breeding cycles which shortens the duration of releasing new improved varieties. The introgression of alleles of preferred traits can be attained after six backcrosses with genome recovery of recurrent parent by 99.2% (Hasan *et al*., 2015). The recurrent parent genome is recovered by half of each backcross for instance, two backcrosses

recover 87.5%. Plant breeders use marker assisted backcrossing as the tool for selecting traits which contain alleles with high recovery of recurrent parent genome. It helps to compare phenotypes and genotypes of large sorghum populations with consistence across the environment; nevertheless, QTLs are influenced by variation in environmental factors (Saxena *et al*., 2002). Testing the effect of water stress on sorghum across environments is important. Researchers have concentrated on developing new varieties which are drought-tolerant because of climate change occurring in most parts of the world. To achieve exploitation of QTLs from the donor parents to recurrent parents, a large number of polymorphic, high resolutions, high throughput, co-dominant and informative molecular markers are used (Baloch *et al*., 2017). This helps to identify genes associated with complex drought tolerance due to small genome size of sorghum (Paterson *et al*., 2009). By using molecular markers, a number of sorghum linkage maps associated with drought tolerance have been identified (Mace *et al*., 2012). Molecular markers for donor and recurrent parents play a major role for elimination of linkage drag to enhance the recovery of recurrent parent's genome. QTLs for STG originated from donor parent B35 and E 36-1 are commonly used for introgression into sorghum lines (Borrell *et al*., 2014; Edema and Amoding, 2015). In addition, epistatic interactions within and between STG loci have been reported in sorghum genome, these interactions within the phenotypes can be exploited by the use of Near-isogenic lines (NILs) and applied in the sorghum breeding program. MABC remains the important tool for introgression of traits of interest from the donor parent to the recurrent parent for sorghum improvement.

MABC involve Foreground and background selection, foreground selection refers to selection of molecular makers associated with the donor parent in particular target locus. The aim of foreground selection markers in MABC is to maintain locus of target in heterozygous state of both parents up to final backcrosses (Gorthy *et al*., 2017). Foreground selection is

achieved by marker-assisted backcrossing. Some of the traits which have been introgressed into recurrent parents by backcrossing include drought tolerance, high yielding, shoot fly resistance, and *Striga species* resistance in sorghum (Ouedraogo *et al*., 2017). Foreground selection markers mSBCIR238, Xtxp72, mSBCIR222, mSBCIR314, Xtxp225 and Xtxp285 have been mapped for STG using SSR markers (Ouedraogo *et al*., 2017). High yield is a complex trait contributed by several QTLs in chromosome 1, while sorghum flowering QTLs are located in chromosome2 (Sukumaran *et al*., 2016). These QTLs together significantly contribute to enhance grain yield in sorghum. Background selection reduces donor parent alleles increasing the recovery of the recurrent parent genome. Background selection occurs when trait of recurrent parent is recovered while receiving new specific traits from the donor parent for enhancement (Gorthy *et al*., 2017). It aims to minimize the size of introgressed region, to remain with the target gene following selection of non-deleterious loci of particular traits during backcrossing. It is achieved by identifying the best recombinants between the genes of target which are closely linked to the markers (Hospital, 2001). Single nucleotide polymorphism markers could work better to detect linkage groups due to higher efficiency compared to SSR markers. Figure 2.1 illustrate basic procedures to follow when improving trait of choice from the donor parent to preferred recurrent parents in sorghum. MABC focuses on specific trait when introgressing to the recurrent parent for instance drought tolerance, disease and pest resistance and *Striga* tolerance. It involves phenotyping and genotyping the genotypes to develop new lines with high proportion of trait of target (Kamal *et al*., 2017).

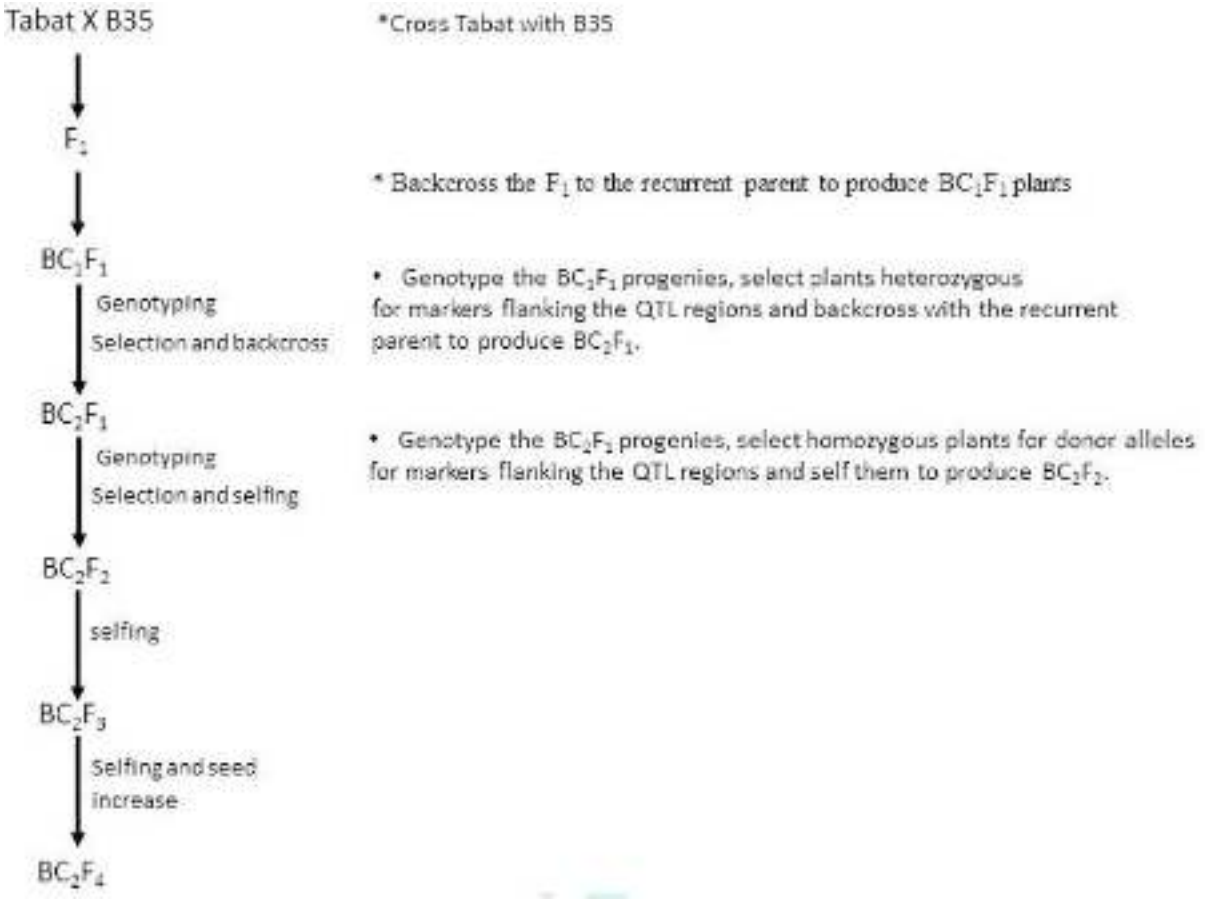


Figure 2. 1. Marker assisted backcrossing procedures for introgression of stay green QTLs from the donor parent B35 to the recurrent parent Tabat

2.7. QTL mapping for yield in sorghum

QTL mapping is the exploitation of QTLs associated with genes coding for expression of a trait of interest using molecular markers. For instance, the yield-related components in sorghum are conferred by a number of genes (Hamidou *et al*., 2018). These genes are associated with QTLs which express agronomic-related traits, such as yield components and abiotic stress tolerance. These QTLs are traced using molecular markers. Molecular markers are useful for mapping of QTL located close to the genes expressing the traits. The markers which are successful and lead to identification of QTL of agronomic traits of importance are recorded as the markers of that QTL (Zou *et al*., 2012). Marker assisted selection has a role to

play for the improvement of sorghum yield. During optimization, only markers which are tightly linked with candidate QTLs for high yield are screened for sorghum improvement by marker assisted selection (Fakrudin *et al*., 2013). The study of genetic diversity focusing on characters of drought-tolerant, yield and yield related traits such as plant height, number of leaves, root biomass is important to increase heritability and genetic gain for sorghum improvement. A number of QTLs for yield and yield related traits have been exploited for sorghum improvement. For instance, PSTOL1 and Sb07g02840 genes are associated with increase of root diameter which enhance sorghum grain yield. The QTLs Gy-3, SA2–3 located in sorghum chromosome 3 (SBI-03), Gy/SA2–3, at position ~ 71Mb in SBI-03; and Gy/RD-7 QTL at 3.6Mb in SBI-07) are linked to yield related traits such as root morphology and surface area of fine roots which help to increasing yield in sorghum (Bernardino *et al*., 2019). In addition, Reddy *et al*. (2013), found 3 QTLs for sorghum yield in SBI-09, 1 in SBI-04 and 1 in SBI-06. According to Reddy *et al*. (2013), at least one QTL for panicle was detected on SBI-09, SBI-04 and SBI-06. QGy-dsr06-1 is the major QTLs of panicle weight in sorghum contributing 11.4% of phenotypic variance. QTL qYLD1.1 in SBI-01 enhances sorghum grain yield during water stress and moisture conditions. Such QTLs should be incorporated to sorghum lines which lack them for sorghum yield improvement. Yield trait is controlled by several QTLs available in every chromosome in sorghum (Kapanigowda *et al*., 2013; Rajkumar *et al*., 2013; Reddy *et al*., 2013; Shehzad and Okuno, 2015). Of the QTLs which have been exploited enhancing sorghum yield, 20% of them are allocated in SBI-01 (Mace and Jordan, 2011). This calls for further research to explore new QTLs which are useful for improvement of yield than the current QTLs. Plant height, number of tillers, panicle weight, grain weight, stay green are associated with grain yield in sorghum (Mace and Jordan, 2011; Reddy *et al*., 2014). Nevertheless, further studies on mapping QTLs for

yield and yield component require more attention to exploit specific QTLs which are closely linked to the gene coding for yield to maximize yield.

2.8. QTLs pyramiding for drought tolerance sorghum

QTL pyramiding involves crossing of one near inbred line (NIL) to other composing different useful QTLs by subsequent marker-assisted selection to produce new lines with both advantageous traits. The transfer of traits of interest from the donor parents to the recipient is recommended for the same species (Takeda and Matsuoka, 2008). QTL pyramiding improves drought tolerance in sorghum compared to introgression lines. For instance, introgression lines with Stg 1 and 2 reduce yield by 23% in water stressed trial compared to fully irrigated trial. The combined stay-green introgression lines with pyramided Stg 1 and 2 introgression lines reduce yield by 11% in water stressed trial compared to fully irrigated trial (Kamal *et al*., 2017). Pyramiding involves combination of QTLs with different efficiency to enhance the expression of stay-green traits (Sanchez *et al*., 2002). Use of molecular markers helps to detect QTLs which could be pyramided to improve drought tolerance in sorghum (Harris, 2007). Pyramiding of QTLs may fail to improve stay-green in sorghum due to incompatible gene action, which does not allow expression of intended traits (Kassahun *et al*., 2010). The success of pyramiding is subject to genetic architecture and correct mapping of QTL of the trait (Takeda and Matsuoka, 2008). During QTLs pyramiding, only the best expression of trait of interest is selected for further evaluation in multi-location trials before approval as new varieties. Studies have suggested that, mapping of QTLs for expression of stay green using molecular markers are important for pyramiding to enhance heritability of new lines (Sakiyama *et al*., 2014). Every QTL contributes a small percent to enable expression of the stay-green trait which needs pyramiding to assemble QTLs for expression (Kassahun *et al*., 2010; Sanchez *et al*., 2002). Pyramiding of stay green QTLs during pre-flowering and post-flowering drought in sorghum increases chance of developing new promising lines with

valued traits by marker assisted backcrossing (Gorthy *et al*., 2017; Kabede *et al*., 2001). Other traits which have been suggested to be exploited by pyramiding method are Striga resistance (Ali *et al*., 2016), high yield and plant height. These findings assist plant breeders to map the potential QTLs of traits that are introgressed and can be expressed after pyramiding (Ongom, 2016). To achieve pyramiding, efficiency molecular markers for mapping QTLs need to be exploited to facilitate molecular breeding programs in sorghum. With gradual increase of temperature and drought especially in semi-arid areas, pyramiding QTLs for drought tolerant and yield is recommended (Kadam and Fakrudin, 2017). It should be noted that some of the pyramided QTLs do not express phenotypically, therefore, only pyramided QTLs which express their traits are recommended for selection in sorghum improvement (Kassahun *et al*., 2010).

2.9. Combining ability for addressing tolerance in sorghum

Combining ability is defined as the ability of the parent lines to combine during crossing and transmitting traits of interest to progenies (Fasahat *et al*., 2016). There are two categories of combining ability, general combining ability which deals with additive genetic variance and specific combining ability which deals with non-additive genetic variance (Sory, 2015). Combining ability is determined in parents and their crosses. Some of the traits of interest evaluated in these combinations include high yield, disease resistance, plant height, early maturity and drought tolerance (Tadesse *et al*., 2008). The parent crosses with good combining ability of progenies at different or specific environment are selected for release or further advancement (Assefa, 2012). Drought is one of the constraints most stressing sorghum productions. The best parents for stable combining ability are the base for addressing drought challenge is sorghum (Assefa, 2012). This is an agreement with Tadesse *et al*. (2008) who commented that, the adoption of sorghum hybrid is one of the goals to

improve drought tolerance in sorghum. Crossing different parent lines develop hybrid sorghum which can tolerate drought stress. To achieve sorghum hybrid development, a successive screening the best parents for drought tolerance and yield is important. MABC is one of the options to identify parents which produce genotypes that can cope with drought stress and other yield related traits in sorghum. The gene actions for enhancing the heritability of drought and high yielding traits have been reported by Sory (2015). This can be confirmed by use of molecular markers to identify the QTLs/genes associated with traits of interest.

CHAPTER THREE

3.0. FARMERS' PRODUCTION CONSTRAINTS AND TRAITS PREFERENCE OF SORGHUM IN CENTRAL TANZANIA

3.1. Introduction

Sorghum is commonly grown in sub-Saharan Africa for food. The crop is grown in drought prone areas which are characterized by low annual rainfall (Mavhura *et al*., 2015). In East Africa, Tanzania is leading in sorghum production followed by Uganda (Tenywa *et al*., 2018). Sorghum is cultivated as small scale agriculture by most of smallholder farmers in Dodoma, Singida, Mara, Simiyu, Tabora and Shinyanga regions (Brown, 2013). Sorghum is grown for food and animal feeds as the source of energy and minerals and it is gluten free (Dial, 2012). However, in Tanzania, the production is below 1 ton per hectare compared to mean yield of 1.3 tons per hectare in East Africa (Mrema *et al*., 2017). Efforts have been made to increase sorghum production by importing new varieties into Tanzania. Nonetheless, only 5% of the local farmers have adopted these new varieties and the productivity has remained stagnant (Orr *et al*., 2016). Low yield advantage to some of the improved sorghum varieties versus landraces causes a low adoption rate (Smale *et al*., 2018). Nevertheless, sustainable exploitation of potentials in sorghum is low as the crop is drought-tolerant and can, therefore, be cultivated in drought-prone areas to safeguard food security for the people and livestock (Wagaw, 2019). Apart from drought, poor soil fertility persists in sorghum farms in semi-arid regions of Tanzania. This is caused by high soil erosion, deforestation, burning of vegetation, poor agricultural practices, and unpredicted rainfall (Omoro, 2013). The depletion of nutrients such as nitrogen, phosphorus, and potassium is high, but the application rate of fertilizers in Tanzania is 15.9 kg/ha, which is far below the average world

recommendation of 162 kg/ha (FAO, 2018). Socio-economic activities have impacts on the adoption of improved sorghum varieties. Gender, age and education can either negatively or positively enhance adoption of technologies. Access to land, cultural uses, performance of varieties influence farmers adoption (Diale, 2011). Farmers' traits preferences determine type of varieties to develop to enhance the adoption of improved sorghum seeds. Sorghum farmers lack knowledge to differentiate improved and local sorghum varieties' traits performance because of poor agronomic practices of crop. There is a gap on adoption rate of improved seed; most farmers grow local varieties which yield is low. There is a gap of technologies to control pests and diseases and other agronomic practices suited for sorghum production.

Therefore the aims of this study were to

i. determine constraints of sorghum production in the study area,
ii. identify sorghum traits preferred by farmers in Central Tanzania, and
iii. determine coping strategies for drought by farmers in the study area

3.2. Materials and methods

3.2.1. Study area

The study was conducted in Dodoma and Singida regions of Tanzania where sorghum is the main crop cultivated. The Dodoma region was represented by the Kongwa district (6.200°S 36.417°E/-6.200; 36.417) and the Singida region was represented by the Ikungi (-5°07'60.00" S 34°45'59.99" E) and Iramba (-4°25'16.14" S 34°18'9.65" E) districts. Temperature ranges between 25-32^{O}C, whereas rainfall ranges from 400-600 mm annually. Rainfall starts in December to the end of April. The areas are characterised by dry spell, which lasts for 2–4 weeks during rainy season each year. These regions are located in the centre of the country, where soil fertility is low and soil erosion is high. The representative villages in this study

were purposively selected as sorghum producers including Laikala (-6.193647, L36.617899), Sagara (-6.24983848, L36.55242241), Msambu (4° 20′ 0″ S, 34° 10′ 0″ E), and Nkonkilangi (4° 15′ 0″ S, 34° 12′ 0″ E). Areas selected for this study in Tanzania are shown in Figure 3.1.

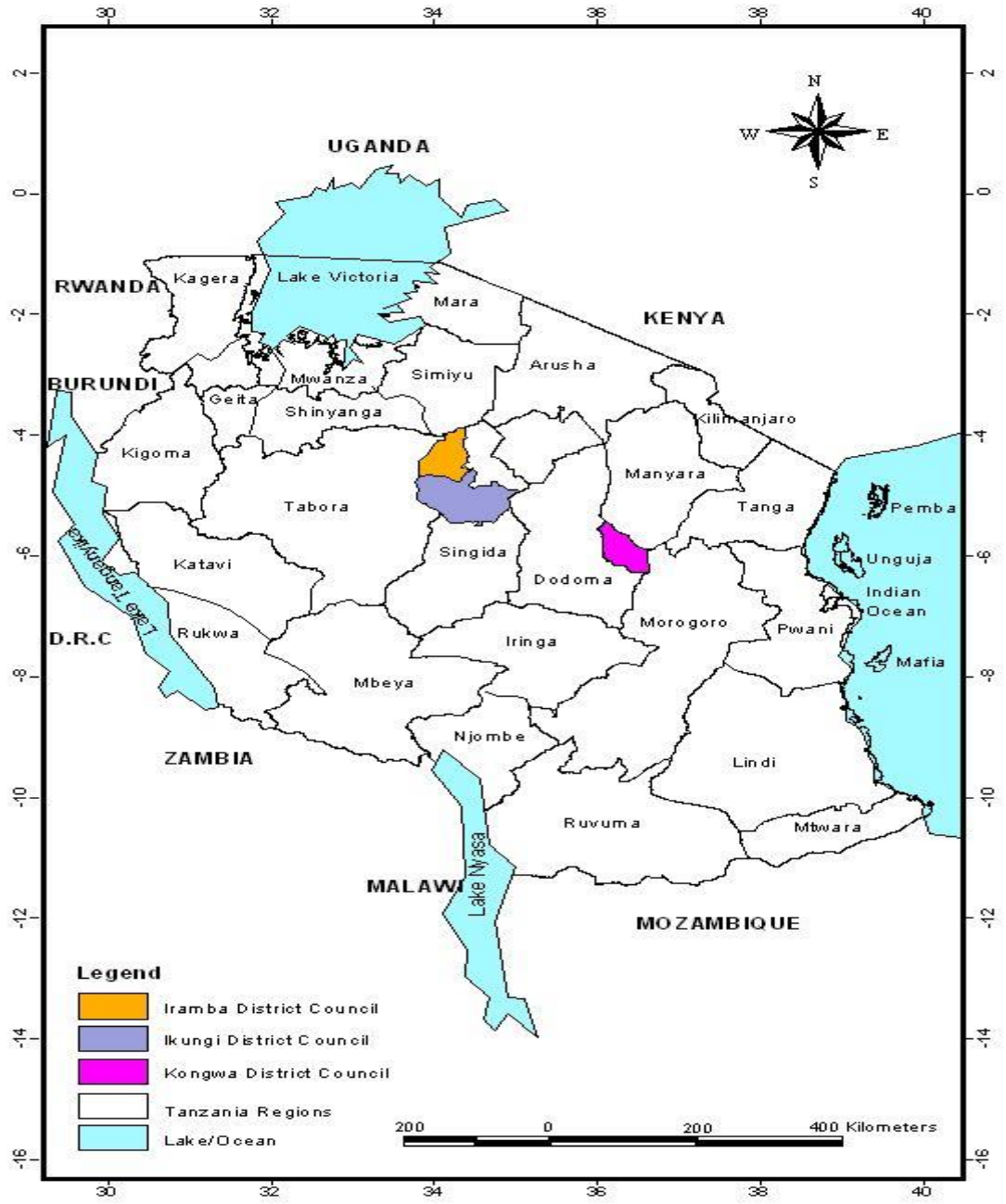


Figure 3. 1. Map showing areas purposively selected for PRA survey in Tanzania

3.2.2. Sampling and respondents

Two regions of Dodoma and Singida, two districts (Iramba and Ikungi) from Singida region and one district (Kongwa) from Dodoma region, were purposively selected for this study

based on the sorghum production, importance of sorghum to many households as food security crop and number of sorghum producers within the districts. Each district was represented by two villages which were randomly selected. Ten experienced sorghum farmers per village (total 60 farmers for six villages) were randomly selected to participate in the focus group discussions facilitated by the Village Chairman (VC) and Agricultural Extension Officer (AEO). About thirty farmers per village, for a total of 180 farmers, were randomly selected for individual interviews. Supplementary information was collected from key informants in each village. The sample size formula used for obtaining study sample was as follows (equation 1):

$$n = no / [1 + (no - 1) / N] \quad (1)$$

Where: n is the sample size, N is the population size, and no is the calculated sample size for an infinite population (Cochran, 1963). Various participatory rural appraisal (PRA) tools were used including focus group discussion, individual interviews, and preference ranking with the total of 240 farmers.

3.3. Research design and data collection

A cross-sectional survey approach was used to collect data at a single time point (Kothari, 2004; Saunders *et al*, 2007). Semi-structured questionnaires and checklists were used to collect data from 180 farmers in households, 60 farmers in the focus groups discussion, and key informants. The information obtained from the focus group discussions and other observations was cross-checked and confirmed with a semi-structured questionnaire. Finally, biographic data were recorded for each respondent.

3.4. Statistical data analysis

Data were processed with SPSS v. 20 (IBM Corp., Armonk, NY, USA). Descriptive and inferential statistics were used for data analysis. Comparisons of the Mean yields of improved and local varieties were estimated by use of the one-way Analysis of Variance (ANOVA). The data analysis on improved sorghum varieties (%) cultivated by farmers per district, constraints of sorghum production and coping strategies proposed by farmers to overcome drought were subject to determination by chi-squares in SPSS software.

The factors influencing the productivity of improved sorghum varieties were estimated by employing the multiple linear regression analysis models as shown below (model equation 2):

$$Y = B_0 + B_1X_1 + \ldots + B_nX_n + \mathcal{E} \qquad \ldots\ldots\ldots\ldots(2)$$

where Y = dependent variable, X = independent variable, B0 = constant value of Y if values of X = 0, n = number of independent variables, and B1 to Bn = estimate of effects of X on Y as X increases by one unit; and Ɛ= error term for the unknown variations in dependent variable Y. On the other hand, factors influencing the improved sorghum variety adoption rates were determined by using a binary logistic regression analysis model below (Model equation 3) in accordance to Wuensch (2020), $Ln(Odds)=ln[p/(1-p)] = E(Y) = \alpha + \beta x$ (3) where Ln is the natural logarithm, p=the predicted probability that farmers adopted the production of improved sorghum varieties, whereas 1-p = the probability that farmers didn't adopt, and p/1-p is the probability of the odds for adoption. But Y is the dependent (outcome) variable and X is the predictor of the factors affecting improved sorghum variety adoption, α is the constant value of Y when all values of X = 0, while β is the estimated effect of X on Y.

3.5. Results

The number of farmers who participated in the interview was balanced by half between males and females per villages in each district (Table 3.1). The analysis of socio economic

characteristics of smallholder farmers growing sorghum in the study area, indicated that, females (51.7%) had higher participation in sorghum production than males (48.3% and farmers with age from 40-50 years played a major role in farming activities while 18-20 years participated least (Table 3.2). The family size had impact on levels of sorghum production by farming families. A family with 7-8 members showed high sorghum production (27.8%) while family with 1-2 members showed the least production (6.8%) (Table 3.2). Out of the 180 farmers interviewed about farm size, 28.4% had above 18.125 hectares while 10.2% had farm size from 14.375-18.125 hectares.

Table 3. 1. Number of males and females participants at two villages in each district

Village	Male	Female	Total	District
Laikala	20	20	40	Kongwa
Sagara A	20	20	40	Kongwa
Msambu	18	20	38	Ikungi
Nkuninkana	18	20	38	Ikungi
Nkonkilangi	20	19	39	Iramba
Mseko	20	19	39	Iramba

Table 3. 2. Socio economic characteristics of smallholder farmers growing sorghum in Central Tanzania

Variable	Frequency	Percent
Gender		
Male	85	48.3
Female	91	51.7
Age category		
18-28 years	22	12.5
29-39 years	38	21.6
40-50 years	53	30.1
51-60 years	37	21.0
Above 60 years	26	14.8
Family size category		
1-2 members	12	6.8
3-4 members	37	21.0
5-6 members	45	25.6
7-8 members	49	27.8
9-10 members	12	6.8
Above 10 members	21	11.9
Farm size category		
1.25-5.0 hectares	42	23.9
5.625-9.375 hectares	26	14.8
10-13.75 hectares	40	22.7
14.375-18.125 hectares	18	10.2
Above 18.125 hectares	50	28.4

3.5.1. Sorghum farm size

Majority of farmers (59%) cultivated 0.2-0.8 hectares per farmer followed by 17% who cultivated 0.9-1.5 hectares. Six percent of farmers cultivated above 2.9 hectares (Figure 3.2).

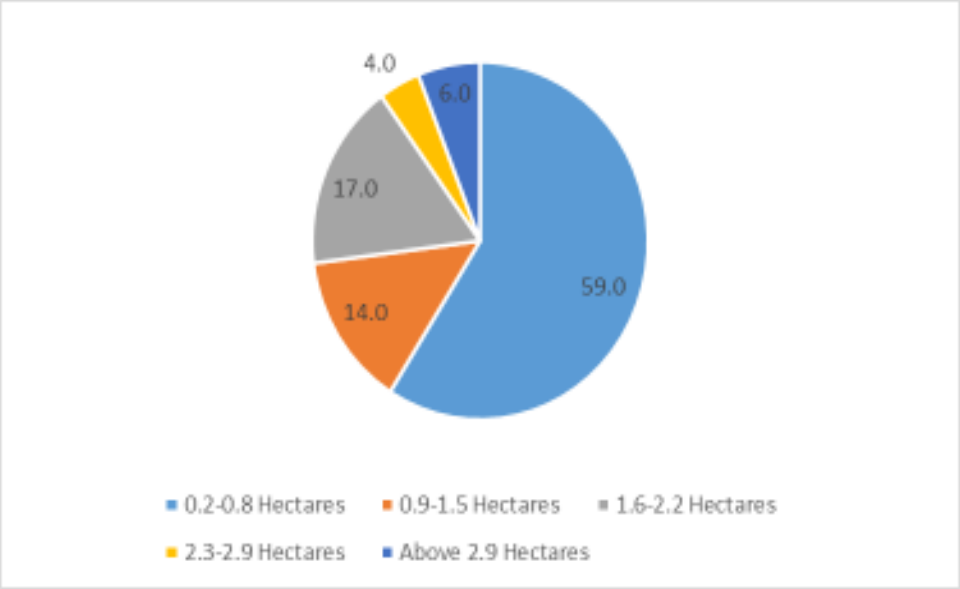


Figure 3. 2. Sorghum farm size distribution in study area

3.5.2. Types of crops grown by farmers in the study area

The major crops cultivated by farmers were sorghum (86.3%) followed by maize (53.1%), pearl millet (37.7%). The least crops cultivated by farmers included grapes (0.6%), bambara nut (1.1) and cowpea, 1.7%. Notwithstanding, maize was given the first priority as food crop. Pearl millet and sweet potatoes used as alternative source of food while sunflower (36.6%) was used as the main cash crop (Table 3.3). Apart from crops above, farmers kept cattle, chicken, sheep and goats for household's income generation.

Table 3. 3. Types of crops grown in the study areas

Crop type	Responses	Percent
Sorghum	151	28.7
Maize	93	17.7
Pearl millet	66	12.5
Sunflower	64	12.2
Sweet potato	56	10.6
Groundnut	37	7
Rice	35	6.7
Cassava	11	2.1
Bambara nut	7	1.3
Cow pea	3	0.6
Finger millet	2	0.4
Grape	1	0.2

3.5.3. Sorghum varieties cultivated by farmers

About 81.3% of farmers interviewed cultivated local sorghum varieties, 6.8% cultivated improved varieties the rest cultivated both. The reasons for growing local varieties were due to availability and accessibility of seed, pest and disease resistance, and food and market demand of local seeds. The reasons for growing improved varieties were due to early maturing, high yielding and appealing colour of improved sorghum seed. Findings revealed that, farmers in Kongwa, Ikungi and Iramba districts mainly cultivated two improved sorghum varieties, Tegemeo and Serena. Serena was cultivated by 23.2%, 16.4%, and 28.8% in Kongwa, Iramba and Ikungi districts, respectively, while 35.7%, 4.9% and 5.1% cultivated Tegemeo variety in Kongwa, Iramba and Ikungi districts, respectively. Farmers in Kongwa districts cultivated the largest number (8) of improved varieties while the adoption rate was very low in Ikungi district (Table 3.4).There were differences in the cultivation of improved varieties among districts, particularly Tegemeo, Macia, NACO-Mtama 1 and Pato. The

cultivation of improved sorghum varieties was the highest in Kongwa district and the lowest in Iramba district. The list of local sorghum cultivated by farmers is presented in Figure 3.3.

Table 3. 4. Improved sorghum varieties (%) cultivated by farmers by district

Variety name	Percentage of improved varieties cultivated per district			Chi-Square
	Kongwa	Iramba	Ikungi	
Serena	23.2	16.4	28.8	0.266
Tegemeo	35.7	4.9	5.1	0.000***
Macia	32.1	1.6	0.0	0.000***
NACO-Mtama 1	17.9	1.6	0.0	0.000***
Pato	16.4	0.0	0.0	0.000***
Okoa	3.6	0.0	1.7	0.329
Sila	3.6	0.0	0.0	0.114
Wahi	0.0	1.6	1.7	0.623
Lulu	0.0	3.3	0.0	0.149
Pirila	1.8	0.0	0.0	0.340
Hakika	0.0	1.6	0.0	0.388

*** Significant at $P < 0.001$

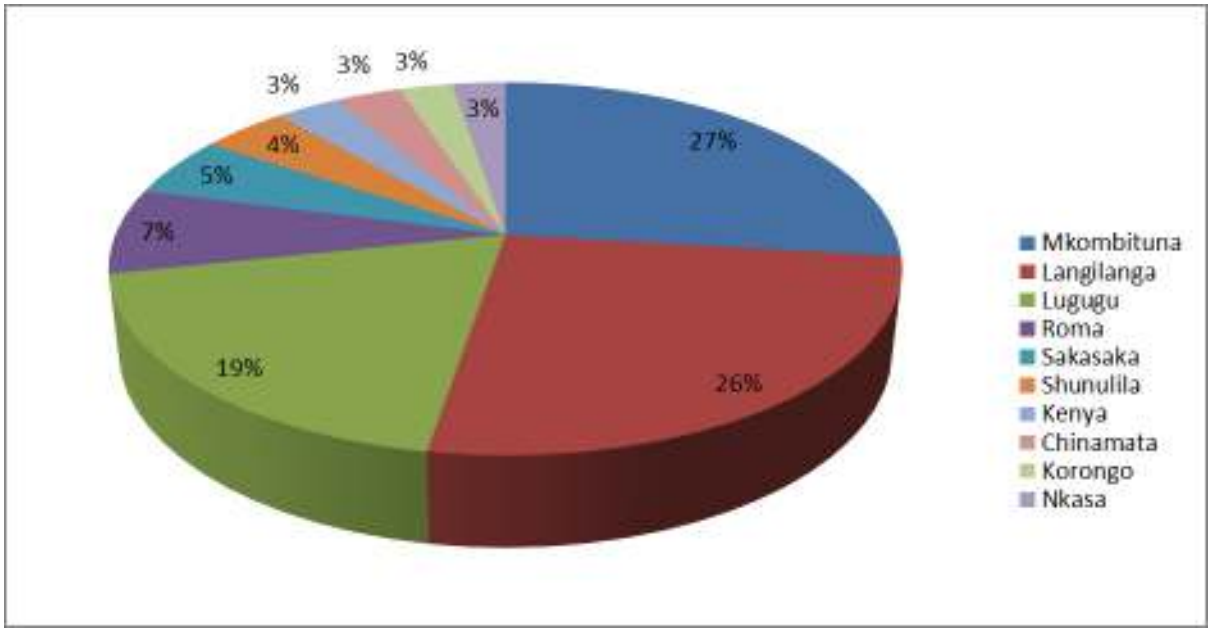


Figure 3. 3. List of local sorghum varieties commonly adopted by farmers

3.5.4. Source of sorghum seed adopted by farmers

The main source of sorghum seed was own saving (89.8%), 5.7% farmers had access to seed from Agro-dealers. Although, extension officers are responsible to coordinate farmers on the adoption of improved technologies, 0.6% of farmers depended on extension officers to get seed (Figure 3.4).

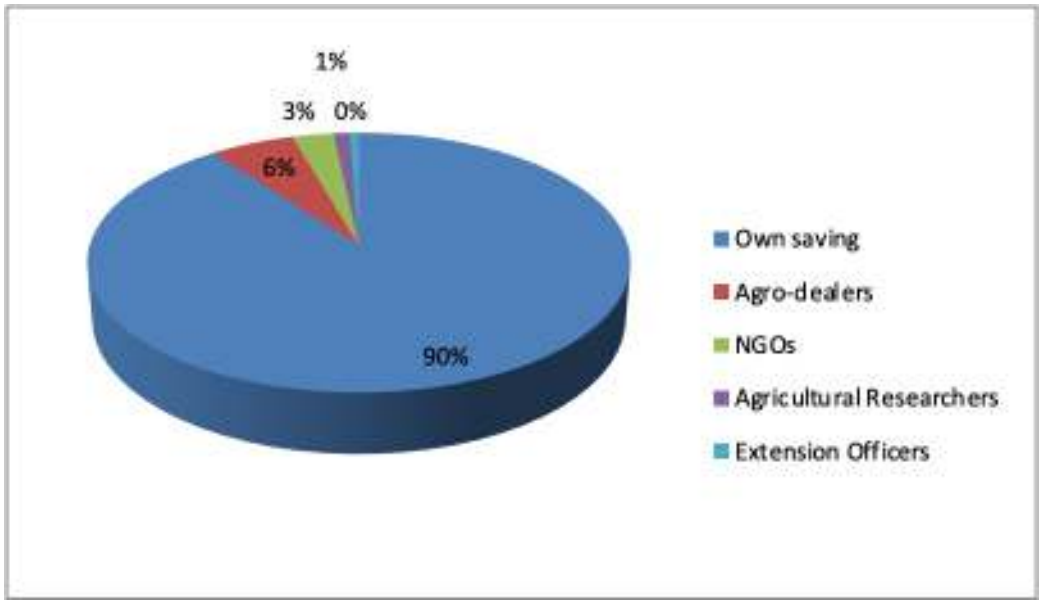


Figure 3. 4. Source of sorghum seed

3.5.5. Constraints facing sorghum farmers

Farmers cited various constraints on sorghum production in their villages. Of these, bird damages and poor soil fertility were reported as major constraints by at least 55% of respondents in each district followed by drought (Table 3.5). Farmers reported that, rainfall had been decreasing annually, resulting in prolonged droughts which affected grain filling during the post-flowering period. It was observed that, limited knowledge of fertilisers, lack of capital to buy them, and few subsidies from the government contributed to soil erosion and

low soil fertility. It was noted that, skills and knowledge of soil and fertility management is needed by farmers to maximize the production and conservation of biodiversity. Despite low soil fertility within the three districts, yet farmers did not use manure from their livestock to support plant growth which could enhance the final yield.

Farmers in all districts complained that, pests and diseases were critical sorghum production constraints (Table 3.6). The risk of these was highest in Iramba (47.5%) district. The pests in sorghum included *Busseola fusca*, *Rhopalosiphum maidis* and *Spodoptera frugiperda*, whereas the common disease was the *Sphacelotheca reiliana*; reported in all three districts. Prevalence of wide range of diseases and pests was the problem which faced farmers as they lacked prior experience in identifying and naming other sorghum diseases. According to the focus group participants in Kongwa, Ikungi and Iramba districts, pests and diseases for sorghum caused great losses. Of the constraints reported drought was reported to be the most common constraint, followed by lack of knowledge on the good agronomic practices related to sorghum field management. The low adoption rate for the new technologies reported by the key informants specifically extension officers, included: perception of crop growers that sorghum is a minor crop, and hence gave it low priority. Low prices of sorghum produce output and lack of reliable markets as well as limited number of products from the same crop. On part of improved sorghum production adopters, the main constraint was lack of extension services emanating from limited extension staff contacts.

Furthermore, sparsely distributed rainfall was reported as constraint by participants in all districts; however, the concern was highly reported in Iramba district. The participants reported that the shortage of rainfall resulted in drought stress during the post-flowering stage. In these areas a dry spell for 2–4 weeks was common such that, post-flowering and grain filling in sorghum were greatly affected. In Iramba district farmers cited fertiliser shortage to be among the challenges to sorghum production due to lack of input subsidy as it

is for other grain crops such as maize, cashew nut, and cotton which the government used to subsidize. Another constraint reported by farmers of Nkonkilanga in Iramba district was that limited number of tractors which reduced their capacity to adopt mechanization and consequently, used hand hoe and ox ploughs which do not cultivate well and thus lead to run short of timely land preparation, planting and harvesting their sorghum crop. Due to rainfall challenges, majority (96%) of the farmers had the need for seeds of drought tolerant sorghum varieties, especially those which do better in the period of late January and February months.

Table 3. 5. Constraints of sorghum production

Constraint	Percentage of constraint per district			Chi-Square
	Kongwa	Iramba	Ikungi	
Birds	96.4	68.9	94.9	0.000***
Problem of market	1.8	4.9	1.7	0.482
Poor soil fertility	58.9	86.9	55.9	0.000***
Drought	33.9	63.9	57.6	0.003**
Pest and diseases	8.9	47.5	18.6	0.000***
Poor agronomic management	14.3	19.7	13.6	0.607
Lack of improved varieties	17.9	23	20.5	0.792
Shortage of fertilisers	5.4	8.2	22	0.012*
Poor mechanization	1.8	3.3	2.03	0.808
Scarce of land	7.1	0	13.6	0.013*

*, **, *** Significant at $P < 0.05$, $P < 0.01$, and $P < 0.001$, respectively.

Table 3. 6. Common pests and diseases affecting sorghum

Village	Pest	Effect	Disease	Effect
Sagara A	Army worm	High	Head smut	Medium
	Stem borer	High		
	Birds	High		
Laikala	Crickets	High	Head smut	High
	Stem borer	High	Leaf rust	Medium
	Armyworm	High		
	Aphids	High		
	Birds	High		
Nkuninkana	Stem borer	Medium	Head smut	High
	Aphids	Medium		
Msambu	Armyworm	High		
	Locust	Medium		
	Stem borer	High		
Nkonkilangi	Armyworm	High	Head smut	medium
	Stem borer	High		
	Aphids	High		
Mseko	Stem borer	High		
	Birds	High		
	Fall armyworm	High		
	Aphids	Medium		
	Strigasp	High		

3.5.6. Sorghum variety trait preference

Of all respondents, 82.4% indicated that early maturity was their preferred sorghum variety trait, followed by drought tolerance (75.6%) and high yield. Other desired traits were taste, pest and disease resistance, post-harvest storage life, bird resistance and *Striga* resistance ranked last. Participants in the focused discussion groups at Sagara A, Msambu, and Nkonkilangi ranked drought tolerance first. The focus discussion groups at Laikala and Nkuninkana ranked high yield as the second most important sorghum variety selection criterion. The Sagara A and Msambu focused discussion groups rated early maturity as their

third most preferred trait, whereas Nkonkilangi, Mseko, and Laikala ranked high yield, drought tolerance, and good germination as their second most preferred traits, respectively. Nonetheless, drought tolerance, and resistance to birds, diseases, and pests ranked last for the sorghum growers at Sagara A, Laikala, Nkuninkana, Msambu, Nkonkilangi, and Mseko villages. Flavour and grain colour criteria were ranked fourth by participants from Nkuninkana village in Ikungi district and Konkilangi village in Iramba district (Table 3.7).

Table 3. 7. Sorghum variety preference criteria ranked by farmers

Criterion	Rank Sagara A	Rank Laikala	Rank Nkuninkana	Rank Msambu	Rank Nkonkilangi	Rank Mseko
Disease and pest resistance	5th	3rd	3rd	5th	§	5th
High yield	3rd	1st	1st	3rd	2nd	1st
Drought tolerance	1st	5th	2nd	1st	1st	2nd
Striga tolerance	7th	†	†	†	†	†
Grain color	†	†	†	†	4th	†
Early maturing	2nd	4th	†	2nd	†	†
Long post-harvest storage life	6th	†	†	†	†	†
Market availability	†	†	5th	4th	3rd	†
Flavor	†	†	4th	†	†	†
Tolerance to bird predation	†	†	†	7th	5th	3rd
Grain weight	4th	†	†	6th	†	†
Good germination	†	2nd	†	†	†	†

†Criteria not ranked

3.5.7. Drought coping strategies used by sorghum farmers

Early sowing was implemented by the highest number of participants (18%) in Iramba district followed by Ikungi (15.3%) and Kongwa (14.3%). Drought tolerant varieties were the second strategy to cope with drought in the particular districts (Table 3.8). Most farmers in Kongwa and Iramba districts cited early-maturing varieties as one of the strategies to overcome drought. Other strategies, such as cropping calendar which guide farmers the dates of planting were used in one district only. The other techniques used by the sorghum farmers to overcome drought are listed in Table 3.8. There were significant differences in coping

strategies to overcome drought stress which were cited by farmers from some of the districts. There were no significant differences for the common responses from participants' districts.

Table 3. 8. Coping strategies proposed by farmers to overcome drought in sorghum

Technique	Number of respondents (%) per district			Chi-square
	Kongwa	Iramba	Ikungi	
Early planting	14.3	18	15.3	0.895
Drought tolerant varieties	8.9	11.5	11.9	0.860
Practice contour farming	12.5	3.3	0.0	0.007**
Fertilisers application	7.1	0.0	0.0	0.012*
Timely weeding	1.8	1.6	0.0	0.599
Deep cultivation	7.1	0.0	0.0	0.012*
Tied ridging	12.5	9.8	5.1	0.373
Early maturing varieties	16.1	14.8	6.8	0.259
Irrigation	1.8	0.0	3.4	0.357
Cropping calendar	0.0	18	0.0	0.000***
Intercropping with legumes	0.0	3.3	1.7	0.392
Staggered planting	1.8	0.0	0.0	0.340

3.5.8. Factors influencing sorghum productivity

Factors enhancing sorghum productivity included low production cost, cultivation experience, market demand, Macia, Pato, sorghum as a food source, and varieties adapted to the specified locations. All of these positively influenced sorghum production (Table 3.9).

Gender (male), age, Tegemeo and area cultivated negatively influenced sorghum productivity in the study areas however, the level of influence differed among factors.

Table 3.9. Regression analysis of the factors influencing the productivity of the improved sorghum varieties

Model	Unstandardized Coefficients		Standardized Coefficients	*t*	*P*-values
	B	SE	*β*		
(Constant)	-404.4	307.9		-1.313	0.237
Gender (1 = male; 0 = otherwise)	-269.2	51.2	-0.46	-5.254	0.002**
Age	-20.7	2.5	-0.96	-8.212	0.000***
Number of men	143.7	15.7	1.05	9.160	0.000***
Number of acres	41.34	9.1	0.58	4.571	0.004**
Acres for sorghum production	-137.7	15.9	-1.37	-8.663	0.000***
Sorghum as a food source	178.4	72.3	0.27	2.469	0.049*
Market demand dummy (1 = high; 0 = low)	244.0	58.4	0.42	4.181	0.006**
Low production cost (1 = yes; 0 = no)	1,710.6	169.8	1.28	10.072	0.000***
Length of time growing sorghum (y)	366.0	75.6	0.55	4.838	0.003**
Sorghum varieties grown (1 = improved; 0 = local)	166.7	40.2	0.49	4.144	0.006***
Tegemeo (1 = grown; 0 = not grown)	-636.3	59.4	-1.07	-10.710	0.000***
Macia (1 = grown; 0 = not grown)	195.1	63.5	0.31	3.074	0.022*
Pato (1 = grown; 0 = not grown)	643.3	85.0	0.89	7.567	0.000***

a. Dependent variable: yield of improved varieties per acre

$R^2 = 0.977$ Adjusted $R^2 = 0.929$ $F = 20.042$ $P = 0.001$

*, **, *** Significant at $P < 0.05$, $P < 0.01$, and $P < 0.001$, respectively.

3.5.9. Factors influencing adoption of improved sorghum varieties

Binary logistic regression results on the factors influencing the adoption of improved sorghum varieties revealed that, some factors negatively influenced adoption whereas others positively influenced the adoption. Those promoting the adoption of improved sorghum

varieties included early maturity with the probability Exp (9.171) of the adoption indicate that [Odds/(1+Odds)] early maturity will result into significant crop adopted by 90.2% above if it was not early maturity, while the value of the Odds for area (EXP (B)) = 1.058 indicates that unit increase in area under sorghum would significantly increase the adoption rate by 51.4% than otherwise (Table 3.10). Similarly, market accessibility Odds value EXP (2.851) indicates that access to the sorghum market would significantly increase adoption rate by 74% than otherwise ($P > 0.05$). The factors negatively influencing the adoption of improved sorghum varieties included lack of drought tolerant variety whose EXP (0.169) indicate that adoption rate would significantly decrease by 15%, and cultivation experience (EXP (B) = 0.131 would significantly decrease adoption rate by 16%. This implies that with much experience in production of the same crop the farmer become reluctant of adopting new technologies believing that his own experience matters more than new ones. The values of cox and Snell R^2 and Nagelke R^2 indicated that about 20.3-32.8% of the variations in the rate of adoption of improved sorghum varieties among the farmers were contributed by the variables specified in the logistic regression model, implying that the factors are very important to consider.

Table 3. 10. Binary logistic regression of the factors influencing the adoption of improved

Variables	B	SE	Wald	df	P -Value	Exp(B)
Constant	-1.47	0.193	57.651	1	0.000***	0.231
ED (1 = primary; 0 = otherwise)	1.61	0.749	4.613	1	0.032*	5
Age dummy (1 = > 35 y; 0 = otherwise)	1.1	0.534	4.252	1	0.039*	3.006
Decreasing rainfall trend (1 = yes; 0 = no)	-0.99	0.506	3.83	1	0.05	0.371
Exp-D (1 = > 3 y; 0 = otherwise)	-2.03	0.732	7.703	1	0.006**	0.131
Area in acres (farm size)	0.06	0.068	0.683	1	0.409	1.058
Lack of DT varieties (1 = yes; 0 = no)	-1.78	0.672	7.015	1	0.008**	0.169
RH (1 = adopted; 0 = otherwise)	1.93	1.11	3.026	1	0.082	6.899
Early maturity	2.22	0.868	6.525	1	0.011*	9.171
Food security	-0.74	0.782	0.9	1	0.343	0.476
Market accessibility	1.05	0.484	4.688	1	0.030*	2.851
Decreasing rainfall trend	-0.4	0.554	0.521	1	0.47	0.671
Model Summary						
		Cox and	Nagelkerke			
-2 log likelihood		Snell R2	R2			
	129.862	0.2	0.328			

3.6. Discussion

The present study noted that, farmers were aware of the improved sorghum varieties though, the number of farmers who cultivated was low. Low adoption rates for the improved sorghum varieties among famers within districts indicates that, either technologies transfer were not sufficiently done or farmers perceived negatively the technology package for the same crop production. Similarly, the cost for adopting the technology could have been too high compared to the use of crop varieties that farmers used to grow on their fields such that farmers saw it as irrational to grow the new varieties. Other reasons could be due to lack of

stockists within farmers reach to ensure supply of the seeds during planting season. Sorghum varieties cultivated in Tanzania are open pollinated varieties (OPV) which have lower yield than hybrid varieties cultivated in countries producing the highest yield like Oman and USA (FAOSTAT, 2018). Strengthening agro dealers who sell improved sorghum seed, subsidizing the seed input cost by the government and extension service delivery may contribute to seed and knowledge access to farmers and subsequently increasing the levels of adoption of improved sorghum varieties among farmers across the districts.

According to the key informants, birds attack farm fields where improved sorghum varieties are grown due to their taste and other physiological characters of the sorghum grains. Aphids and mealybugs affect sorghum leaves, stems, and grains. Similar pests in addition to *Striga* weed and stem borers were reported to affect sorghum production (Muui *et al*., 2013; Suleiman and Rugumamu, 2017). The same finding was reported by Mofokeng *et al*. (2017) that bird damage and drought stress are the major sorghum production constraints which hinder production causing food insecurity to smallholder farmers. Furthermore, limited soil nutrients result into poor physiological plant growth. Dimkpa and Bindraban (2013) asserted that fertiliser application supplements soil nutrients, supports physiological plant growth, and boosts grain yield.

Thus, farmers need to supplement their soil fertility with application of manure and inorganic fertilisers for improved crop growth and production. Moreover, inconsistent rainfall and dry spells during the growing season hamper fertiliser application in the semi-arid areas. Based on these results, it is recommended that farmers work closely with Tanzania Meteorological Authority (TMA) and extension officers to access weather condition forecasts which are recommended for fertilisers application. Farmers' lack of knowledge to understand life cycles of pests and diseases contributed to failure to control them timely. This suggests for use of a participatory approach system and demonstration plots in order to disseminate

technologies for diagnosing of pests and diseases' symptoms in sorghum before it is too late for controlling them. Moreover, this calls upon the strengthening collaboration between agricultural researchers, extension officers and farmers in order to address pests and diseases affecting sorghum production.

The findings from these results suggest that sorghum breeders should develop new improved sorghum varieties which can tolerate post-flowering drought which is most common within the semi-arid areas which have been reported to experience decrease of rainfall due to climate change (Nhamo *et al*., 2019). On the other hand, agricultural officers, researchers and extension officers should consider dissemination of techniques for diagnosis of pests and disease symptoms in sorghum production areas. This will help farmers to be aware of the control measures against the pests and diseases of sorghum to reduce economic yield losses. Breeding for drought-tolerant sorghum varieties can reduce drought stress especially during the post-flowering period when plants need sufficient water for grain filling. This study suggests that farmers to grow improved and local sorghum varieties with traits of drought tolerance and promising yield as an alternative coping strategy to drought to ensure food availability.

Farmers practiced early sowing before rainfall as the strategy to cope with drought however this approach may not be effective as it affects the germination and emergence of seed because, more than 70% of the seeds are affected by pests while some decay, similar report was contributed by Basavaraj *et al*. (2015) who used conjoint analysis to rank five sorghum production traits and two market traits in drought prone areas under limited early rainfall before the seed germinate and emerge. Delayed planting is another strategy especially in the regions with high dry spell period, this helps fast recovery of plants from drought effect. Application of rainwater harvesting technologies like tied-ridging and contour farming has been reported to sustain moisture for plant use after ceasation of rainfall (Ouedraogo *et al*.,

2017). Application of organic fertilizers allows decomposition of nutrients while keeping soil moist for a long period than in organic fertilizers (Gaufichon *et al*., 2010). Mixed cropping assure getting yield even if some crops in combination fail others will succeed for instance farmers mix sorghum with legumes such as groundnuts, cow pea, bambaranut which contribute to reduce drought risk.

Technique of planting in holes conserves water and nutrients to support plants (Gaufichon *et al*., 2010).The finding reported by Mrema *et al*. (2017) shows that, *Striga* weed resistance was ranked the third preference by farmers who participated in the interview. This is contrary to findings in this study which ranked *Striga* weed resistance the last of choice suggesting that the variation of weed availability differ from one location to another nevertheless, further study is needed to explore scientific information.

This study noted that, some traits such as post-harvest storage, shelling and taste were ranked the last preferences notwithstanding, these traits are important for maintaining post-harvest grain quality and enhancing adoption rate of sorghum by-products. Therefore, traits of interest are key factors for enhancing adoption of improved sorghum varieties to increase sorghum grain production. Traits of interest by farmers should be considered before developing new improved sorghum varieties which are demand driven; similar suggestion was reported by Ajambo *et al*. (2017). The findings of this study, both men and women participated equally in sorghum farming, contrary to Ogeto *et al*. (2013) who reported a stronger influence of women than men. Men and women in the study areas believed that working as teams in the families have impact on the success of farming activities. However, some participants reported that, men take over when it comes to selling of agricultural produce. Climatic conditions have influence on the adoption of improved varieties from one location to another. For instance, the current study found Tegemeo, an improved sorghum

variety, was negatively influencing productivity in central zone, different from the lake zone where the adoption rate was high (Mafuru *et al*., 2007).

Even though, there were number of factors positively influencing sorghum productivity, market demand was a key factor for many farmers. Apart from the constraints raised by respondents, the findings of this study noted that, farmers are willing to produce high yielding sorghum varieties when there is assurance of market demand. For instance, some brewing industries buy white sorghum grains for beer processing nonetheless; they are not consistent for the reason of lacking legal contract between producers and brewing companies. For assured crop market and quality products there should be contractual arrangements between farmers and buyers which are legally binding.

3.7 Conclusion and recommendations

The main constraints on sorghum production in central Tanzania were poor soil fertility, drought, pests, and diseases. Stem borers, aphids and *Quelea* birds were the most destructive sorghum pests in the region. The traits preferred by farmers in the studied areas included high yield, early maturity, drought tolerance, and pest and disease resistance. Coping strategies used for addressing drought stress in sorghum comprised; early planting, use of drought tolerant varieties and use of cropping calendar. Therefore, plant breeders must factor in drought tolerance, pest and diseases resistance and other related traits when they develop improved sorghum varieties to stimulate farmers' adoption rate. To further address these constraints, collaboration among plant breeders, pathologists, entomologists, socioeconomists, soil scientists, extension officers, local government authorities, and the ministry of agriculture must be enhanced. Coping strategies should be well demonstrated to farmers to increase awareness of addressing drought effects in sorghum production. Low adoption of improved sorghum varieties is contributed by market problem. Plant breeders

should consider pest and disease resistance, market access, long-term post-harvest storage and bird damage tolerance when they develop new sorghum varieties. There is need to strengthen sorghum value chain to diversify end products and market options among stakeholders as these will motivate farmers to expand farms for sorghum production. Furthermore, the finding suggests uses of better ways of transfer of improved technologies to the target communities for increased adoption of improved sorghum varieties to maximize the productivity per hectare.

CHAPTER FOUR

4.0. INTROGRESSION OF QUANTITATIVE TRAIT LOCI FOR DROUGHT TOLERANCE INTO FARMERS' PREFERRED SORGHUM VARIETIES

4.1. Introduction

Post flowering drought that occurs during dry spells is among the major challenges of sorghum production in Tanzania. Dry spells affect physiological plant growth and sorghum grain yield (Barron, 2004). In sorghum, both pre and post flowering drought stress affects the crop, though post flowering drought stress is the major concern (Premachandra *et al*., 1994). Post flowering drought stress occurs when plants are at grain filling stage, which is critical for determination of yield performance (Kamal *et al*., 2018). Drought affects water and nutrients uptake to plant parts by causing leaf rolling, leaf senescence and reduced vegetative plant growth (Kamal *et al*., 2018). It affects plant stands, grain size, grain quality and quantity (Borrel *et al*., 2014). Farmers can escape long dry spells by planting the crop later than normal where the post-flowering period does not fall within the dry spell period (Slegers, 2008). Drought stress in sorghum has been addressed using both conventional and modern breeding tools such as genome-wide association, marker-assisted selection, marker assisted backcrossing (MABC), and bioinformatics (Serraj *et al*., 2003).

The common traits indicative of drought tolerance include plant height, leaf rolling, leaf senescence, chlorophyll content, days to 50% flowering, days to 50% maturity and root biomass (Velazco *et al*., 2019). Besides, panicle length, panicle weight, and number of seeds per panicle, number of tillers, grain size, dry matter and inflorescence exsertion can also be indicative traits for drought tolerance (Sory, 2015). Nevertheless, drought stress is a highly complex trait, where environmental and crop management greatly contribute to the variability

found (Weeden *et al.*, 1993; Boyles *et al.*, 2019). Therefore, the identification of plants that associate correctly with the trait is quite challenging. It has been indicated that, retaining greenness in leaves during limited water condition accelerates carbon fixation for the production of starch (Van Oosterom *et al.*, 1996). The greenness of leaves allows photosynthesis process to produce food for the plant and minimizes premature death of flowers which ultimately maintains yield under drought stress conditions (Rosenow and Clark 1981).

The exploitation of drought tolerance traits in sorghum has been achieved by mapping STG QTLs which are associated with the expression of phenotypic traits such as delay of leaf senescence and maintaining of green leaves at maturity in the field. The STG in sorghum is a recessive trait character that expresses retention of greenness of the leaf area at maturity (Borrell *et al.*, 2014). The efficiency of retention of green leaf area depends on the genetic makeup of sorghum genotypes under limited water condition (Borrell *et al.*, 2014). It is difficult to select plants by phenotyping only because variations may not be due to genetic differences but rather it may be due to environmental effects that may mislead screening (Ribaut and Betran, 1999). To screen for drought tolerance, researchers have developed molecular markers that are tightly linked to STG QTLs associated with drought tolerance. Studies have been conducted to map STG QTLs in sorghum genotypes SC56, B35, KS19 and E36-1 using SSR and SNPs markers (Xu *et al.*, 2000).

The parental lines B35 and E36-1 contain QTL which contribute to expression of the STG phenotypes in sorghum (Kabede *et al.*, 2001). The B35 parental line is characterized by high chlorophyll content, STG and stable root system traits. These agro-morphological characteristic traits enable plants to grow and survive under limited moisture conditions and can be used as phenotypic markers to aid the introduction of drought tolerance into sorghum genotypes in the development of superior varieties for cultivation under arid drought stress

environments. Within the several traits, STG is the major trait that enhances photosynthesis in plants for growth development and yield. Sorghum genotypes with STG have high efficiency of utilizing nitrogen nutrients from the soil which lower leaf senescence under limited moisture conditions (Ngugi *et al*., 2013).

Molecular markers that are used for QTLs mapping are suggested to be of high polymorphism. AFLP, RAPD and RFLP markers have been applied in breeding for drought tolerance, however; AFLP and RAPD has been poor mainly because they fail to differentiate between homo and heterozygous alleles. RFLP requires high molecular weight DNA and also because of its tedious experimental assays, has not been amenable to automation, these days it is not used. SSR markers are cost-effective, simple and can distinguish heterozygous versus homozygous but not as high throughput as SNP markers (Gimode *et al*., 2016; Mammadov *et al*., 2012). KASP markers are useful for screening genotypes that contain traits of interest for a short time compared to field phenotyping. In sorghum, the use of KASP markers can shorten breeding cycles of releasing the new inbred lines by half of the cycles used for the development of inbred lines by conventional breeding method (Semagn *et al*., 2013). KASP markers identify heterozygous alleles that are important for the improvement of traits through introgression to the deficient varieties by backcrossing (Thomson, 2014). KASP markers are simple and amenable for use however it is medium throughput (Meade *et al*., 2019). For efficiency, marker-assisted backcrossing for STG in sorghum, KASP markers are being employed. Furthermore, the SNPs markers in sorghum breeding are used to fine-map the stay-green QTLs which are closely linked to the genes that express stay-green during post-flowering drought. Similarly, SNPs markers have the ability to detect clearly and efficiently the existing polymorphism among genotypes, including in sorghum. High effective marker-assisted selection shortens the genetic distance of the flanking DNA markers of particular QTLs of target (Hospital, 2001). First, mapping starts with many markers followed by

successive reduction based on the number of backcrosses used. Screening targets recurrent parent alleles with markers linked to QTLs of a target in the genome which depends on the study objective (Morris *et al*., 2003). It is advised that mapping of STG QTLs should be done at BC_2F_1 or BC_3F_1 onwards as at this stage the introgression of targeted traits is attained. One of the approaches that currently is being used to tackle drought in sorghum is the introgression of QTLs from the donor parents to the recurrent parents until the genome or main features of the recurrent parent is recovered (Meru, 2010). Notwithstanding, this approach needs several backcrosses and high polymorphic markers for successive recovery of recurrent parent traits.

Therefore, the objective of this study was to:

i. introgress STG QTLs from donor parents B35 and S35 to the recurrent parents NACO Mtama 1, Seguifa, Tegemeo, Macia, Pato, Wagita, Wahi, Hakika and Kenya by using MABC; and

ii. identify STG QTLs transferred to the recurrent parents using SNPs markers

4.2. Materials and methods

4.2.1. Location of the study

The study was conducted at TARI- Makutupora Centre located 23 km North of Dodoma city in Dodoma region (Longitude: 35º, 46.093'E and Latitude: 05º, 58.669'S) (Altitude: 1070 m). The annual rainfall at TARI- Makutupora ranges from 300-500 mm with poor distribution and over 500 mm in few years, and the temperature varies from 15-35.1°C (Tanzania Meteorological Agency, 2014). The area is classified as semi-arid which is characterized by a mono-modal rainfall pattern. Rainfalls occur between December and April with dry spell in February.

4.2.2. Markers used for phenotyping and genotyping of BC_2F_1 population

Morphological and molecular markers that track the presence of drought tolerance and related traits of agronomic importance were used in this study. The utilization of these markers made the selection of genotypes of interest effective.

The morphological markers used in this study were leaf rolling, leaf senescence, plant height, days to 50% flowering, days to 50% plant maturity, total number of green leaves at plant maturity, grain colour and grain size, grain weight, root biomass, stem biomass and chlorophyll content. These traits are easy for visual analysis during field evaluation. The molecular markers were used to trace favourable alleles for the STG QTLs as described by Burow *et al*. (2019) and introgressed into preferred sorghum varieties. A total of 30 SNP Kompetitive allele specific PCR (KASP) markers were used to screen materials at population stage BC_2F_1 at the first genotyping (da Silva *et al*., 2020). Seven out of these markers were for STG 1, 2 and 3; the remaining 23 SNP markers were for traits contributing to STG in plants such as heat shock domain, programmed cell death triggering, aspartic proteases and chloroplast precursor, these traits are expressed in sorghum plants during drought. Others were used for the recurrent specific traits (Table 4.1.).

A total of 10 SNP (KASP) markers were used to genotype the BC_2F_3 population in the second genotyping. Three out of these markers were for STG 1, one marker for STG 2 and three SNP markers were for STG 3; the remaining 3 SNP markers were for traits contributing to STG in plants such as heat shock domain, programmed cell death triggering, aspartic proteases and chloroplast precursor. One marker was used for the recurrent specific traits (Table 4.2.).

Table 4. 1. List of SNPs markers associated with STG QTLs for drought tolerance in sorghum for first genotyping

S.n	Physical_Mbp	SNP Ids	alleles	Donor allele	Alternate allele	possible traits
1	S2_56112177	snpSB0035	C/T	C	T	grain types
2	S2_56918230	snpSB0037	C/T	C	T	
3	S2_56918233	snpSB0038	A/T	A	T	
4	S2_57495274	snpSB0039	A/G	A	G	PCD triggering
5	S2_59000770	snpSB0040	C/T	C	T	
6	S2_59047449	snpSB0041	C/G	C	G	
7	S2_59255001	snpSB0042	T/C	T	C	
8	S2_59281892	snpSB0043	T/C	T	C	
9	S2_59472716	snpSB0044	A/T	A	T	
10	S2_59821923	snpSB0049	G/A	G	A	N2 mobilization/ SAG/SGR1
11	S2_60059010	snpSB0053	A/G	A	G	
12	S2_60098184	snpSB0054	G/A	G	A	
13	S2_61811307	snpSB0072	G/A	G	A	APETALA2 and EREBPs
14	S2_62145285	snpSB0075	A/G	A	G	Recurrent parent specific
15	S2_62155735	snpSB0076	T/A	T	A	Heat shock domain
16	S2_62155778	snpSB0077	T/C	T	C	
17	S2_62378269	snpSB0080	C/T	C	T	Recurrent parent specific
18	S2_63375987	snpSB0083	T/C	T	C	Chloroplast precursor/ubiquitionus
19	S2_63690795	snpSB0087	A/G	A	G	
20	S2_65453155	snpSB0089	C/A	C	A	SGR2
21	S2_67306935	snpSB0091	A/C	A	C	panicle compactness
22	S2_67357372	snpSB0092	C/G	G	C	Salt responsive

Comments

Favourable allele

Most important for STG traits

23	S2_67423749	snpSB0093	A/T	A	T	
24	S2_67664290	snpSB0094	T/C	T	C	
25	S2_67710384	snpSB0095	A/G	A	G	
26	S2_69739036	snpSB0098	C/G	G	C	Aspartic proteases through SA
27	S2_69859850	snpSB0099	C/T	C	T	
28	S2_70523721	snpSB0101	C/G	C	G	SGR3
29	S2_71360153	snpSB0102	A/G	A	G	
30	S2_71419274	snpSB0103	C/G	G	C	

Table 4. 2. List of SNPs markers used for the second genotyping of BC_2F_3 population

S.n	Physical_Mbp	SNP Ids	alleles	Donor allele	Alternate allele	possible traits
1	S2_59255001	snpSB0042	T/C	T	C	PCD triggering
2	S2_59821923	snpSB0049	G/A	G	A	
3	S2_60059010	snpSB0053	A/G	A	G	N2 mobilization/ SAG/SGR1
4	S2_60098184	snpSB0054	G/A	G	A	
5	S2_61811307	snpSB0072	G/A	G	A	APETALA2 and EREBPs
6	S2_65453155	snpSB0089	C/A	C	A	SGR2
7	S2_69739036	snpSB0098	C/G	G	C	Aspartic proteases through SA
8	S2_70523721	snpSB0101	C/G	C	G	
9	S2_71360153	snpSB0102	A/G	A	G	SGR3
10	S2_71419274	snpSB0103	C/G	G	C	

Comments

Favourable allele

Most important for STG traits

4.3. Introgression of STG QTLs from donor parents to the recurrent parents and genotyping

Marker-assisted backcrossing (MABC) method was used to introgress STG QTLs alleles from the donor parents B35 and S35 into the farmers' preferred sorghum varieties (NACO-Mtama 1, Macia, Seguifa, Hakika, Wahi and Pato) in Tanzania. The donor parent B35 is drought tolerant and contains STG 1, STG 2, STG 3 and STG 4 QTLs. Parental line S35 is an introgression line with B35 as STG QTL donor (STG A). These donor parents were introgressed into recurrent parents Pato, Macia, Wahi, Hakika, Seguifa and NACO Mtama1 to develop generations of F_1, BC_1F_1 BC_2F_1, BC_2F_2 and BC_2F_3 populations for strengthening the stability of drought tolerance in sorghum. The recurrent parent Pato was originated in Ethiopia but was released in Tanzania after multi-location screening. This is a white grain variety that is used for food, and it is characterized by fast recovery from drought, having a maturity period between 95-100 days, highly palatable with hard grain endosperm and no testa. Macia and Seguifa are also white grained and used for food while NACO Mtama1 is

white grain used for food and brewing (Simtowe and Mausch, 2018). The introgressions of STG QTLs in drought susceptible sorghum varieties were achieved using marker-assisted backcrossing (MABC) method used by Reddy *et al*. (2014).

4.3.1. The development of F_1, BC_1F_1 and BC_2F_1 populations

A total of 12 crossing plots with 5 rows each were planted on two crossing blocks. Inter-row and intra-row spacing was 0.75 m and 0.30 m respectively. The distance between crossing plots was 1.5 m whereas the distance between crossing blocks was 2 m. The plastic bag method was used to emasculate anthers in the florets by clipping the top and bottom parts of panicle at initial flowering. The remaining portion of the panicle was covered by using a white plastic bag to allow high temperature and humidity to accelerate anthers formation and death to avoid selfing during crossing. Scissor, cotton wool, rubber band, tweezers, dissecting needles and sharp pencil points were used for emasculation. Pollen was collected in paper bags from the donor parent plants to the preferred recurrent parent plants during morning time from 7:00 to 11:00 am, precisely when flowers are blooming and shedding pollen. This was followed by dusting the pollen on the stigma of emasculated panicles of the recurrent parents to obtain the F_1 seeds. Each head of the selected recurrent parent was cross pollinated by a single donor parent and both were bagged soon after pollination. Pollination bags were removed at the soft dough stage 10- 15 days from pollination and the seed set on bagged heads was assessed visually using a scale of 0 to 100% where 0% represented a completely sterile head without seed set, and 100% represented a completely fertile head with complete seed set. Birds were controlled from eating seed set after removing the bags by using nets which were covered for each panicle.

To generate BC_1F_1 populations, the F_1 seeds from each of the F_1 generation were planted and backcrossed to their respective recurrent parents. Seeds from the F_1 and recurrent parents

were planted in different crossing blocks on 17th and 22nd August, 2018 at TARI-Makutupora Centre. Each of the F_1 generation corresponding to each of the recurrent parent was planted side by side with spacing a of 0.75 m between rows and 0.30 m within rows where each row was 5.4 m long. The emasculation of anthers was conducted on the recurrent parents to leave female parts of plant which were pollinated with pollen from the F_1 plants to produce BC_1F_1 populations.

BC_1F_1 seeds were used to generate BC_2F_1 populations; seeds from each of the BC_1F_1 population were planted and backcrossed to their recurrent parents. Seeds from the BC_1F_1 and recurrent parents were planted in different crossing blocks on 24th and 30th December 2018 at TARI-Makutupora Centre, Tanzania. Each of BC_1F_1 corresponding to each recurrent parent was planted in a length of 5.4 m side by side using the same spacing as ealier indicated. The plastic bag method was used for anthers emasculation on the recurrent parents for each BC_1F_1 populations as previously explained. The emasculated recurrent parents' flowers in both trials were pollinated with BC_1F_1 pollen to produce BC_2F_1 populations. The number of seeds was recorded.

4.3.2. Genotyping of BC_2F_1 populations

Genotyping of BC_2F_1 populations for STG QTLs was performed with a KASP marker (LGC, Middlesex, UK) in an agreement with Intertek-AgriTech

(https://www.intertek.com/agriculture/agritech/: accessed on 20 July 2021) in Sweden. A total of 200 seeds per BC_2F_1 population and 17 seeds per parent were sown on 14th June 2019 at TARI- Makutupora Centre in Dodoma region, Tanzania using the spacing of 0.75 m between rows and 0.3 m between intra-row. A total of 150 leaf samples per BC_2F_1 populations and 3 samples per donor and recurrent parent population making a total of 752

samples were collected from the breeding nursery 46 days from planting. Prior to leaf sample collection, the 96 well plates were labelled for sample tracking. Three leaf discs of approximately 4-5 cm were collected per plant sample using single-hole punching methodology and placed into the 96 well plates. The plates were kept in ice-box containing ice packs to avoid deterioration of the genomic (gDNA) concentration while collecting samples in the field. Further, samples were oven dried at a temperature of 40^{O}C for 48 hours followed by packing and shipping to Intertek laboratory in Sweden for genotyping.

DNA extraction at vendor was conducted as follows; plant tissue samples were homogenised before commencing the extraction. The extraction was done using the sbeadex™ magnetic microparticles Kit from LGC (UK). Briefly, a concentration of 250 µL lysis buffer PN was added to each homogenised sample, and then samples were incubated at 65°C for 10 minutes followed by centrifugation at 2500 x g for 10 min to pellet the debris. The total of 520 µL binding buffer PN and 60 µL sbeadex particle suspension was added to a fresh sample tube after mixing well with the sbeadex particle suspension. About 200 µL lysate was transferred into the prepared tube containing binding buffer PN and sbeadex particle suspension. It was followed by thoroughly mixing and setting the pipette volume to 700 µL and pipetting up and down 5 times. The sample solution was incubated at room temperature for 4 min to allow binding to occur. Shake tube(s) by magnetic for 1 min at room temperature which enabled sbeadex particles to form a pellet. The supernatant was removed and discarded. The magnetics were separated from the sample tubes, then 400 µL wash buffer PN1 was added followed by mixing thoroughly by pipetting 350 µL to fully re-suspend the pellet. The sample tubes were incubated at room temperature for 10 min. The samples were agitated periodically by using a shaker or vortex. The magnet was brought into contact with the tubes for 1 min at room temperature where, sbeadex particles formed a pellet. The supernatant was removed

and discarded while taking care not to dislodge the pellet. The magnet was separated from the sample tubes. The addition of 400 µL wash buffer PN2 and 400 µL sterile ultrapure water were repeated to separate supernatants and DNA pellets. One hundred micro litre elution buffer PN was added to the pellet. Elution buffer and pellets were mixed thoroughly by pipetting 75 µL to fully re-suspend the pellet. Sample tubes were incubated at 55 °C for 10 min, and agitated periodically using a heated shaker or vortex. An elution was performed at a temperature of 55 °C which is recommended for high DNA yield. The magnet was brought into contact with the tubes for 3 min at room temperature where sbeadex particles formed a pellet. An eluate of 80 µL was transferred to a new tube by pipetting and the DNA quality and concentration were checked using nanodrop spectrophotometer. Isolated DNA samples were loaded in 0.8% agarose gel followed by electrophoresis and Lambda (λ) DNA standards were compared to The DNA content. Samples with good quality and quantity of 2 to 2.5 ng per µl were diluted as working solution before amplification. The DNA samples were arrayed into a 96-well with a total of 752 samples. No template controls (NTCs) were included on each of 8 PCR plates. KASP genotyping mix (LGC, UK) and genotyping mix was prepared according to the intended number of reactions plus an additional dead volume. Dry DNA method was used for genotyping, n/a of DNA, 2.5 µL of 2x KASP Master mix, 0.07 µL KASP assay mix and 2.5 µL of water with the total reaction volume of 5 µL. All reagents were vortexed before use. The required amount of genotyping mix to each DNA sample in the reaction plate using a pipette or dispensing robot was added. The plate with an optically clear seal was sealed. The plate was centrifuged at 550 *x* g. The thermal cycle for genotyping was activated at 94^{O}C for 15 minutes, denatured at 94^{O}C for 20 minutes, Annealing/elongation temperature 61-55^{O}C for 60 seconds (drop 0.6°C per cycle). Denaturation at step three was set at 94^{O}C and annealing temperature of 55^{O}C. The reaction

plate in a FRET-capable plate reader was read after completion of the thermal cycle where all plates were read below 40^{O}C (Gimode *et al.*, 2016).

4.3.3. Development of B_2F_2 and BC_2F_3

The genotypes W82, NA241 and NA307 with heterozygous alleles and two genotypes NA316 and SE438 with homozygous alleles were selected from BC_2F_1 population as the best performing after field and genotyping analysis. These genotypes were planted in the field for evaluation of their performance on grain yield, days to 50% flowering, days to 50% maturity, plant vigour, panicle weight and appealing of grain seeds to generate BC_2F_2 population. The genotypes NA241A, NA241B, NA316A, NA316B, NA316C, NA307, SE408 and SE438 were screened from the population BC_2F_2 as the best performing under field condition. Each genotype of BC_2F_2 populations was planted to generate genotypes of BC_2F_3 populations.

4.3.4. Genotyping of BC_2F_3 population

The genotyping of BC_2F_3 population was conducted to evaluate availability of favourable alleles for STG trait in BC_2F_3 populations. Seven genotypes (NA241A, NA241B, NA316A, NA316B, NA316C, SE408 and SE438) were chosen and planted for the second genotyping and field evaluation. The genotype NA307 was evaluated under field conditions only following the limitation of samples needed for genotyping. At least a total of 100 leaf samples from at least 100 plants were sampled per genotypes and 3 samples per control genotype and per parent making a total of 752 samples. The young leaf samples with quality DNA were collected 35 days from planting. Sampling was done on the fresh dry leaf to avoid molds when shipping samples to molecular genotyping facility. Samples were kept in the icebox with ice parks inside to avoid DNA denaturation. The samples were oven-dried for

two days at 40^{O}C. Dried samples were packed and labeled well for shipping to Intertek laboratory in Sweden for molecular data analysis.

The procedures for DNA extraction, genotyping and molecular data analysis of BC_2F_3 population were conducted in the same way as it was done in the genotyping of BC_2F_1 population above. The genotyping of BC_2F_3 population used 10 SNP markers which indicated high number of favourable alleles for STG expression with corresponding high STG under field conditions in BC_2F_1 population (Table 4.2).

4.4. Molecular data analysis

Marker-assisted selection data analysis was conducted using flapjack (https://excellenceinbreeding.org/toolbox/tools/flapjack) tool and the genotypic data input files for flapjack were prepared using a data file conversion tool in galaxy online tool accessed through the EiB galaxy portal (http://galaxy-demo.excellenceinbreeding.org/). The galaxy input file containing genotype file, sample file, SNP summary, original genotype file, original sample file, original SNP information, sample list in order, matched id results and header were generated and imported into the EiB galaxy online software (https://excellenceinbreeding.org/toolbox/tools/eib-galaxy-instance).The data inputs generated in the galaxy tool were converted into the https://ics.hutton.ac.uk/flapjack/ file which was used for further analysis.

4.5. Results

4.5.1. BC_2F_1 genotyping results

Seventy one (10%) out of 728 BC_2F_1 samples collected for genotyping was heterozygous (Table 4.3). Of these, SNP markers snpSB00075, snpSB00102 and snpSB00103 were scored

as heterozygous alleles in a total of 7 samples with BC_2F_1 of B35*Wahi background. The markers snpSB00102 and snpSB00103 were expressed as heterozygous alleles in the total of 8 samples of BC_2F_1 with BS35*Hakika background. The markers snpSB00049, snpSB00077, snpSB00102 and snpSB00103 indicated heterozygous allele in 37 samples of S35*Pato background. The markers snpSB00072, snpSB00077 and snpSB00103 expressed heterozygous alleles in 10 samples of BC_2F_1 with B35*Seguifa background. The markers snpSB00037, snpSB00041, snpSB00098, snpSB00102 and snpSB00103 showed heterozygous allele in 10 samples with B35*Macia background and the markers snpSB00042, snpSB00098, snpSB00102 and snpSB00103 expressed heterozygous allele in the total of 18 samples of B35*NACO Mtama 1 background (Table 4.3). The rest (19) SNP markers showed homozygous allele of the BC_2F_1 samples used for genotyping.

Table 4. 3. BC_2F_1 sorghum populations scored with heterozygous alleles using SNP markers

Population	Sample number	SNP markers	H.Allele	DP alleles	Rp alleles
BC_2F_1 (B35*Wahi)	22	snpSB00102	G/A	AA	GA
	44	snpSB00103	G/C	CC	CC
	59	snpSB00103	G/C	CC	CC
	70	snpSB00103	G/C	CC	CC
	82	snpSB00102	G/A	AA	GA
	90	snpSB00102	G/A	AA	GA
	94	snpSB00102, snpSB00103	G/A, G/C	AA, CC	GA, CC
BC_2F_1 (S35*Hakika)	111	snpSB00103	G/C	CC	CC
	117	snpSB00103	G/C	CC	CC
	125	snpSB00103	G/C	CC	CC
	127	snpSB00103	G/C	CC	CC
	128	snpSB00103	G/C	CC	CC
	131	snpSB00102, snpSB00103	G/A, G/C	AA, CC	GA, CC
	138	snpSB00103	G/C	CC	CC
	157	snpSB00103	G/C	CC	CC
BC_2F_1 (NACO Mtama 1)	175	snpSB00102	G/A	AA	AA
	190	snpSB00102	G/A	AA	AA
	207	snpSB00102	G/A	AA	AA
	210	snpSB00103	G/C	CC	CC
	213	snpSB00103	G/C	CC	CC
	241	snpSB00102,snpSB00103	G/A, G/C	AA, CC	AA
	255	snpSB00102	G/A	AA	AA
	266	snpSB00103	G/C	CC	CC
	272	snpSB00103	G/C	CC	CC
	276	snpSB00103	G/C	CC	CC
	284	snpSB00103	G/C	CC	CC
	304	snpSB00102	G/A	AA	AA
	307	snpSB00102,snpSB00103	G/A, G/C	AA, CC	AA, CC
	319	snpSB00103	G/C	CC	CC
	320	snpSB00103	G/C	CC	CC
BC_2F_1 (B35* Macia)	347	snpSB00103	G/C	CC	CC
	348	snpSB00102, snpSB00103	G/A, G/C	AA, CC	AA, CC
	352	snpSB00103	G/C	CC	CC

Table 4.3: Continue

	391	snpSB00103	G/C	CC	CC
	395	snpSB00103	G/C	CC	CC
BC_2F_1 (B35*Seguifa)	418	snpSB00072	A/G	GG	AA
	441	snpSB00072	A/G	GG	AA
	463	snpSB00072	A/G	GG	AA
	566	snpSB00072	A/G	GG	AA
	567	snpSB00103	G/C	CC	CC
	569	snpSB00103	G/C	CC	CC
	581	snpSB00103	G/C	CC	CC
	582	snpSB00103	G/C	CC	CC
BC_2F_1 (S35*Pato)	586	snpSB00103	G/C	CC	CC
	589	snpSB00102, snpSB00103	G/A, G/C	A,CC	
	595	snpSB00049, snpSB00102	G/A, C/T	GG,AA	AA,AA
	601	snpSB00103	G/C	CC	CC
	606	snpSB00049	G/A	GG	AA,CC
	610	snpSB00049, snpSB00103	G/A, G/C	GG,CC	
	624	snpSB00102	G/A	AA	AA
	625	snSB00103	G/C	CC	CC
	631	snpSB00102	G/A	AA	AA
	634	snpSB00102	G/A	AA	AA
	637	snSB00102, snpSB00103	G/A, G/C	AA,GG	AA,CC
	638	snpSB00102	G/A	AA	AA
	639	snpSB00102, snpSB00103	G/A, G/C	AA,GG	AA,CC
	640	snpSB00103	G/C	CC	CC
	644	snpSB00103	G/C	CC	CC
	645	snpSB00102	G/A	AA	AA
	647	snpSB00103	G/C	CC	CC
	656	snpSB00103	G/C	CC	CC
	659	snpSB00102	G/A	AA	AA
	661	snpSB00102, snpSB00103	G/A, G/C	AA,CC	A,CC
	663	snpSB00103	G/C	CC	CC
	68	snpSB00103	C/T, G/C	CC	CC
	671	snpSB00103	G/C	CC	CC
	679	snpSB00103	G/C	CC	CC
	681	snpSB00103	G/C	CC	CC
	685	snpSB00102, snpSB00103	G/A, G/C	AA,CC	AA,CC
	703	snpSB00102	G/A	AA	AA
	725	snpSB00102	G/A	AA	AA

Eighteen SNP markers showed favourable alleles among 728 of BC_2F_1 samples genotyped (Table 4.4). The list of markers that indicated favourable alleles that were most important for STG included: snpSB00049 and snpSB00054 for STG 1, and snpSB00102 and snpSB00103 for STG 3. The rest of alleles were favourable for other roles related to STG in sorghum, for instance triggering programmed cell death, heat shock domain and salt regulation in plants during drought conditions.

Table 4. 4. Favourable alleles identified and the roles in sorghum under drought condition

SNP Markers	Favourable allele	Role
snpSB00039	A	Triggering programmed cell death
snpSB00040	C	Triggering programmed cell death
snpSB00041	C	Triggering programmed cell death
snpSB00043	T	Triggering programmed cell death
snpSB00044	A	Triggering programmed cell death
snpSB00049	G	STG 1
snpSB00053	A	STG 1
snpSB00076	T	Heat shock domain
snpSB00077	T	Heat shock domain
snpSB00083	T	Chloroplast precursors
snpSB00087	A	Chloroplast precursors
snpSB00089	C	STG 2
snpSB00093	A	Salt regulation
snpSB00094	T	Salt regulation
snpSB00095	A	Salt regulation
snpSB000101	C	STG 3
snpSB00102	A	STG 3
snpSB00103	G	STG 3

Three samples of plants (W82, NA241 and NA307) with heterozygous alleles and two samples (NA316 and SE438) with homozygous alleles were chosen as samples which contained favourable alleles for STG QTL for planting to generate BC_2F_3 populations. Plant W82 possessed snSB00102, NA241 (snSB00102 and snSB00103) and NA307 was linked

with snSB00101 and snSB00102 markers for STG 3. Moreover, plant sample NA316 recorded homozygous alleles for SNP markers SNsb00049, snSB00053 and snSB00054 for STG 1 and snSB00102 for STG 3 while plant SE438 showed homozygous alleles of SNP marker snSB00054 for STG 1, and snSB00102 for STG 3. Plants which expressed heterozygous alleles after genotyping were categorized as the first priority because of the availability of traits from both parents for successful STG introgression from either of the donor parents B35 or S35 to the recurrent parents. Prior to selection, phenotypic data were recorded for traits such as days to 50% flowering, plant height, days to 50% plant maturity, panicle weight, grain weight per plant, seed vigour, stability of plants, leaf senescence, susceptible to pest and diseases. Plants with good performance of the traits above were selected and compared with genotyping data. Only plants with favourable alleles for STG 1, STG 2 or STG 3 and good performance under field conditions were selected.

4.5.2. BC_2F_3 genotyping results

Seven out of ten SNP markers (snpSB00049, snpSB00053, snpSB00054, snpSB00089, snpSB00098, snpSB00101 and snpSB00102) used for genotyping of BC_2F_3 population identified favourable homozygous alleles for STGs in sorghum. snpSB00049, snpSB00053, snpSB00054 identified favourable homozygous alleles for STG 1, snpSB00089 for STG 2 and snpSB00101 and snpSB00102 identified STG 3 (Table 4.5). The snpSB00042, snpSB00072 and snpSB00103 SNP markers failed to identify favourable alleles.The SNP markers snpSB00054, snpSB00089 and snpSB00101 the highest number (124) of samples in almost all genotypes genotyped while snpSB00042 and snpSB00098 failed to identify favourable alleles for STG. The donor parent B35 showed favourable alleles, the recurrent parents NACO Mtama 1, Seguifa and control Wahi did not show favourable alleles.

Table 4. 5. BC_2F_3 populations scored by at least six favourable homozygous alleles using 10 SNP markers

Population	Genotype	SNP markers	FA	Number of FA
BC_2F_3 (B35*NACO)	NA241A	snSB00042	TT	0
		snSB00049	GG	115
		snSB00053	AA	122
		snSB00054	GG	124
		snSB00072	AA	6
		snSB00089	CC	124
		snSB00098	GG	0
		snSB00101	CC	124
		snSB000102	AA	123
		snSB00103	GG	0
	NA241B	snSB00042	TT	0
		snSB00049	GG	113
		snSB00053	AA	122
		snSB00054	GG	122
		snSB00072	AA	4
		snSB00089	CC	122
		snSB00098	GG	0
		snSB00101	CC	122
		snSB000102	AA	122
		snSB00103	GG	0
	NA316A	snSB00042	TT	0
		snSB00049	GG	116
		snSB00053	AA	120
		snSB00054	GG	120
		snSB00072	AA	2
		snSB00089	CC	121
		snSB00098	GG	0
		snSB00101	CC	121
		snSB000102	AA	121
		snSB00103	GG	0
	NA316B	snSB00042	TT	0
		snSB00049	GG	70
		snSB00053	AA	115
		snSB00054	GG	115

Table 4.5. continue

		snSB00072	AA	40
		snSB00089	CC	115
		snSB00098	GG	0
		snSB00101	CC	115
		snSB000102	AA	115
		snSB00103	GG	0
	NA316C	snSB00042	TT	0
		snSB00049	GG	92
		snSB00053	AA	122
		snSB00054	GG	122
		snSB00072	AA	27
		snSB00089	CC	122
		snSB00098	GG	0
		snSB00101	CC	121
		snSB000102	AA	121
		snSB00103	GG	0
BC_2F_3 (B35*Seguifa)	SE408	snSB00042	TT	0
		snSB00049	GG	55
		snSB00053	AA	55
		snSB00054	GG	55
		snSB00072	AA	0
		snSB00089	CC	73
		snSB00098	GG	0
		snSB00101	CC	73
		snSB000102	AA	73
		snSB00103	GG	0
	SE438	snSB00042	TT	0
		snSB00049	GG	23
		snSB00053	AA	51
		snSB00054	GG	51
		snSB00072	AA	26
		snSB00089	CC	52
		snSB00098	GG	0
		snSB00101	CC	52
		snSB000102	AA	52

Table 4.5. continue

		snSB00103	GG	0
DP	B35	snSB00042	TT	2
		snSB00049	GG	2
		snSB00053	AA	2
		snSB00054	GG	2
		snSB00072	AA	0
		snSB00089	CC	2
		snSB00098	GG	2
		snSB00101	CC	2
		snSB000102	AA	2
		snSB00103	GG	2
Control	Wahi	snSB00042	TT	0
		snSB00049	GG	3
		snSB00053	AA	3
		snSB00054	GG	0
		snSB00072	AA	0
		snSB00089	CC	3
		snSB00098	GG	0
		snSB00101	CC	2
		snSB000102	AA	3
		snSB00103	GG	0
RP	NACO Mtama 1	snSB00042	TT	0
		snSB00049	GG	0
		snSB00053	AA	3
		snSB00054	GG	3
		snSB00072	AA	3
		snSB00089	CC	3
		snSB00098	GG	0
		snSB00101	CC	3
		snSB000102	AA	3
		snSB00103	GG	0
	Seguifa	snSB00042	TT	0
		snSB00049	GG	3
		snSB00053	AA	3
		snSB00054	GG	3

Table 4.5. continue

	snSB00072	AA	0
	snSB00089	CC	3
	snSB00098	GG	0
	snSB00101	CC	3
	snSB000102	AA	3
	snSB00103	GG	0

DP- Donor parent, FA- Favourable allele, no- Number, RP- Recurrent parent,

The SNP marker snpSB00089 indicated the highest (729) total number of samples with favourable homozygous alleles for STG followed by snpSB00101 (728) (Table 4.6). The marker snpSB00103 indicated the lowest (0)

Table 4. 6. Favourable alleles identified and the roles in sorghum under drought condition

SNP Markers	FA	Role	Number of samples with favourable alleles
snpSB00042	T	Triggering PCD through SA	0
snpSB00049	G	STG 1 and N mobilization	584
snpSB00053	A	STG 1 and N mobilization	707
snpSB00054	G	STG 1 and N mobilization	709
snpSB00072	A	APETALA2) and EREBPs	105
snpSB00089	C	STG 2	729
snpSB00098	G	Aspartic proteases through SA	0
snpSB00101	C	STG 3	728
snpSB00102	A	STG 3	727
snpSB00103	G	STG 3	0

EREBPs- ethylene-responsive element binding proteins, FA- Favourable allele, N- Nitrogen, PCD- programmed cell death, SA- salicylic acid, STG- stay-green

4.6. Discussion

Findings in this study revealed the importance of molecular markers to shorten breeding cycles in sorghum. The genotyping results indicated plants which contained favourable alleles for STG. Such information is crucial when exploiting traits from the parents to improve new lines, as STG is a complex trait associated with several genes. The introgression of STG QTLs to senescence genotypes by MABC enhances post-flowering drought tolerance in sorghum (Kamal *et al*., 2018; Kiranmayee *et al*., 2020). The transfer of STG trait from the donor parent B35 and S35 to the farmers' preferred varieties in the current study was the basis of drought tolerance improvement of new sorghum genotypes generated. The introgression of STG QTL uses SNP markers which have been screened for post-flowering drought tolerance. This study identified favourable alleles for donor and recurrent parents on STG 1, STG 2 and STG 3 in the samples of the genotypes BC_2F_1 and BC_2F_3 populations indicating successful introgression. The incorporation of STG QTLs improves plants' delay leaf senescence from post flowering to physiological plant maturity. Sukumaran *et al*. (2016) reported 8 to 24% phenotypic variations contributed by STG QTLs with the flanking SNP markers. Other studies which used SSRs markers for improvement of STG 1, STG 2, STG 3 and STG 4 in sorghum were reported successful introgression (Kimani *et al*., 2012; Ouedraogo *et al*., 2017; Reddy *et al*., 2014). However, this study was focused on SNP markers which produce clear map resolution for clear polymorphism. The SNP markers indicate bialleles arrangement in the whole of genome. The markers identified alleles with high rate of recovery in the genome which made their precision higher than other markers (Da Silva *et al*., 2020). Findings in the current study show that, some of the new genotypes generated by backcrossing method performed well in grain yield compared to control and recurrent parents. The outperformance of new genotypes over farmers' cultivated varieties shows the advantages of SNP markers in sorghum improvement. STG 1 QTL adapts post

flowering drought stress using SSR markers. STG 1 QTL increases yield grain more than other QTLs thus, it is named as the best trait using SSR marker (Kamal *et al*., 2017). The STG 3A and 3B QTLs are flanked by SNP markers from the target region of 56 Mbp to 72 Mbp for delaying leaf senescence at post-flowering drought in sorghum because of their water use efficiency. Currently, SNP are the most commonly used markers for identification of genetic polymorphism which are used in the study of genetic variation among populations (Adu *et al*., 2019; Nelimor *et al*., 2020). Da Silva *et al*. (2020) used two backcrosses to introgress bmr6 allele for biomass from the donor parent CMSXS170 line to elite lines CMSXS652 and IS23 of sweet sorghum using SNP markers. Results revealed a successful transfer of the bmr6 allele to the recurrent parent for biomass improvement. For successful backcrosses, the promising genotypes selected for traits of interest should contain favourable alleles either one of the parents and must be expressed phenotypically. Backcrosses must be done at least three to four times for sufficient transfer of the QTLs from the donor to the recurrent parents followed by successive selfing until the backcross line is close to the original recurrent parent (Da Silva *et al*., 2020). SNPs markers narrow the specific location of QTLs which is closely linked to gene expressing STG trait. The SNP markers identify favourable alleles for STG expression thus; fasten screening of drought tolerance sorghum genotypes. The current study identified favourable alleles for STG that were screened for further studies of sorghum improvement. It was noted that; some of the plants performed well in the field condition by expressing STG, low leaf rolling, high yield and high vigour of grains sorghum but were not linked by SNP markers used for genotyping. This indicates that there is need to map large number of SNP markers to widen chances of identifying the right STG QTLs. If SNP markers fail to locate the position of the QTL expressing STG in the chromosomes, SSR and DArT markers can alternatively be used (Kiranmayee *et al*., 2020). Moreover, findings have reported that there are some QTLs which are detected by molecular

markers in the backcrosses but fail to express under field condition; such genotypes do not qualify for selection of crop improvement (Platten *et al*., 2019). Differential transcriptome analysis encompasses bioprocess to map genes controlling the expression of drought tolerance in sorghum (Azzouz-Olden *et al*., 2020). The STG expression on drought tolerant sorghum varieties is due to the reaction of genes which increase the production of antioxidant capacity, regulatory factors and the repressors of early senescence (Azzouz-Olden *et al*., 2020). The application of modern technology supplements conventional breeding techniques to address drought stress in sorghum. The modern technology for instance, molecular markers map traits of interest which save time of breeding cycles. In this study, SNP markers identified favourable alleles for STG QTLs from the genotypes of BC_2F_1 and BC_2F_3 population which simplified comparison with field data to screen the best plants with traits of target. It is important to use markers that are powerful and reliable to identify the favourable alleles for STG, QTLs and chromosome position of genotypes for inclusion in drought tolerance sorghum improvement.

4.7. Conclusions and recommendations

Eighteen out of 30 SNP markers used for genotyping BC_2F_1 population indicated favourable alleles for STG 1 STG 2 and STG 3 among the sorghum genotypes employed in this study. Seven out of 10 SNP markers used for genotyping BC_2F_3 population indicated favourable alleles for STG 1 STG 2 and STG 3. These markers are recommended for screening drought tolerance QTLs in sorghum. This study recommends further mapping of SNPs markers that are tightly linked to STG QTLs for drought tolerance in sorghum. This will help to widen chances of identifying favourable alleles for STG in sorghum. Further marker assisted backcrossing by SNPs markers is recommended to enhance transferring further important target traits from the donor parents to the farmers' cultivated sorghum varieties in Tanzania.

CHAPTER FIVE

5.0. DETERMINATION OF PHYSIOLOGICAL TRAITS CONTRIBUTING TO DROUGHT TOLERANCE OF SORGHUM

5.1. Introduction

Drought tolerance in sorghum is enhanced by different genes; each gene contributes part to drought tolerance (Phuong *et al*., 2019). Breeding in sorghum for drought tolerance trait focuses on the incorporation of many traits, for instance; lower leaf canopy, leaf rolling, and reduced transpiration. These traits are negatively correlated with yield in sorghum during drought stresses (Ali *et al*.., 2009). Plants with deep root system show resistance to post-flowering drought stress under limited water conditions. Deep roots help plants to uptake sufficient water and nutrients required for plants (Khatab *et al*., 2017). Sorghum root system is well developed that helps to absorb water and nutrients from the soil for survival of the crop under limited water condition (Naoura *et al*., 2019). On top of that, leaf dry matter, root biomass, flag leaf area, leaf weight, grain yield and plant height are among the parameters used for multivariate analysis to screen drought tolerant crops (Misra *et al*., 2002; Khan *et al*., 2004; Kapanigowda *et al*., 2013; Jabereldar *et al*., 2017). Multivariate analysis helps to identify traits which contribute drought tolerance in sorghum (Negarestani *et al*., 2019). Combinations of different traits contribute to drought tolerance in sorghum for instance; plants with high root length and root dry weight have higher drought tolerance (Brunner *et al*., 2015). Other traits used for multivariate analysis of drought tolerance in sorghum include; above ground dry matter, specific leaf area and heading date (Phuong et al., 2019). Furthermore, leaf rolling is used as an indicator to identify drought tolerant plants (Kadioglu and Terzi, 2007). In sorghum, lines with high tolerance to drought show leaf rolling under

limited moisture content in the soil. Leaf rolling is correlated with leaf water potential, a leaf starts to roll following reduction of leaf water potential however; it depends on varieties (Amelework *et al*., 2015). Plants adjust osmotic water potential under low water leaf potential to reduce leaf rolling during drought stress condition which favours plant physiological growth and final yield (Morka, 2015). Leaf rolling trait is used for screening drought tolerance and high yield in sorghum. Drought tolerant varieties show reduced leaf rolling due to effective adjustment of osmotic potential in low leaf water potential under water stress. Plants reduce leaf area, root biomass, dry matter content and yield in drought condition as the mechanism of survival (Maqsood and Ali, 2007). In addition, traits performance can be estimated by the heritability which indicates proportion of variations which is important for selection of the lines (Waqar-Ul-Haq *et al*., 2008). High heritability for days to flowering, panicle length, days to maturity and plant height has been reported in sorghum (braha *et al*., 2015). Drought indices such as MP, GMP, SSI, STI and tolerance index (TOL) are also useful for determination of trait performance in sorghum which simplifies selection of best breeding materials of sorghum (Kharrazi and Rad, 2011). Highly drought tolerant lines produce relative higher traits of targets compared to susceptible lines. Therefore, during screening for drought tolerance, lines which are tolerant to drought are screened for improvement to produce new varieties.

The objectives of this study were to:

i. determine yield performance of sorghum lines under well irrigated and non-irrigated;
ii. identify physiological traits contributing to post-flowering drought tolerance in sorghum;
iii. determine indices of drought tolerance in sorghum lines; and
iv. determine heritability of traits in sorghum population developed.

5.2. Materials and Methods

5.2.1 Location of the study

The study was conducted at TARI- Makutupora Centre located at 22 km North of Dodoma municipality in Dodoma region (Longitude: 35º, 46.093'E and Latitude: 05º, 58.669'S) (Altitude: 1070 m). The annual rainfall ranges from 300-500 mm with poor distribution, temperature varies from 15-35.1 °C (TMA- 2014). The area is classified as semi-arid which is characterized by mono-modal rainfall pattern. The area gets rainfall between December and April of the year.

The management of BC_2F_1 plants were collected for genotyping until pre-flowering. Plants were self-pollinated before flowering to avoid outcrossing. Each plant was selfed using pollination bag until the soft dough stage. Seeds were harvested from plants of each BC_2F_1 populations. Plants with the best performance in terms of heading, grain size and other yield related traits were chosen for generating BC_2F_2 populations. A total of six genotypes from BC_2F_1 populations, that is, W82, SE408, SE438, NA307, NA316 and NA241 with B35*Wahi, B35*Seguifa, and B35*NACO Mtama 1 parents background were selected as the best genotypes under genotypic and phenotypic evaluation to generate BC_2F_2 populations. These genotypes were planted on 29th December, 2019 with the spacing of 0.75 m between rows and 0.3 m within rows. Agronomic management such as fertilizers application, weeding, pest and diseases control were conducted. Prior to pre-flowering, BC_2F_2 plants were self-pollinated. Birds were controlled by scaring in support with the use of mosquito net that were used to cover the panicle to reduce pressure of birds attacking from grain filling to early physiological plant maturity. Phenotypic data including number of seeds per panicle, plant height, and number of leaves were recorded from the breeding site. Plant height was measured at the distance from the ground to the top of panicle using ruler/tape measure. Leaf

length, leaf width, panicle length and panicle width and panicle exsertion length was measured using a ruler. Data collection was recorded to identify the plants that were responding well on yield related traits which were then used for field evaluation. A total of eight genotypes from BC_2F_2 populations include NA241A, NA241B, NA316A, NA316B, NA316C, NA307, SE408 and SE438 were selected as the best genotypes from BC_2F_2 populations for generating BC_2F_3 populations.

5.2.2. Experimental design

The split plot on the complete randomized block design (CRBD) with three replications and twelve entries per replication was used for this study. The total area for each trial was 1000 m^2. Genotypes of BC_2F_3 sorghum populations were planted in five rows per replication with spacing of 0.75 x 0.3 m between row and within row respectively. The genotypes were evaluated in well watered irrigation and stressed water environments in one location. The main treatment in the main plot was irrigation regimes while sub treatment in the sub-plot was sorghum genotypes of BC_2F_3 populations. Well watered irrigation trial was fully irrigated from planting to 50% physiological maturity. Water irrigation was withheld in the water stressed trial at days to 50% flowering stage to physiological maturity. The yield related traits were evaluated in well watered and stressed plots.

The interaction between sorghum genotypes generated and irrigation regimes were determined using model $Y_{ij} = \mu+g_i+e_j+ (ge)_{ij} +e_{ij}$ where; Y_{ij} is the measured mean of i^{th} genotype and j^{th} environment, μ is the grand mean, g_i is the main effect of genotype, e_j is the main effect of environment, ge_{ij} is the interaction of i^{th} genotype and j^{th} environment, e_{ij} is an experimental error associated with i^{th} genotype and j^{th} environment. 5.5.2 Evaluating physiological traits contributing to drought tolerance in sorghum

5.2.3. Determination of indices of drought tolerance in sorghum genotypes

Six selection indices including stress susceptibility index (SSI), stress tolerance index (STI) (Fernandez, 1992), stress tolerance (TOL) (Hossain *et al*., 1990), mean productivity (MP) (Hossain *et al*., 1990), geometric mean productivity (GMP) (Fernandez, 1992), stress intensity (SI) and yield stability index (YSI) were calculated based on grain yield under drought-stressed and irrigated conditions. Stress tolerance attributes were computed based on the formula:

$SSI = [\ 1-(\ Ys\ )\ /\ (\ Yp\ )\ ]\ /\ SI.$

SI is the stress intensity and calculated as:

$SI = [\ 1-(\ \bar{Y}s\ )\ /\ (\ \bar{Y}p\ )\ ]$

$GMP = \sqrt{\ (\ YP \times YS\ )}$

$TOL = (\ Yp - Ys\ )$ and

$MP = (\ Yp + Ys\ )\ /\ 2$

$STI = [\ (\ Yp\ ) \times (\ Ys\ )\ /\ (\ \bar{Y}p\ )^2\ ]$ (Fernandez, 1992)

$YSI = Ys\ /\ Yp$

$Reduction\ (\%) = (\ Yp - Ys\ )\ /\ Yp$

Where Ys and Yp are the yields of B_2F_3 populations assessed under water stress and normal irrigation conditions trials and $\bar{Y}s$ and $\bar{Y}p$ are the mean yields over all populations assessed in two conditions (Fernandez, 1992; Hossain *et al*., 1990).

5.3. Determination of heritability of traits among sorghum parents

The heritability of donor and recurrent parents was determined based on the interaction between genotypes and environments. The interactions of sorghum lines with the environment were determined based on the phenotypic parameters that were collected as described in Chapter 3. Broad sense heritability was computed using the formula below;

The broad sense heritability (H) of the parent lines was computed as follows;

$H = \sigma^2 g\ /\ (\ \sigma^2 g + \sigma^2 gxe + \sigma^2 e\ )$

Where, $\sigma^2 g$ = genotypic variance, $\sigma^2 gxe$ = genotype × environment variance, and $\sigma^2 e$ = environmental variance (residual error).

Genotypic variance was computed as follows;

$$\sigma^2 g = (MSg - MSgxe - MSe) / re$$

Where, MSg = mean square of the genotypes, MSgxe = mean square of the genotype x environment interactions, MSe = mean square of the residual error (environmental variance), r =number of replications, and e = number of environments.

Genotype x environment interaction variance was computed as follows;

$$\sigma^2 gxe = (MSgxe-MSe) / r$$

5.4. Data collection and statistical data analysis

Ten plants were tagged with labels per plot for data collection. The parameters recorded for drought tolerance indices and yield related traits were root biomass, stem biomass, chlorophyll content, 1000 seed weight, STG, Inflorescence exsertion, panicle weight. Yield of grain under full irrigation and stress managed trials were measured using a digital weighing balance. Ten sample plants per plot were used for root and stem biomass evaluation. Root biomass was collected below ground level of the plant, stem biomass was taken from above the ground to the base of panicle, and these two parameters were subjected to oven drying at 70^{oC} for three days. Chlorophyll content was measured from single leaf on top, middle and bottom part of the plant using atLEAF chlorophyll meter where the average chlorophyll content of three leaves was recorded. Leaf rolling was scored using a scale of I to 5 where I - no leaf rolling, 2 - low leaf rolling, 3 - intermediate leaf rolling, 4 - High leaf rolling and 5 – extremely high leaf rolling (death of leaves). STG was scored using scale from 1 to 9 where 1- very low or no visible sign of leaf drying, 3 - low, 5 - intermediate, 7 -

high and 9 - very high. Inflorescence exsertion was recorded using the scale of 1 to 4 where 1 - slightly exserted (<2 cm but ligule of flag leaf definitively below inflorescence base), 2 - exserted (2-10 cm between ligule and inflorescence base) 3 - well-exserted (>10 cm between ligule and inflorescence base), 4 - peduncle recurved (inflorescence below ligule and clearly exposed splitting the leaf sheath). Total number of green leaves per plant at physiological maturity was counted in both full irrigation and water stress trials based on ICRISAT sorghum descriptors.

Multivariate statistical analyses such as the principal component analysis were computed by using GENSTAT 12th edition, SAS software version 9.4 and "R" software of the current version. The mean separation was determined using least significance difference at 5% confidence interval.

5.5. Results

Results showed segregation among populations evaluated after selfing BC_2F_1 generation. Ten percent of the total plants planted per BC_2F_1 populations were screened for the best performing of traits (appendix 3. Table 6.1, 6.2 and 6.3). A total of six genotypes (NA307, NA241, NA 316, SE408, SE438 and W82) were selected from the best 10% of screened plants (Table 5.1).

Table 5. 1. Selected best performing BC2F2 seeds of sorghum genotypes from BC2F1 populations

Background of parent	Genotype	PH	STG	GLM	PL	PW	IEX	GWT
B35*NACO_NA241	NA241	146	5	2	24	8	2	113.4
B35*NACO_NA307	NA307	147.8	5	3	26	8.5	4	147.3
B35*NACO_NA316	NA316	145	5	5	24	8	3	154.2
B35*Seguifa_SE408	SE408	143.8	5	4	20	7	3	114.7
B35*Seguifa_SE438	SE438	147.3	5	3	21.5	7	3	103.4
B35*Wahi_W82	W82	123	5	2	34	8	2	101

PH- Plant height (cm), STG- Stay green, GLM- Total number of green leaves at maturity, PL- Panicle length (cm), PW- Panicle width (cm), IEX- Inflorescence exersion, GWT- Grain weight (g), NACO- NACO Mtama 1

5.5.1 Grain yield performance of new sorghum populations under well and stressed water environments

In water irrigation environment, the population NA316C yielded the highest with the mean yield of 3415 kg/ha followed by NA307 with the mean yield of 3163 kg/ha, Seguifa (recurrent parent) mean yield of 2911 kg/ha and SE 438 with 2770 kg/ha (Table 5.2). The lowest yield was recorded by the donor parent B35 with mean yield of 1770 kg/ha. Under stress environments, the best performing genotypes included SE438 with mean yield of 2652 kg/ha followed by NA316C with mean yield of 2585 kg/ha) and NA307 with 2556 kg/ha. The genotype B35 resulted in the lowest mean yield (1711 kg/ha) of all populations evaluated.

Table 5. 2. Grain yield of sorghum within genotypes under normal water irrigation and water stress conditions

	Grain yield (kg/ha)	
Population	Water irrigated	Water stressed
NA241A	2430	2081
NA241B	2430	2170
NA307	3163	2556
NA316A	2400	2252
NA316B	2296	2230
NA316C	3415	2585
SE408	2044	2296
SE438	2770	2652
Wahi	2030	2415
NACO	2630	1978
Seguifa	2911	2326
B35	1770	1711
SE	0.008	0.005
LSD	0.016	0.009
CV (%)	54.5	34.3

SE- Standard error mean, LSD- Least significance difference of the mean, CV(%)- Coefficients of variations, NACO- NACO Mtama 1

5.5.2 Distribution of chlorophyll content in sorghum genotypes

The evaluation of chlorophyll content distribution indicated that the highest content was found in the middle leaves of sorghum plant in both environments. Results in the water stressed environment indicated that the donor parent B35 expressed the highest chlorophyll content on both positions of the plant with 59.04 g/l at the top leaf, 60.71 at the middle and 51.4 g/l at the bottom leaf (Table 5.3). The genotype SE438 expressed the highest chlorophyll content (47.18, 52.14 and 44.96 g/l) at top, middle and bottom leaves of the plant respectively. Majority of the genotypes (NA316C, NA307, NA241B, SE408, Seguifa, NACO Mtama 1, Wahi, S438 and B35) had chlorophyll content above 30 g/l in all parts of the plant which has been recommended for sorghum. However, the overall chlorophyll content was higher in donor parent and the recurrent parents except genotype SE438.

Results in the well watered environment showed above 35 g/l of chlorophyll content on top, middle and bottom leaves of all sorghum genotypes. The genotypes SE438, NA316C and the

parents B35, Seguifa resulted in above 46 g/l of chlorophyll content in both parts of the plant. B35 showed the highest chlorophyll content (66.01, 63.94 and 58.23 g/l) on top, middle and bottom plant leaves of all genotypes evaluated in well watered environment (Table 5.3).

Table 5. 3. Mean values of top, middle and bottom leaves of chlorophyll content on sorghum genotypes in stressed and un-stress environments

	Stress environment				Un-stress environment		
Genotypes	CH-Top leaf	CH-Middle leaf	CH-Bottom leaf	Genotypes	CH-Top leaf	CH-Middle leaf	CH-BOT leaf
NA241A	27.29a	34.97ab	29.91a	NA241A	39.77a	46.89abc	40.67abc
NA316B	29.75a	34.38a	31.97a	NA316B	48.14bc	49.97bcd	42.23abc
NA316A	29.99a	38.62abc	33.63a	NA316A	46.52abc	50.91bcd	44.56bc
NA316C	31.46ab	38.72abc	32.88a	NA316C	50.99c	56.57de	46.26c
NA307	33.49abc	43.16bc	34.79a	NA307	44.46abc	46.53abc	40.54abc
NA241B	33.70abcd	40.93abc	38.10ab	NA241B	39.84a	42.28a	35.44a
SE408	39.49bcde	43.20bc	38.01ab	SE408	41.85ab	44.46ab	35.99ab
Seguifa	40.05bcde	42.43abc	36.75ab	Seguifa	52.23c	53.77cd	47.80c
NACO	40.98cde	44.61cd	38.85ab	NACO	42.44ab	50.14bcd	41.36abc
Wahi	42.67de	46.86cd	35.29a	Wahi	47.03abc	49.15abcd	39.04abc
SE438	47.18e	52.14de	44.96bc	SE438	47.80abc	51.09bcd	43.73abc
B35	59.04f	60.71e	51.46c	B35	66.01d	63.94e	58.23d
SE	2.65	2.56	2.74	SE	2.41	2.23	2.6
LSD	5.21	5.04	5.39	SLD	4.73	4.39	5.12
CV (%)	27	22.9	28.5	CV (%)	19.7	17.1	23.5

[a,b,c,d,e] and [f] are comparisons for the Bonferroni mean test, BOT- Bottom, CH- Chlorophyll content (g/l), CV (%)- Coefficient of variations of means, SE- Standard error of deviation, LSD- Least Significance difference of means, NACO- NACO Mtama 1

There were significant differences in traits (plant height, leaf rolling, grain weight per plant, panicle weight per plant and STG) of sorghum genotypes at $P < 0.001$ under water irrigation and water stress treatments (Table 5.4 and 5.5). The mean performance of plant height was the highest (142.2 cm) in population NA316 followed by NA316B with 139 cm in the stressed environment. The donor parent exhibited the lowest (95.5 cm) plant height in the watered stress trial; however, there were no significant differences among populations tested. In the watered irrigation trial, the genotype NA316A, NA316C indicated the high performance of plant height with 143.3 cm and140.3 cm while

the lowest plant height (94.6 cm) was recorded in the donor parent B35. There were significance differences of leaf rolling in the genotypes NA241B, NA316A, NA316BB, SE408, local check Wahi and the donor parent B35 in the water stressed trial (Table 5.4). The score of leaf rolling for the genotypes NA241A, NA316C, NA307, NACO Mtama 1, Seguifa and SE438 varied but were not significant. The genotypes SE438 and NA316C scored low leaf rolling in both environments. The parents Seguifa and NACO Mtama 1 indicated low score in water irrigation and the genotype NA307 had lower score leaf rolling than the rest of the genotypes genotypes in water stressed condition.

In this study, there were variation ($P < 0.05$) of the total number of leaves counted at physiological plant maturity among sorghum genotypes evaluated in irrigated and water stressed conditions. The genotype NA307 showed the highest total number of leaves across the environments. The donor parent B35 and check Wahi showed higher total number of leaves than backcross genotypes except genotype NA307. The population NA241B showed the lowest (1.97) total number of leaves.

Each backcross genotype showed lower total number of green leaves than check and donor parent B35 across the environments. The total number of green leaves at maturity was significantly affected by water irrigation and water stress environments. The highest number (3.5) of green leaves was recorded from the donor parent B35 and check Wahi (2.7) in the stressed environment where the genotypes NA241B and NA316A showed higher (2.1 and 1.7) total number of green leaves than the rest (Table 5.5).

The genotypes NA316C and NA316A showed 3.87 and 4.2 rating of STG similar to the check Wahi and B35 with 3.73 and 4.13 rating of STG respectively in irrigated environment (Table 5.4). The genotype NA241B and NA316A had 5.6 and 5.8 rating of STG similar to the check Wahi which showed 5.7 STG under water stressed environment (Table 5.4). The

genotype NA316A performed well across the environments. The lowest STG rating (5.8) was recorded from the genotype NA241B in water irrigated and the lowest STG (7.3) in the genotype NA241A in water stressed environment.

Based on the root biomass, genotypes NA316C and NA316A produced higher (68.33 and 51.67 g) than other backcross genotypes except parents B35 and Seguifa which produced the highest root biomass in water irrigated condition (Table 5.4). The lowest (40 g) root biomass was reported on genotype SE408 in unstressed conditions. Similarly, there were significant differences among genotypes evaluated in the stressed condition whereas, check Wahi recorded the highest (58.3 g) root biomass followed by B35 (48.3 g). The genotypes SE438 and NA307 (45 g) performed well in root biomass accumulation. The genotypes NA241A and SE408 were significantly affected by post flowering drought, resulting in less than 37 g of root biomass (Table 5.5).

The yield of genotypes NA307, NA316C and S438 per panicle weight per plant in well watered irrigation and in water stressed environments were 0.88 kg, 0.8 kg, 0.74 kg and 0.79 kg, 0.7 kg and 0.74 kg respectively (Tables 5.4 and 5.5). The panicles per plant showed significant differences among the genotypes at $P < 0.05$.

The genotype NA316C and NA307 produced high yield across the environments. Under water irrigation, genotypes NA316C recorded (0.077 kg) grain yield per plant and NA307 (0.071 kg) in well watered condition (Table 5.4). These genotypes produced the same amount (0.58 kg) of root biomass per plant in water stressed condition (Table 5.5).

Table 5. 4. Means of traits performance of sorghum genotypes under well watered irrigation

Genotype	PH	LR	NLM	GLM	STG	RB	PWT	GWT
NA241A	132.6cd	1.9abc	6.73ab	2.90abc	4.93abc	45.00abc	0.073abcde	0.055abc
NA241B	134.9cd	2.0bc	6.87ab	1.90a	5.80c	43.33ab	0.071abcde	0.055abc
NA316A	143.3d	1.8abc	6.23a	2.70abc	4.20ab	51.67bc	0.073bcde	0.054abc
NA316B	134.4cd	1.9abc	6.13a	2.43ab	5.00abc	46.67abc	0.067abcd	0.052abc
NA316C	140.3cd	1.7ab	6.63a	3.33bcd	3.87a	68.33d	0.080de	0.077c
NA307	133.9cd	2.0bc	7.77c	2.57abc	5.60bc	46.67abc	0.088e	0.071bc
SE408	129.2c	2.1c	6.83ab	2.87abc	5.00abc	40.00a	0.056abc	0.046ab
SE438	138.6cd	1.7ab	6.73ab	3.13bc	4.07a	48.33abc	0.074bcde	0.062abc
Wahi	111.9b	2.0bc	7.70c	4.37d	3.73a	48.33abc	0.054ab	0.046ab
NACO	136.4cd	1.6a	6.13a	2.77abc	4.80abc	51.67bc	0.075cde	0.059abc
Seguifa	135.4cd	1.7a	6.70ab	3.07bc	4.53abc	53.33c	0.079de	0.066abc
B35	94.6a	1.9abc	7.50bc	3.57cd	4.13ab	73.33d	0.052a	0.040a
SE	3.6	0.1	0.24	0.33	0.44	2.9	0.006	0.008
LSD	7.08	0.19	0.47	0.64	0.86	5.71	0.012	0.016
CV	10.7	20.3	13.6	42.6	36.3	21.9	32.7	54.5

[a,b,c,d] and [e] are comparisons for the Bonferroni mean test, CV- Coefficient of variation, GWT- Grain weight per plant (kg), GLM- Total number of green leaves at maturity, LSD- Least significance difference of means, LR- Leaf rolling, NLM- Total number of leaves at maturity, PWT- Panicle weight per plant (kg), RB- Root biomass, SE- Standard error of means, STG- Stay green

Table 5. 5. Mean values of traits of sorghum genotypes under water stress condition

Genotype	PH	LR	NLM	GLM	STG	PWT	GWT	RB
NA241A	138.3b	2.3b	6.9ab	1.3ab	7.3e	0.06abc	0.05ab	33.3a
NA241B	135.3b	2.03ab	6.5a	2.1bc	5.6ab	0.064abcd	0.05ab	41.7bcd
NA316A	134.5b	1.98ab	7.0ab	1.7ab	5.8abcd	0.066bcd	0.051ab	41.7bcd
NA316B	139.0b	2.03ab	6.9ab	1.5ab	6.7bcde	0.062abcd	0.05ab	41.7bcd
NA316C	142.2b	1.85a	6.9ab	1.2a	6.9de	0.07bcd	0.058b	40abc
NA307	135.1b	1.8a	7.6bc	1.3ab	6.9cde	0.079d	0.058b	45cd
SE408	134.2b	2.1ab	7.0ab	1.6ab	6.3bcde	0.064bcd	0.052ab	38.3abc
SE438	135.9b	1.8a	6.7ab	1.6ab	6.1bcde	0.074cd	0.06b	45cd
Wahi	106.9a	1.97ab	8.1c	2.7cd	5.7abc	0.071bcd	0.054b	58.3e
NACO	136.2b	1.9a	6.4a	1.4ab	6.7bcde	0.064bcd	0.052ab	36.7ab
Seguifa	134.5b	1.9a	6.9ab	1.3ab	6.3bcde	0.054ab	0.045ab	36.7ab
B35	95.5a	2.1ab	7.5bc	3.5d	4.8a	0.046a	0.039a	48.3d
SE	3.39	0.11	0.26	0.26	0.36	0.005	0.005	2.08
LSD	6.66	0.22	0.51	0.52	0.7	0.01	0.009	4.1
CV	10	22	14.3	57.9	22.1	32	34.3	19.1

[a,b,c,d] and [e] are comparisons for the Bonferroni mean test, CV- Coefficient of variation, GWT- Grain weight per plant (kg), GLM- Total number of green leaves at maturity, LSD- Least significance difference of means, LR- Leaf rolling, NLM- Total number of leaves at maturity, PWT- Panicle weight per plant (kg), RB- Root biomass, SE- Standard error of means, S (1- full leaf with green Colour, 9-Complete leaf death)

The mean square values of various traits evaluated between genotypes by environments revealed interactions (at $P < 0.05$ and $P < 0.01$) (Table 5.6). The interaction were recorded from eight traits including days to 50% flowering, leaf rolling, chlorophyll content, total number of leaves at physiological plant maturity, total number of green leaves at plant maturity, STG, panicle weight, grain weight per panicle and root biomass. There was no interaction between genotypes by environments for trait of plant height and grain weight. The mean squares for genotypes were significant (at $P < 0.05$ and $P < 0.01$) for all traits except root biomass. Majority of the traits evaluated for mean squares performance between genotypes by environments showed significant differences except plant height and grain yield per plant.

Table 5. 6. Mean squares for performance of sorghum genotypes in well water and watered stressed environments

SV	DF	DFW	PH	LR	CH	GLM	STG	RB	GWT
Rep	2	129.38	2430**	0.8	1700**	6*	11*	2224**	0.006**
Gen	11	1263.39**	11389**	1.1**	2359**	18.9**	18**	2437**	0.003**
Env	1	79.33	6	1.8**	13432**	260**	469**	15116**	0.006*
Gen xEnv	11	97.97**	266	0.4*	405**	7**	10**	1777**	0.001
Residual	694	19.83	187	0.2	59.43	1.4	2	101	0.0001
Total	719								

*, ** Significant differences at p ≤0.05 and p ≤0.01 respectively, DF- Degree of freedom, DFW- Days to 50% flowering, PH- Plant height (cm), LR- Leaf rolling, CH- Chlorophyll content (g/l), GLM- Total number of green leaves, STG- Stay green, RB-Root biomass (g), GW- Grain weight per plant (kg), SV- Sources of variation, Rep- Replication, Gen- Genotype, Env- Environment.

5.5.3 Correlation of drought tolerance and yield related traits in sorghum

There were positive and negative correlation coefficients among traits contributing to drought tolerance and yield in sorghum populations (Table 5.7). The traits of panicle weight, panicle width and panicle length were significantly correlated with grain yield. STG and inflorescences exsertion were negatively correlated with grain yield. Panicle width and panicle weight were strongly correlated to grain yield and each other. Chlorophyll content was positive and significantly correlated with total number of green leaves at maturity. Plant height was correlated with panicle weight however the correlation was low with grain yield. STG and inflorescences exsertion showed negative correlation with most traits. However, both showed non-significant positive correlation with plant height. The highest negative correlation was between traits STG and total number of green leaves at maturity and STG and chlorophyll content.

Table 5. 7. Correlation coefficients of traits contributing to drought tolerance and grain yield of sorghum genotypes under water irrigated and water stressed environments

Trait	GW	PWT	PH	NLM	BM	NGLM	PW	IEX	CH	STG	PL
GW	-										
PWT	0.72**	-									
PH	0.27**	0.41**	-								
NLM	0.08	0.17**	-0.13**	-							
BM	0.26**	0.29**	-0.21**	0.04	-						
NGLM	0.18**	0.2**	-0.2**	0.21**	0.39**	-					
PW	0.53**	0.75**	0.5**	0.04	0.17**	0.06	-				
IEX	-0.17**	-0.18**	0.2**	-0.07	0.01	0.1	-0.08	-			
CH	0.25**	0.27**	-0.26**	-0.02	0.6	0.53**	0.2**	0.1	-		
STG	-0.21**	-0.22**	0.09	-0.03	-0.41**	-0.84**	-0.11	-0.15**	-0.56**	-	
PL	0.35**	0.51**	0.18**	1	0.29**	0.07	0.46**	-0.09	0.14**	-0.1	-

Note: BM= Root biomass, IEX- Inflorescence exsertion, GW= Grain weight per panicle (kg), PWT= Panicle weight (kg), PH= Plant height (cm), CH= Chlorophyll content (g/l), STG= Stay green, NGLM= Total number of green leaves at maturity, NLM= Total number of leaves at maturity, PL- Panicle length (cm), PW- Panicle width (cm).

5.6. Heritability estimates

The broad sense heritability estimates varied from 88.2% of plant height to 64.2% of grain weight in water irrigated condition and from 89.1% of plant height to 65% of leaf rolling in water stressed condition (Table 5.8). The traits of stem biomass, root biomass, 1000 seed weight and chlorophyll content recorded above 85% of broad sense heritability estimates in both environments. The average heritability for both environments was about 80%.

Table 5. 8. Broad sense heritability (%) of parents and backcrosses

Trait	Water irrigation	Water stress
Plant height (cm)	88.2	89.1
Stem biomass (kg)	87.3	89.7
Chlorophyll content (g/l)	85.9	88.4
Root biomass (g)	87.1	86.9
1000 seed wt	86	86.3
Panicle length (cm)	84.7	88.5
Leaf width (cm)	85.7	83.6
Number of green leaves at maturity	78.9	84.4
Number of leaves at maturity	83.4	78.3
Panicle width (cm)	82	70.4
Inflorescence exsertion	74.3	77.4
Panicle weight (g)	76.1	75.3
Leaf senescence	71	79.5
Leaf length (cm)	75.2	75.1
Leaf rolling	74	65
grain weight (g)	64.2	65.8

Plant height showed the highest (89.52%) broad sense heritability of all traits determined by days to 50% flowering (80.88%) and 80% of stem biomass between genotypes by environments. The root biomass showed the lowest broad sense heritability (12.38%) followed by STG (17.64%) across the environments (Table 5.9).

Table 5. 9. Heritability of sorghum genotypes traits accross water management environments

Trait	Heritability (%)
Plant height (cm)	89.52
Days to 50% flowering	80.88
Stem biomass (kg)	80
Chlorophyll content (g/l)	64.39
Leaf width (cm)	62.33
Total number of leaves at maturity	62.26
Panicle length (cm)	61.26
Panicle width (cm)	43.67
Grain weight per plant (g)	42.86
Leaf length (cm)	38.95
1000 seed weight (g)	38.4
Total number of green leaves at maturity	34.86
Panicle weight (g)	30
Inflorescence exsertioon	29.36
Leaf rolling	23
STG	17.64
Root biomass	12.38

5.7. Principal components analysis of the traits

PCA of grain yield under water stressed and well watered condition showed two principal components of which PC1 explained 85.99% of the total variation while the PC2 explained 14.01% of the variation (Figure 5.1)

Based on grain yield results, the highest MP and GMP were recorded for populations NA316C (3415 and 2585 kg/ha) and NA307 (3163 and 2556 kg/ha), suggesting genotypes variation for yield performance across the environments (Table 5.10). Yield based on STI indicated that the population NA316C (with mean yield 3415 and 2585 kg/ha), NA307 (3163 and 2556 kg/ha and SE438 (2770 and 2652 kg/ha) performed the best across the environments with the STI values (1.39, 1.27 and 1.15) respectively. The highest values (830,

652 and 607) of TOL were recorded in the populations NA316C (3415 and 2585 kg/ha) Seguifa (2630 and 1978 kg/ha and NA307 (3163 and 2556 kg/ha). The lowest values (-385,-252 and 66) were recorded in the populations Wahi with mean yield of 2030 and 2415, SE408 (2044 and 2296 kg/ha) and NA316B (2296 and 2230 kg/ha. These populations performed differently across the environments.

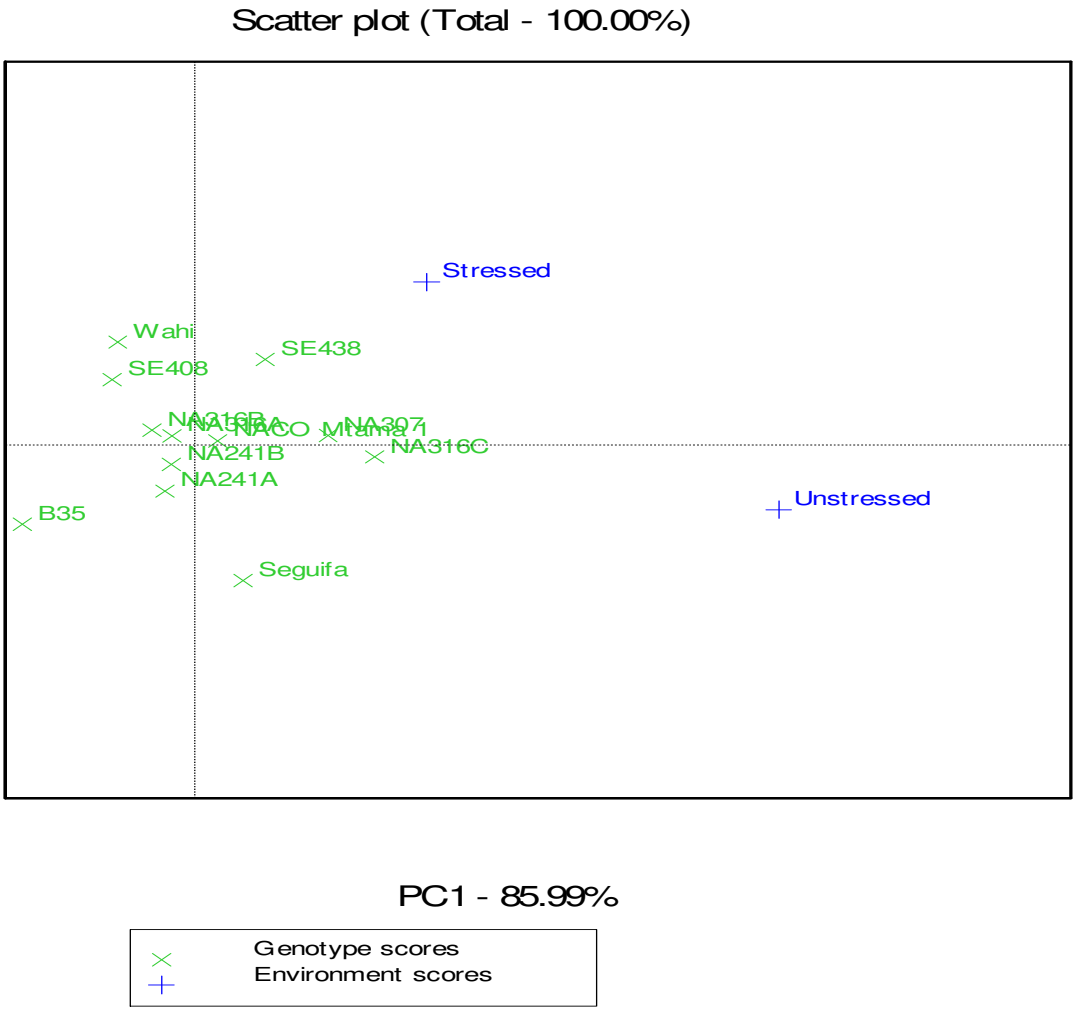


Figure 5. 1. Principal component analysis of (PCA) of grain yield under water stressed and well watered conditions

Correlation coefficients were used as tool to determine the promising criteria for drought tolerant populations (Table 5.11). The indices GMP, MP were highly correlated with YP and

YS and each other. TOL was positively correlated with yield by 0.83 under water irrigated environment and low correlation with yield by 0.12 under water stressed condition. The contrasting correlation between TOL and YP and YS suggested the reduction of grain yield under water irrigation conditions thus is not recommended for selection of promising populations for drought tolerance indices.

Table 5. 10. Drought stress indices and yield under water irrigated and water stressed conditions

Population	YP	YS	TOL	MP	GMP	SI	SS1	STI	YSI	Red (%)
NA241A	2430	2081	349	2255.5	2248.7	0.14	1.03	0.79	0.86	14.4
NA241B	2430	2170	260	2300	2296.3	0.11	0.97	0.83	0.89	10.7
NA307	3163	2556	607	2859.5	2843.4	0.19	1.01	1.27	0.81	19.2
NA316A	2400	2252	148	2326	2324.8	0.06	1.03	0.85	0.94	6.2
NA316B	2296	2230	66	2263	2262.8	0.03	0.97	0.8	0.97	2.9
NA316C	3415	2585	830	3000	2971.2	0.24	1.01	1.39	0.76	24.3
SE408	2044	2296	-252	2170	2166.3	-0.12	1.03	0.74	0.12	-12.3
SE438	2770	2652	118	2711	2710.4	0.04	1.08	1.15	0.96	4.3
Wahi	2030	2415	-385	2222.5	2214.2	-0.19	1	0.77	1.19	-19
Seguifa	2630	1978	652	2304	2280.8	0.32	0.78	0.82	0.75	24.8
NACO	2911	2326	585	2618.5	2602.1	0.12	1.68	1.06	0.8	20.1
B35	1770	1711	59	1740.5	1740.3	0.03	1.1	0.48	0.97	3.3

Note: NACO- NACO Mtama 1, YS- Yield under water stress condition, YP- Yield under normal irrigation conditions, TOL- Tolerance inde, MP- Mean productivity,GMP- Geometric mean productivity, SSI- Stress susceptibility index, STI- Stress tolerance index, Red (%)- Percentage reduction.

There were no significant correlations between SSI and the rest of indices. SSI is the best indices for selection of the populations with low yield and drought tolerance to water stressed conditions. YSI was strongly correlated with STI and negatively correlated with YP, TOL, MP, GMP and SSI. Red was only positively correlated with TOL and MP, rest of the indices were negatively correlated and non-significant. TOL was positively correlated with MP and GMP but not strongly correlated with YP. Furthermore, SSI indicated non-significant

correlation with MP and GMP. The lowest correlation was found between Red and YSI and indices between YSI and TOL.

Table 5. 11. Correlation coefficients of drought stress indices and yield under water irrigated and water stressed conditions

Indices	YP	YS	TOL	MP	GMP	SS1	STI	YSI	Red
YP	-								
YS	0.648*	-							
TOL	0.832**	0.116^{ns}	-						
MP	0.955**	0.846**	0.628*	-					
GMP	0.95**	0.853**	0.617*	0.997**	-				
SS1	0.168^{ns}	0.129^{ns}	0.125^{ns}	0.168^{ns}	0.171^{ns}	-			
STI	-0.599*	0.118^{ns}	-0.866**	-0.373**	-0.362^{ns}	0.322^{ns}	-		
YSI	-0.748^{ns}	0.009^{ns}	-0.982**	-0.52**	-0.511^{ns}	-0.109^{ns}	0.897**	-	
Red	0.748^{ns}	-0.009^{ns}	0.982**	0.52**	0.511^{ns}	0.109^{ns}	-0.897**	-1**	-

Note: *= Significance at P= 0.05, ** = significant at P= 0.01, ns= not significance at 5% or 1% of probability level, YS- Yield under water stress condition, YP- Yield of sorghum grain under normal irrigation conditions, TOL-Tolerance index, MP-Mean productivity, GMP- Geometric mean productivity, SSI-Stress susceptibility index, STI-Stress tolerance index, Red- Percentage reduction, YSI- Yield stability index

Four principal axes were generated in the study of PCA in drought tolerance indices (Table 5.12). The first principal axis (PC1) accounted for 59.1% of the total variation and positively correlated with YP, YS, TOL, MP, GMP, SI, SSI, STI and Red (Table 5.12). The first dimension of principal component may be selected as the best indicator of yield potential and drought tolerance in this study. The population with high values of PC1 can be high yielding in the water irrigated and water stressed environments. The second principal axis (PC2) showed 20.8% of the variation and explained the positive correlation with TOL, SI, YSI and Red however were negatively correlated with YP, YS, MP, GMP, SSI and STI (Table 5.12). PC3 indicated 10% variation explaining positive correlation of most of the indices except TOL and SSI. The findings revealed that SSI did not separate the drought-susceptible cultivars. However, the populations with highest PCA1 and lowest PCA2 showed good yield performance in both environments. The rest of principal axes were below 10% of proportion.

Table 5. 12. Principal component analysis of potential yield (YP), yield under stressed (YS) and drought tolerance

Variable	PC1	PC2	PC3	PC4
YP	0.411	-0.013	0.002	0.007
YS	0.261	-0.527	0.033	0.124
TOL	0.346	0.367	-0.022	-0.08
MP	0.39	-0.215	0.014	0.053
GMP	0.389	-0.224	0.018	0.052
SI	0.289	0.486	0.008	0.104
SS1	0.071	-0.129	-0.098	-0.97
STI	0.39	-0.21	0.017	0.051
YSI	-0.02	0.017	0.994	-0.1
Red	0.314	0.441	0.006	-0.1
Eigenvalue	5.905	2.082	0.999	0.987
Proportion	0.591	0.208	0.1	0.099
Cumulative	0.591	0.799	0.899	0.997

YP = yield under well-watered conditions, YS = yield under drought-stress, TOL = tolerance index, MP = Mean productivity, GMP = Geometric mean productivity, SSI = stress susceptibility index, STI = stress tolerance index, Red = percentage reduction, YSI- Yield stability index, SI- Stress intensity

There was correlation of angles between vectors in the biplot diagram of PCA (Figure 5.2). There was similarity between STI and MP and strong correlation with GMP. YP and YS were distantly correlated indicating that water stress reduced the yield.

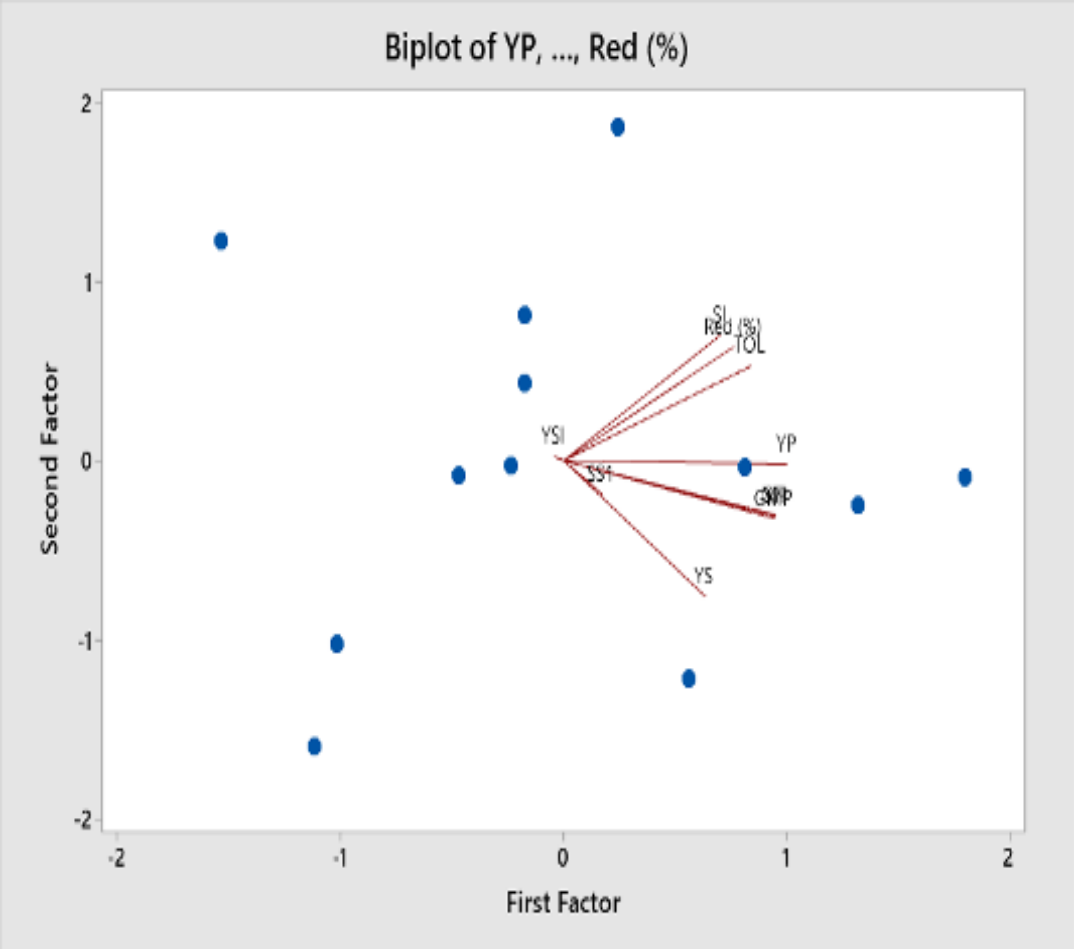


Figure 5. 2. Biplot diagram of PCA of genotypes on yield irrigated (YP) and water stressed conditions

5.8. Discussion

Chlorophyll content is an important trait which determines the ability of a plant to photosynthesize food for use. The current study noted variation of chlorophyll content in plant parts. The chlorophyll content distribution in the plant differs from one part to another depending on factors like age and temperature (Dwyer *et al*., 1991). It is concentrated more in the centre portion of the sorghum plant than top and bottom parts (Dwyer *et al*., 1991). This aligns with the current study which noted higher distribution of chlorophyll content at the middle part of the plant than top and bottom parts. Results showed that the bottom leaves of the plant have the lowest chlorophyll content because leaf senescence starts at that part.

Chlorophyll content reduces with increase of water stress which affects water and nutrients uptake from the soil to the plants. In sorghum, chlorophyll content is helpful especially during post flowering drought to enable a plant manufacture its own food for use and maintain physiological plant growth and grain yield. The presence of high chlorophyll content during drought stress condition delays the process of leaf senescence to retain STG which accounts for grain yield on sorghum. Xu *et al*. (2000) reported the correlation of leaf chlorophyll content and STGT in sorghum during post flowering drought suggesting the use of these traits for screening drought tolerance and yield in sorghum. The amount of chlorophyll content recorded in all parts of the donor parent B35 and the introgressed genotypes in our study indicate that the donor parent B35 contributed part of the improvement. Although water stress reduced chlorophyll content in all plant parts compared to irrigated condition, majority of the genotypes showed above 30 g/l chlorophyll content however, the highest chlorophyll content in the donor parent indicates that the introgression of STG to the recurrent parent was not exploited enough to some of the genotypes for improvement. The reduction of chlorophyll content during post flowering drought initiates leaf senescence and lowers STG (Li *et al*., 2019). The resistance of sorghum genotypes delay leaf senescence to allow grain filling and other physiological processes to take place (Wanous *et al*., 1991).

Drought is the major constraint of crop production globally. This study noted different responses of genotypes in stressed and non-stressed environments. The yield performance of sorghum crop depends on the genetic diversity, heritability and genetic gain of the generated population. The traits associated with yield in sorghum include biomass, plant height, days to 50 flowering, days to 50% maturity, number of tillers per plant, number of grains per panicle and panicle weight per plant (Sadia *et al*., 2018). These traits are considered when screening high yielding sorghum. Grain yield is influenced by biotic and abiotic factors. Abiotic stresses such as

drought significantly reduces grain yield of sorghum (Wenzel, 1999). STG 1, 2, 3 and 4 QTLs linked to drought tolerance in sorghum have been useful for introgression to non-stay green sorghum to enhance grain yield during post flowering drought stress (Sabadin *et al*., 2012). The QTLs introgressed to non- STG parents contribute to grain yield increase in water stressed condition. Nonetheless, it depends on the period of dry spell and genetic makeup of genotypes (De Souza *et al*., 2020). The current study recorded the increase of grain yield by 156 kg (7%) under water stressed environments. The difference of performance of grain yield is affected by water management where water irrigation enhances physiological plant growth and yield. Similarly, Ajeigbe *et al*. (2018) reported that, sorghum grain yield depends on amount of water available in the soil for plant use. Low water content hinders roots and leaves growth which is important for water and nutrients uptake and photosynthesis. However, this study identified few genotypes which performed better in water stress environment than in the well watered condition; this is contrary to Sory (2015) who recorded higher yield in irrigated than in water stressed condition. The genotypes with high grain yield in both environments for example, the genotypes NA307 and NA316C in this study indicate that these genotypes can perform well in the multi-location trials in the next step of evaluation. These genotypes may contain high genetic potential which may perform well in physiological growth across environments.

Environmental conditions determine the performance of traits in plant growth. Plant height, days to flowering, root biomass and chlorophyll content of plants respond differently at different environments. Traits of sorghum plant respond different in water managements, some respond positively in one environment and negatively in another environment. Traits which show good response in contrasting environments are suggested for screening of drought tolerant genotypes in sorghum. Water stress reduces chlorophyll content which affects photosynthesis efficiency and grain yield of sorghum (Zhang *et al*., 2019). The current study noted higher chlorophyll content in water irrigation environment than in water stressed condition implying that water management influenced the results. Genotypes with high chlorophyll content delay leaf senescence under limited water. The ability to maintain

chlorophyll contents depends on type of drought. The expression of STG during drought condition prior to flowering and after flowering of plants is the indication of drought tolerance which is contributed by STG. Nonetheless, in water stress conditions plants express STG before physiological maturity. Findings of this study revealed that, STG expression is affected by water stress than in water irrigation condition because of reduction of moisture in the soil. In the study of STG introgression in non-STG sorghum, Kassahun *et al*. (2010) found success introgression of STG from donor parent B35 to the adapted senescence sorghum varieties which suggest the uses of MABC for improvement of post flowering drought tolerance in sorghum. In addition, the balance between nitrogen for grain filling and nitrogen released by parts of the plant and absorbed by root accounts for STG expression in sorghum during water scarcity (Borrell *et al*., 2000b). Plants with plenty of nitrogen content have ability to delay leaf senescence for maintaining photosynthesis for a long period which helps to produce sufficient carbohydrates for developing grains. STG in sorghum has been associated with high grain yield in drought prone environments where post flowering drought is the major challenge (Borrell *et al*., 2000a). Chlorophyll content and STG are important for evaluation of drought tolerant sorghum under water management environments. Findings by Zaeifizade and Goliov (2009) revealed that maize genotypes with high drought tolerance are associated with high chlorophyll content and grain yield compared to drought susceptible genotypes. The current study indicates that plants respond differently in plant height under water irrigation and water stress conditions. The variation of plant height is due to genetic difference of genotypes in the same condition. In addition, this study recorded the majority of the introgression genotypes had higher root biomass at post flowering than the recurrent parents but less than check genotypes and the donor parent B35 at water stress condition indicating the contribution of donor parent to improve target traits. Kassahun *et al*. (2010)

reported similar findings where backcross genotypes had higher root biomass than senescence parents and positive correlation between root biomass and STG.

The interaction of genotypes by environments plays the major role in the study of genetic and phenotypic variance in crop improvement. The genetic and phenotypic variances determine the variation of performance on traits of sorghum in different treatments. Mean squares computed for various traits of sorghum genotypes by environments showed significant differences at $p \leq 0.05$ and $p \leq 0.01$ respectively except plant height and grain yield per plant. The interactions were influenced by water stress environment which limits growth of most parts of the plant. The traits which are positively influenced by the interaction of full irrigation and stressed treatments have been considered as the best indices of drought tolerance in sorghum. The current study noted low variation of plant height of the sorghum genotypes evaluated in the contrasting environments; this could be because of recording days to flowering in the same environmental condition at days to 50% flowering before imposing stressed environments. The same trend was on grain yield except that the mean yield of grain per plant was high in water irrigation environment. Similarly, the interaction of water irrigation and water stress did not influence root biomass and grain yield per plant, this could be due to insufficient transfer of STG QTLs from the donor parent to the recurrent parent, variation of soil nutrients and water uptake by the plant. The interaction of water management and genotypes accounts for grain yield at post-flowering growth stages. Plants with deep root systems can access water from deep soil in water stressed condition to support plant growth and the high grain yield. This study reports significant interactions of genotypes performance of STG in sorghum.These plants can be used as the basis for improvement of drought tolerance and yield in crops including sorghum.

The interaction of genotypes and environments reduces the broad sense heritability of target traits. Broad sense heritability estimates are used to screen variability of traits performance of

genotypes. It focuses on traits such as, grain yield, chlorophyll content and panicle length, root biomass, dry matter, plant height, panicle weight and STG and number of seed per plant (Efisue, 2006). Plant height and days to 50% flowering expressed higher heritability compared to other traits in crops. Variability of heritability estimates in the genotypes in this study was influenced by the interactions among genotypes by environments. Plant height has been useful for estimation of heritability in various crops including sorghum (Phuke *et al*. 2017). Heritability estimates depend on the level at which trait gene is inherited from the parents to the siblings. Hamidou *et al*. (2018) reported high heritability in sorghum for traits of grain yield, plant height and panicle weight. These are similar to this study where the results reported ≥ 80% broad sense heritability of plant height, days to 50% flowering and stem biomass which indicates these traits were not influenced by the environments. Nevertheless, the percentage of heritability in root biomass, STG and leaf rolling were below 25% because of environmental influences (Kiranmayee *et al*., 2020). The exploitation of variability present in the genotypes depends on heritability estimates of the traits evaluated. The interaction of genotypes and water managements in the current study reduces broad sense heritability estimates. The reduction of heritability estimates is due to variation of performance of genotypes in the contrasting environments. Plants respond different in different environments which affects phenotypic and genotypic expression of the genotypes (Ye *et al*., 2006).

In the study of drought tolerance indices, STI index appeared to be the best drought tolerance indicator which is suggested for the screening of drought tolerant genotypes. The genotype NA316C which produced the highest grain yield across the environments however its TOL was the highest too. These findings contradict with report by Sory (2015) who reported high values of TOL in sorghum and concluded that such genotypes were susceptible to drought stress. Therefore, further studies are needed to confirm the findings.

Principal component analysis is essential for grouping genotypes based on the performance of traits of interest. It shows the relationship among genotypes which enable selection of the best traits for different purposes. The study of drought tolerance indices of sorghum in full irrigation and water stress environments identify the genotypes which adapt well in both environments. Basically plants grow well in sufficient water condition compared to limited water condition. However, in sorghum significant variation occurs at post flowering drought which reduces yield. Interestingly in this study, some genotypes performed better in stressed environments than in well watered. This finding suggests that the variation could be due to variation of soil nutrients which are common in semi-arid areas. Various studies (Sory, 2015; Ouedraogo *et al*., 2017) have shown higher grain yield in full irrigation of sorghum when water is withheld at 50% post-flowering growth stage. It is possible for some genotypes to perform well in stressed environments after withholding water at post flowering because moisture retains for some days which support plant growth.

5.9. Conclusions and recommendations

Water irrigation and water stress environments influenced the variation of traits performance. The interaction of genotypes by environment influenced the performance of various traits. The heritability was higher in well watered than in the water stressed conditions but was low on the interaction of genotype by environments. Chlorophyll content in sorghum leaves vary from one part of plant to another but the concentration is highest in the middle part of the plant. For the best screening of plants with drought tolerance traits in sorghum, chlorophyll content is one of the important traits. The genotypes NA307, NA316C and SE438 were selected as the best performing in terms of grain yield across the environments. These genotypes should be further tested for multi-location trials to evaluate the performance before recommending as new varieties.

CHAPTER SIX

6.0. GENERAL CONCLUSIONS AND RECOMMENDATIONS

6.1. General conclusions

In the present study, the constraints of sorghum production identified included bird damages, poor soil fertility, and drought stress. Majority of farmers in Tanzania cultivate local sorghum varieties which are tolerant to storage pests and own seed saving because of unavailability of improved seed of the sorghum crop and high cost of inputs.

The drought coping mechanism practiced by farmers included early planting, use of drought tolerant varieties, planting early maturing varieties. The traits preferences chosen by farmers include drought tolerance, early maturity and high yield. Farmers have knowledge to identify preferred traits of sorghum genotypes suggesting that plant breeders should considered them when planning for breeding new improved sorghum varieties.

There was limited number of extension officers in the villages to satistify extension services to farmers, the available officers failed to train large numbers of farmers due to limited facilitation.

In the study of MABC 71 (10%) out of 728 BC_2F_1 samples collected for genotyping was heterozygous. Eighteen SNP markers showed favourable alleles for STG QTLs and other roles contributing to STG in sorghum. The markers nsSB00049 and nsSB00054 expressed favourable alleles for STG 1, and nsSB00102 and nsSB00103 expressed for STG 3. The favourable alleles identified for STG QTL in the current study showed that, the genotypes generated by marker assisted backcrossing was partly successful.

Three samples of genotypes W82, NA241 and NA307 with heterozygous alleles and two samples of NA316 and SE438 genotypes with homozygous alleles were selected as the best genotypes from BC_2F_1 population after comparing field and genotypic data, these samples were planted as genotypes for advancing to BC_2F_3 populations.

The genotypes NA241A, 241B, NA307, NA316A, NA316B, NA316C, NA408 and NA438 of BC_2F_3 seeds were selected as the best for grain yield, grain vigour and heading from BC_2F_2 populations.

The donor parent B35 contributed to the variation of yield recorded between three genotypes NA307, NA316C, SE438 and the recurrent parents and check in well water and water stressed environments. Three genotypes (NA307, NA316C, and SE438) showed genetic gain after introgression of STG QTL from B35 compared to the recurrent parents NACO Mtama 1 and Seguifa, these genotypes will contribute to increase sorghum productivity when approved as new sorghum varieties in Tanzania.

6.2. General recommendations

Plant breeders should consider drought tolerance, pest and diseases resistance and other related traits when they develop improved sorghum varieties to stimulate farmers' adoption rate. To further address these constraints, collaboration among plant breeders, pathologists, entomologists, socio-economists, soil scientists, extension officers, local government authorities, and the ministry of agriculture is imperative to enhance sorghum productivity.

There is need to strengthen sorghum value chain to diversify use of end products and market options among stakeholders as these will motivate farmers to expand farms for sorghum production.

The government should increase the number of extension officers to facilitate the uptake of new technologies innovated by researchers which will help to increase the productivity of sorghum.

Studies are needed to map further SNP markers which are highly polymorphic and closely linked to QTLs associated with drought tolerance genes. These will increase efficiency of identifying QTLs that are linked to STG and expand genetic variation between parents and backcross populations. Further backcrosses for enhancing STG QTLs in sorghum are needed to widen the possibility of transferring trait of target.

Based on heritability information, the traits plant height and days to 50% flowering which showed the highest heritability are recommended as the basis for differentiating of sorghum populations.

STI is recommended as the indice for screening drought tolerance of sorghum genotypes. Because of contradiction of different reports on the acceptable TOL in crops further studies are needed to find the relevant conclusion.

The genotypes NA307, NA316C and SE438 performed well in terms of grain yield per hectare across the environments. Therefore, these genotypes should be tested in multi-location trials to evaluate the distinction, uniformity and stability before recommended for release as the new varieties.

References

Adu-Boakyewaa, G.A., Badu-Apraku, B., Akromah, R., Garcia-Oliveira, A.L., Awuku, F.J., Gedil, M. (2019). Genetic diversity and population structure of early-maturing tropical maize inbred lines using SNP markers. *PLoS ONE*, 14, e021481

African Agricultural Technology Foundation (AATF). (2011). Feasibility study on Striga control in sorghum. Nairobi, AATF

Ahmed, I.M., Nadira, U.A., Cao, F., He, X., Zhang, G. and Wu, F.J.P. (2016). Physiological and molecular analysis on root growth associated with the tolerance to aluminium and drought individual and combined in Tibetan wild and cultivated barley. *Plant*, 243(4): 973-985

Ajambo, R., Elepu,g., Bashaasha, B. and Okori, P. (2017). Farmers' preferences for maize attributes in eastern and western Uganda. *African Crop Science Journal*, 25(2): 177 – 187

Ajeigbe, H.A., Akinseye,F.M., Ayuba,K. and Jonah, J. (2018). Productivity and Water Use Efficiency of Sorghum [*Sorghum bicolor* (L.) Moench] Grown under Different Nitrogen Applications in Sudan Savanna Zone, Nigeria. *International Journal of Agronomy*, 2018: Article ID 7676058, 11 pages https://doi.org/10.1155/2018/7676058

Agrama, H.A. and Tuinstra, M.R. (2003). Agrama, H.A. and Tuinstra, M.R. (2003). Phylogenetic diversity and relationships among sorghum accessions using SSRs and RAPDs. Afric. J. Biotech. 2003; 2(10): 334-340. *African Journal of Biotechnology*, *2*(10), 334–340

Ahlawat, S. et al. (2008). Genotypic divergence analysis for stay green characters in Wheat (*Triticum aestivum* L. em. Thell). *South Pacific Journal of Natural Sciences*, (26):73-81 2008

Akhare, A.A., Sakhare, S.B., Kulwal, P.L., Dhumale, D.B. and Kharkar, A. (2008). RAPD Profile studies in Sorghum for identification of hybrids and their parents. *International Journal of Integrative Biology*, 3(1): 18

Ali, M.A., Abbas, A., Awan, S.I., Jabran, K. and Gardezi, S.D.A. (2011). Correlated response of various morpho-physiological characters with grain yield in sorghum landraces at different growth phases. *Journal of Animal and Plant Sciences*, 21(4), 671–679

Amelework, B., Shimelis, H., Tongoona, P., and Laing, M. (2015). Physiological mechanisms of drought tolerance in sorghum, genetic basis and breeding methods: A review. *African Journal of Agricultural Research*, 10(31), 3029–3040

Amelework, B.A., Shimelis, H.A., Tongoona, P., Mengistu, F., Laing, M.D. and Ayele, D.G. (2016). Sorghum production systems and constraints, and coping strategies under drought-prone agro-ecologies of Ethiopia. *South African Journal of Plant and Soil* 33(3): 207–217

Amoah, J.N. and Antwi-Berko, D. (2020). Comparative Physiological, Biochemical and Transcript Response to Drought in Sorghum Genotypes. *Biotechnology Journal International,* 24(3): 1-14

Ansarifard, I., Mostafavi, K., Khosroshahli, M., Bihamta, M.R. and Ramshini, H. (2020). A study on genotype–environment interaction based on GGE biplot graphical method in

sunflower genotypes (*Helianthus annuus* L.). *Food Science & Nutrition,* 8(7): 3327-3334

Arunkumar, B., Biradar, B.D and Salimath, P.M. (2004). Genetic Variability and Character Association Studies in Rabi Sorghum. *Karnataka Journal of Agricultural Sciences*, 17(3): 471-475

Ashikari, M. and Matsuoka, M. (2006). Identification, isolation and pyramiding of quantitative trait loci for rice breeding. *Trends Plant Science*, 11: 344–350

Ashraf, M., Athar H.R., Harris P.J.C. and Kwon T.R. (2008). Some prospective strategies for improving crop salt tolerance. *Advances in Agronomy,* 97: 45–110. 10.1016/S0065-2113(07)00002-8

Assefa, A. B. (2012). Genetic diversity analysis of lowland sorghum [*Sorghum bicolor* (L.) Moench] landraces under moisture stress conditions and breeding for drought tolerance in North Eastern Ethiopia. (Thesis). University of Kwazulu-Natal. Retrieved from http://researchspace.ukzn.ac.za/handle/10413/9876

Assefa, Y., Staggenborg, S.A. and Prasad, V.P.V. (2010). Grain Sorghum Water Requirement and Responses to Drought Stress: A Review (PDF Download Available). *Crop Management*, 11. https://doi.org/doi:10.1094/CM-2010-1109-01-RV

Assefa, Y., and Staggenborg, S.A. (2011). Phenotypic changes in grain sorghum over the last five decades. *Journal of Agronomy and Crop science*, 197: 249–257

Ayana, A. and Bekele, E. (2000). Geographical patterns of morphological variation in sorghum (*Sorghum bicolor* (L.) Moench) germplasm from Ethiopia and Eritrea:

Quantitative characters. *Euphytica*, 115(2): 91–104 https://doi.org/10.1023/A:1003998313302

Azzouz-Olden, F., Hunt, A.G. and Dinkins, R. (2020). Transcriptome analysis of drought-tolerant sorghum genotype SC56 in response to water stress reveals an oxidative stress defense strategy. *Molecular Biology Reports*, 47(5):3291-3303. doi: 10.1007/s11033-020-05396-5

Badigannavar, A., Teme, N., de Oliveira, A.C., Li, G., Vaksmann, M., Viana, V.E., Ganapathi, T.R. and Sarsu, F. (2018). Physiological, genetic and molecular basis of drought resilience in sorghum [*Sorghum bicolor* (L.) Moench]. *Indian Journal of Plant Physiology*, 23: 670–688. https://doi.org/10.1007/s40502-018-0416-2

Ballesta, P., Mora, F., Alejandro Del Pozo, A.D. (2018). Association mapping of drought tolerance indices in wheat: QTL-rich regions on chromosome 4A. *Scientia Agricola*, 77(2): DOI: http://dx.doi.org/10.1590/1678-992X-2018-0153

Baloch, F.S., Alsaleh, A., Shahid, M.Q., Çiftçi, V., de Miera, L.E.S, Aasim, M., Nadeem, M.A., Aktaş, H., Özkan, H., Hatipoğlu, R. (2017). A Whole Genome DArTseq and SNP Analysis for Genetic Diversity Assessment in Durum Wheat from Central Fertile Crescent. *Plos One*, 12(1): e0167821. https://doi.org/10.1371/journal.pone.0167821

Baret, F., Madec, S., Irfan, K., Lopez, J., Comar, A., Hemmerlé, M., Dutartre, D., Praud, S. and Tixier, M.H. (2018). Leaf-rolling in maize crops: from leaf scoring to canopy-levelmeasurements for phenotyping. *Journal of Experimental Botany*, 69(10): 2705–2716

Barnaud, A., Trigueros, G., McKey, D. and Joly, H.I. (2008). High outcrossing rates in fields with mixed sorghumlandraces: how are landraces maintained?. *Heredity*, 101: 445-452

Barron, J., Rockström, J., Gichuki, F. and Hatibu, N. (2003) Dry spell analysis and maize yields for two semi-arid locations in East Africa. *Agriculture for Meteorology*, 117 (1–2): 23–37

Basavaraj, G., P. Parthasarathy Rao, P.P., Lagesh, L.A., Pokharkarj, V.G., Gupta, S.K. and Kumar, A.A. (2015). Understanding Trait Preferences of Farmers for Post-Rainy Sorghum and Pearl Millet in India - A Conjoint Analysis. *Indian Journal of Agricultural Economy*, 70(l):130-143

Bernardino, K. C., Pastina, M.M., Menezes, C.B., de Sousa, S.M., Maciel, L.S., Jr, G.C., Guimarães, C.T. Barros, B.A., Silva, L.D.,Carneiro, P.C.S., Schaffert, R.E., Kochian, L.V. and Magalhaes, J.V. (2019). Bernardino et al. The genetic architecture of phosphorus efficiency in sorghum involves pleiotropic QTL for root morphology and grain yield under low phosphorus availability in the soil. *BMC Plant Biology*, 19:87 https://doi.org/10.1186/s12870-019-1689-y

Bibi, A., H. A. Sadaqat, H. M. Akram, and M. I. Mohammed. (2010). Physiological markers for screening sorghum (*Sorghum bicolor*) germplasm under water stress condition. *International Journal of Agriculture and Biology*, 12: 451–455

Bibi, A., Sadaqat, H.A., Tahir, M.H.N. and Akram, H.M. (2012). Screening of sorghum (*Sorghum bicolor* Var moench) for drought tolerance at seedling stage in polyethylene glycol. *The Journal of Animal & Plant Sciences,* 22(3): 671–678

Blum, A. (2005). Drought resistance, water-use efficiency, and yield potential—are they compatible, dissonant, or mutually exclusive? - Google Search. *Australian Journal of Agricultural Research*, 56:1159–1168

Blum, A. (2004). Sorghum physiology. In 'Physiology and biotechnology integration for plant breeding'. (Eds Nguyen, H.T. and Blum, A.) pp. 141–223. Marcel Dekker, New York

Blum, A., Mayer, J. and Golan, G. (1989). Agronomic and physiological assessments of genotypic variation for drought resistance in sorghum. *Australian Journal of Agricultural Research*, 40(1): 49. https://doi.org/10.1071/AR9890049

Boczkowska, M., Baczek, K.B., Kosakowska, O., Rucin´ ska, A., Podyma, W. and Weglarz, Z. (2020). Genome-Wide Diversity Analysis of Valeriana o_cinalis L. Using DArT-seq Derived SNP Markers. *Agronomy*, 10: 1346. doi:10.3390/agronomy10091346

Borrell, A.K. and Hammer, G.L. (2000). Nitrogen dynamics and the physiological basis of stay green in sorghum. *Crop Science*, 40(5): 1295–1307

Borrell, A.K., Hammer, G.L., Douglas, A.C.L. (2000a). Does maintaining green leaf area in sorghum improve yield under drought? I. Leaf growth and senescence. *Crop Science*, 40:1026–1037

Borrell, A.K., Hammer, G.L., and Henzell, R.G. (2000b). Does maintaining green leaf area in sorghum improve yield under drought? II. Dry matter production and yield, *Crop Science*, 40(4): 1037-1048

Borrell, A.K., van Oosterom, E.J., Mullet, J.E., George-Jaeggli, B., Jordan, D.R., Klein, P.E., and Hammer, G.L. (2014). Stay-green alleles individually enhance grain yield in

sorghum under drought by modifying canopy development and water uptake patterns. *New Phytologist*, 203(3): 817–830. https://doi.org/10.1111/nph.12869

Boyles, R.E., Brenton, Z.W. and Kresovich, S. (2019). Genetic and genomic resources of sorghum to connect genotype with phenotype in contrasting environments. *The Plant Journal*, 97: 19–39. doi: 10.1111/tpj.14113

Bradeen, J.M. and Simon, P.W. (1998). Conversion of an AFLP fragment linked to the carrot *Y2* locus to a simple, codominant, PCR-based marker form. *Theoritical and Applied Genetic*, 97: 960-967

Brown, D. (2013). Contribution of sorghum production towards household food security in Tanzania: A case study of Singida region. Sokoine University of Agriculture, Morogoro, Tanzania Brooks, S., Thompson, J., Odame, H., Kibaara, B., Nderitu, S., Karin, F. and Millstone, E. (2009). *Environmental change and maize innovation in Kenya: Exploring pathways in and out of maize'* (STEPS Working Paper 36). Brighton. Retrieved from https:// ww.ids.ac.uk/files/dmfile/STEPSWorking Paper 36

Brunner, I, Herzog, C., Dawes, M, Arend, C. and Sperisen, C. (2015). How tree roots respond to drought. *Frontiers in Plant Sciences*, https://doi.org/10.3389/fpls.2015.00547

Bucheyeki, T.L., Shenkalwa, E.M., Mapunda, T.X. and Matata, L.W. (2010). Yield performance and adaptation of four sorghum cultivars in Igunga and Nzega districts of Tanzania. *Communications in Biometry and Crop Science*, 5(1): 4–10

Burow, G., Chopra, R., Sattler, S., Burke, J., Acosta-Martinez, V. and Xin, Z. (2019). Deployment of SNP (CAPS and KASP) markers for allelic discrimination and easy

access to functional variants for brown midrib genes bmr6 and bmr12 in *Sorghum bicolor*. *Molecular Breeding*, 39:115 https://doi.org/10.1007/s11032-019-1010-7

Burow, G., Franks, C.D., Xin, Z. and Burke, J.J. (2012). Genetic Diversity in a Collection of Chinese Sorghum Landraces Assessed by Microsatellites. *American Journal of Plant Sciences*, 3(12): 1722-1729

Calviño, M., Miclaus, M., Bruggmann, R. and Messing, J. (2009). Molecular Markers for Sweet Sorghum Based on Microarray Expression Data. *Rice*, 2:129–142

Casa, A.M., Pressoir, G., Brown, P., Mitchell, S.E., Rooney, W.L., Tuinstra, M.R., Franks, C.D. and Kresovich, S. (2008). Community resources and strategies for association mapping in sorghum. *Crop Science*, 48:30–40

Cochran, W.G. (1963). Sampling Techniques. 2nd ed. New York: John Wiley and Sons, Inc

Condon, A.G., Richards, R.A., Rebetzke, G.J. and Farquhar, G.D. (2002). Improving intrinsic water-use efficiency and crop yield. *Crop Science*, 42: 122–131

Costa, R., Pereira, G., Garrido, I., Tavares-de-Sousa, M.M. and Espinosa, F. (2016). Comparison of RAPD, ISSR, and AFLP Molecular Markers to Reveal and Classify Orchardgrass (Dactylis glomerata L.) Germplasm Variations. *PLoS ONE*, 11(4): e0152972. doi:10.1371/journal.pone.0152972

Craufurd, P.Q. and Peacock, J.M. (1993). Effect of Heat and Drought Stress on Sorghum (Sorghum Bicolor). II. Grain Yield. *Experimental Agriculture*, 29(1): 77–86

Da Silva, M.J., Pastina, M.M., de Souza, V.F., Schaffert, R.E., Carneiro, P.C.S., Noda, R.W, et al. (2017) Phenotypic and molecular characterization of sweet sorghum accessions

for bioenergy production. *PLoS ONE*, 12(8): e0183504. https://doi.org/10.1371/journal.pone.0183504

Da Silva, M.J., Ribeiro, P.C.O., Silva, R.A., Silva, K.J., Schaffert, R.E., Parrella, R.A.C. (2020). Multi-trait selection of sweet sorghum (Sorghum bicolor (L.) Moench) genotypes for bioenergy production. *Journal of Bioenergy and Food science*, 7(4): e2952020. http://doi.org/10.18067/jbfs.v7i4.295

Derese, S.A., Shimelisa, H., Laing, M. and Mengistu, F. (2017). The impact of drought on sorghum production, and farmer's varietal and trait preferences, in the north eastern Ethiopia: implications for breeding, *Acta Agriculturae Scandinavica, Section B — Soil & Plant Science*, DOI: 10.1080/09064710.2017.1418018

De Souza, A.A., Carvalho, A.J., Bastos, E.A., Portugal, A.F., Torres, L.G., Batista, P.S.C., Julio, M.P.M.,, Julio, B.H.M. and de Menezes, C.B. (2020). Grain Sorghum Grown Under Drought Stress at Pre- and Post-flowering in Semiarid Environment. *Journal of Agricultural Science*; 12(4)

Dial, H.L. (2012). Plant guide for sorghum (*Sorghum bicolor* L.*)*. USDA-Natural Resources Conservation Service, Tucson Plant Materials Center, Tucson, AZ

Diale, N.R. (2011). Socio-economic indicators influencing the adoption of hybrid Sorghum: The Sekhukhune District perspective. *South African Journal of Agricultural Extension*, 39(1): 75-85

Dicko, M.H., Gruppen, H., Traore, A.S., Voragen, A.G.J. and van Berkel, W.J.H. (2006). Sorghum grain as human food in Africa: relevance of content of starch and amylase activities. *African Journal of Biotechnology*, 5(5): 384-395

Dimkpa, C.O. and Bindraban, P.S. (2016). Fortification of micronutrients for efficient agronomic production: a review. *Agronomy for Sustainable Development*, 36(1): 7. 10.1007/s13593-015-0346-6. hal-01532372

Disasa, T., Feyissa, T., Admassu, B., Paliwal, R., deVilliers, S.M. and Odeny, D.A. (2016). Molecular evaluation of Ethiopian sweet sorghum germplasm and their contribution to regional breeding programs. *Australian Journal of Crop Science*, 10(4): 520–527

Dwyer, L.M., Tollenaar, M. and Houwing, L. (1991). A Nondestructive Method to Monitor Leaf Greenness in Corn. *Canadian Journal of Plant Science*, 71: 505–509

ECARSAM, 2005. Sorghum and Millet Research for Development in Eastern and Central Africa 2005-2010, Regional Priorities

Edema, R. and Amoding, G.L. (2015). Validating simple sequence repeat (SSR) markers for introgression of stay-green quantitative trait loci (QTLs) into elite sorghum lines. *African Journal of Biotechnology*, 14(46): 3101–3111

Efisue, A.A. (2006). Studies of drought tolerance in interspecific progenies of *Oryza glaberrima* (steud) and O. *Sativa* (L) and an appraisal of the use of male gametocides in rice hybridisation. PhD thesis University of KwaZulu-Natal, South Africa

Ejeta G. 2007. The Striga Scourge in Africa: a growing pandemic. In: Integrating new technologies for Striga control: towards ending the witch-hunt. Singapore: *World Scientific*; p. 3–16

Ejeta, G. and Knoll, J.E. (2007). Marker-assisted selection in sorghum In: Varshney, R.K. and R. Tuberosa (ed.) Genomic-assisted crop improvement: *Genomics Applications in Crops*, 2: 187-205

FAOSTAT. (2017). Database of agricultural production. Rome: Food and Agriculture Organization of the United Nations. Available at http://faostat.fao.org/default.aspx

FAOSTAT. (2018). Database of agricultural production. Rome: Food and Agriculture Organization of the United Nations. Available from: http://faostat.fao.org/default.aspx

Fakrudin, R.B., Kavil, S.P., Girma, Y., Arun, S.S., Dadakhalandar, D., Gurusiddesh, B.H., Patil, A.M., Thudi, M., Bhairappanavar, S.B., Narayana, Y.D., Krishnaraj, P.U., Khadi, B.M. and Kamatar, M.Y. (2013). Molecular mapping of genomic regions harbouring QTLs for root and yield traits in sorghum (*Sorghum bicolor* L. Moench). *Physiology and Molecular Biology of Plants*, 19(3): 409-419

Fasahat, P., Rajabi, A. Rad, J.M. and Derera, J. (2016). Principles and Utilization of Combining Ability in Plant Breeding. *Biometrics & Biostatistics International Journal*, 4(1), 1–24. https://doi.org/10.15406/bbij.2016.4.00085

Fernandez, G.C.J. (1992). Effective Selection Criteria for Assessing Plant Stress Tolerance. In: Proceedings of the International Symposium on Adaptation of Vegetables and Other Food Crops in Temperature and Water Stress (Kuo, C.G, ed.). Publication, Tainan, Taiwan.

Fiust, A., Rapacz, M., Wójcik-Jagła, M. and Tyrka, M. (2015). Development of DArT-based PCR markers for selecting drought-tolerant spring barley. *Journal of Applied Genetics*, 56: 299–309. DOI 10.1007/s13353-015-0273-x

Fu, J. and B. Huang. (2001). Involvement of antioxidants and lipid peroxidation in the adaptation of two cool-season grasses to localized drought stress. *Environmental and. Experimental Botany*, 45: 105–114.doi:10.1016/S0098-8472 (00)00084-8

Gaufichon, L., Prioul, J. and Bachelier, B. (2010).What are the prospects for genetic improvement in drought-tolerant crop plants?. FARM, November 2010

Gebretsadik, R., Shimelis, H., Laing, M., Tongoona, P. and Mandefro, N. (2014). A diagnostic appraisal of the sorghum farming system and breeding priorities in Striga infested agro-ecologies of Ethiopia. *Agricultural Systems*, 123: 54–61

Getnet, Z., Husen, A., Fetene, M. and Yemata, G. (2015). Growth, Water Status, Physiological, Biochemical and Yield Response of StaynGreen Sorghum (*Sorghum bicolor* (L.) Moench) Varieties-A Field Trial Under Drought-Prone Area in Amhara Regional State, Ethiopia. *Journal of Agronomy*, 14(4): 188-202. doi: 10.3923/ja.2015.188.202

Gierend, A., Ojulong, H., Letayo, E., Mgonja, F.M. (2014). A combined ex-post/ex-ante impact analysis for improved sorghum varieties in Tanzania ICRISAT, Nairobi, Kenya. Socioeconomics Discussion Paper Series, Series Paper Number 20

Gimode, D., Odeny, D.A., de Villiers, E.P., Wanyonyi, S., Dida, M.M., Mneney, E.E. Muchugi, A., Machuka, J., de Villiers, S.M. (2016). Identification of SNP and SSR Markers in Finger Millet Using Next Generation Sequencing Technologies. *PLoS ONE*, 11(7): e0159437. doi:10.1371/journal.pone.0159437

Grieder, C., Trachsel, S. and Hund, A. (2014). Early vertical distribution of roots and its association with drought tolerance in tropical maize. *Plant and Soil*, 377(1–2): 295–308

Jing, H., Bayon, C., Kanyuka, K., Berry, S., Wenzl, P., Huttner, E., Kilian, A. and Hammond-Kosack, K.E. (2009). DArT markers: diversity analyses, genomes comparison, mapping and integration with SSR markers in Triticum monococcum. *BMC Genomics*, 10:458 doi:10.1186/1471-2164-10-458

Gorthy, S., Narasu, L., Gaddameedi, A., Sharma, H.C., Kotla, A., Deshpande, S.P. and Are, A.K. (2017). Introgression of Shoot Fly (Atherigona soccata L. Moench) Resistance QTLs into Elite Post-rainy Season Sorghum Varieties Using Marker Assisted Backcrossing (MABC). *Frontiers in Plant Science*, 8(1494)

Govindaraj, M., Vetriventhan, M. and Srinivasa, M. (2015). Importance of Genetic Diversity Assessment in Crop Plants and Its Recent Advances: An Overview of Its Analytical Perspectives. *Genetics Research International*, Volume 2015, Article ID 431487, http://dx.doi.org/10.1155/2015/431487

Gupta, M., Chyi, Y.S., Romero-Severson, J. and Owen, J.L. (1994). Amplification of DNA markers from evolutionarily diverse genomes using single primers of simple-sequence repeats. *Theoretical and Applied Genetics*, 89: 998–1006

Hamidou, M., Souley, A.K.M., Kapran,I., Souleymane, O., Danquah, E.D., Ofori, K., Gracen, V. and Ba, M.N. (2018). Genetic Variability and Its Implications on Early Generation Sorghum Lines Selection for Yield, Yield Contributing Traits, and Resistance to Sorghum Midge. *International Journal of Agronomy*, 2018, Article ID 1864797, https://doi.org/10.1155/2018/1864797

Harris, H., Subudhi, P.K., Borrell, A., Jordan, D., Rosenow, D., Nguyen, H., Klein, P., Klein, R. and Mullet, J. (2007). Sorghum stay-green QTL individually reduce post-flowering drought-induced leaf senescence, *Journal of Experimental Botany*, 58(2): 327–338, https://doi.org/10.1093/jxb/erl225

Hasan, M.M., Rafii, M.Y., Ismail, M.R., Mahmood, M., Rahim, H.A., Alam, M.A. S. Ashkani, S., Malek, M.A. and Latif, M.A. (2015). Marker-assisted backcrossing: a

useful method for rice improvement. *Biotechnology and Biotechnological Equipment*, 29(2): 237–254. doi: 10.1080/13102818.2014.995920

Hash, C.T., Bhasker Raj, A.G., Lindup, S., Sharma, A., Beniwal, C.R., Folkertsma, R.T., Mahalakshmi, V., Zerbini, E. and Michael, B. (2003). Opportunities for marker-assisted selection (MAS) to improve the feed quality of crop residues in pearl millet and sorghum. *Field Crop Research*, 84: 79 – 88

Hayat, S., Hayat,Q., Alyemeni, M.N.A., Wani,A.S., Pichtel,J. and Ahmad, A. (2012). Role of proline under changing environments. *Plant Signaling and Behavoir*, 7(11): 1456–1466

Habyarimana, E., Laureti, D., Di Fonzo, N. and Lorenzoni, C. (2002). Biomass production and drought resistance at the seedling stage and in field conditions in sorghum. *Maydica*, 47: 303-309

Hasan, S.A., Rabei, S.H.., Nada, R.M. and Abogadallah, G.M. (2017). Water use efficiency in the drought-stressed sorghum and maize in relation to expression of aquaporin genes. *Biologia Plantarum*, 61(1): 127-137

Hayward, A.C, Tollenaere, R., Dalton-Morgan, J. and Batley, J. (2015). Molecular marker applications in plants. *Methods in Molecular Biology*, 1245: 13–27

Hodnett, G.L. and Rooney, W.L. (2018). Male sterility induction of sorghum using chemical hybridizing agent TFMSA, trifluoromethanesulfonamide. *Canadian Journal of Plant Science*, 98: 1102-1108

Hospital, F. (2001). Size of donor chromosome segments around introgressed loci and reduction of linkage drag in marker-assisted backcross programs. *Genetics*, 158(3):1363-1379

Hossain, A.B.S., Sears, A.G., Cox, T.S. and Paulsen, G.M. (1990). Desiccation tolerance and its relationship to assimilate partitioning in winter wheat. *Crop Science*, 30: 622-627

House, L.R. (1985). A guide to Sorghum Breeding. Second Edition. International Crops Research Institute for the Semi-Arid Tropics ICRISAT Patancheru P.O. Andhra Pradesh 502 324, India

Idris, A.E., Hamza, N.B., Yagoub, S.O., Ibrahim, A.I.A. and El-Amin, H.K.A. (2012). Maize (*Zea mays* L.) Genotypes Diversity Study by Utilization of Inter-Simple Sequence Repeat (ISSR) Markers. *Australian Journal of Basic and Applied Sciences*, 6(10): 42-47

Iqbal, M.A. and Iqbal, A. (2015). Overview on Sorghum for Food, Feed, Forage and Fodder: Opportunities and Problems in Pakistan's Perspectives. *American-Eurasian Journal of Agriculture and Environmental Sciences*, 15(9): 1818-1826

Izanloo, A., Condon, A. G., Langridge, P., Tester, M. and Schnurbusch, T. (2008). Different mechanisms of adaptation to cyclic water stress in two South Australian bread wheat cultivars. *Journal of Experimental Botany*, 59(12): 3327–3346

Jabereldar, A.A., El Naim, A.M., Dagash, Y.M., Abdalla, A.A. and Ahmed, S.E. (2017) Effect of water stress on drought tolerance index of sorghum (Sorghum bicolor L. Moench) in North Kordofan state. *University of Kordofan Journal of Natural Resources and Environmental Studies*, 10: 11–20

Jenhan, T. and Lakhanpaul, S. (2006). Single Nucleotide Polymorphism (SNP)-Methods and application in plant genetics: A review. *Indian Journal of Biotechnology*, 5: 435-459

Jiang, G. (2013). Molecular Markers and Marker-Assisted Breeding in Plants. Plant Breeding from Laboratories to Fields, Andersen, S.B. IntechOpen, DOI: 10.5772/52583. Available from: https://www.intechopen.com/books/plant-breeding-from-laboratories-to-fields/molecular-markers-and-marker-assisted-breeding-in-plants

Jordi, W., Schapendonk, A., Davelaar, E., Stoopen, G. M., Pot, C.S., De Visser, R. and Amasino, R.M. (2000). Increased cytokinin levels in transgenic PSAG12–IPT tobacco plants have large direct and indirect effects on leaf senescence, photosynthesis and N partitioning. *Plant, Cell & Environment*, 23(3): 279–289

Kadioglu, A. and Terzi, R. (2007). A dehydration avoidance mechanism**:** Leaf rolling. *Botanical Review*, 73: 290-302

Kalia, R.K., Rai, M.K., Kalia, S., Singh, R., Dhawan, A.K. (2011). Microsatellite markers: an overview of the recent progress in plants. *Euphytica*, 177: 309–334

Kaliba, A.R, Mazvimavi, K., Gregory, T.L., Mgonja, F.M. and Mgonja, M. (2018). Factors affecting adoption of improved sorghum varieties in Tanzania under information and capital constraint. *Agricultural and Food Economics*, 6:18

Kamal, N.M., Gorafi, Y.S.A. and Ghanim, A.M.A. (2017). Performance of Sorghum stay-green introgression lines under post-flowering drought**.** *International Journal of Plant Research*, 7(3): 65-74

Kamal, S.M., Gorafi, Y.S.A., Tsujimoto, H. and Ghanim, A.M.A. (2018). Stay-Green QTLs response in adaptation to post flowering drought depends on the drought deverity.

BioMed Research International, Article ID 7082095:15. https://doi.org/10.1155/2018/7082095

Kapanigowda, M.H., Payne, W.A., Rooney, W.L., Mullet, J.E. and Balota, M. (2014). Quantitative trait locus mapping of the transpiration ratio related to pre flowering drought tolerance in sorghum (*Sorghum bicolor*). *Functional Plant Biology*, 41(11):1049-1065. doi: 10.1071/FP13363

Kapanigowda, M.H., Perumal, R., Djanaguiraman, M., Aiken, R.M., Tesso, T., Vara Prasad, P.V. and Little, C.R. (2013). Genotypic variation in sorghum [*Sorghum bicolor* (L.) Moench] exotic germplasm collections for drought and disease tolerance. *Springer Plus*, 2:650

Kassahun, B., Bidinger, F.R., Hash, C.T. and Kuruvinashetti, M.S. (2010). Stay-green expression in early generation sorghum [*Sorghum bicolor* (L.) Moench] QTL introgression lines. *Euphytica*, 172(3): 351–362. https://doi.org/10.1007/s10681-009-0108-0

Kebede, H., Subudhi, P.K., Rosenow, D.T. and Nguyen, H.T. (2001).Quantitative trait loci influencing drought tolerance in grain sorghum (Sorghum bicolor L. Moench). *Theoretical and Applied Genetics*, 103(2–3): 266–276 https://doi.org/10.1007/s001220100541

Khan, I.A., Habib, S., Sadaqat, H.A. and Tahir, M.H.N. (2004). Selection criteria based on seedling growth parameters in maize varies under normal and water stress conditions. *International Journal of Agriculture and Biology*, 6: 252–156

Kharrazi, M.A.S. and Rad, MRN. (2011). Evaluation of sorghum genotypes under drought stress conditions using some stress tolerance indices. *African Journal of Biotechnology*, 10(61): 13086-13089

Khatab, I.A., El-Mouhamady, A.A., Abdel-Rahman, H.M., Farid, M.A. and El-Demardash, I.S. (2017). Agro-morphological and molecular characterization of sorghum (Sorghum vulgare L.) for water stress tolerance. *International Journal of Current Research in Biosciences and Plant Biology*, 4:37–55

Kothari, C.R. (2004). Research Methodology: Methods and Techniques, New Age International (P) Ltd, Publishers, New Delhi

Kudadjie, C.Y., 'Struik, P.C., Richards, P. and Offei, S.K. (2004). Assessing production constraints, management and use of sorghum diversity in north-east Ghana: a diagnostic study. *Wageningen Journal of Life Sciences*, 52(3-4): 371-391

Kimani, W., Ngugi, K., Kiambi, D., MutituI, E.W., De Villiers, S. (2012). Marker-assisted introgression of stay-green trait in a Kenyan sorghum variety. *East African Agricultural and Forestry Journal*, 78(2): 109-115

Kiranmayee, K.N.S.U., Hash, C.T., Sivasubramani, S., Ramu, P., Amindala, B.P., Rathore, A., Kishor, P.B.K., Gupta, R. and Deshpande, S.P. (2020). Fine-Mapping of Sorghum Stay-Green QTL on Chromosome10 Revealed Genes Associated with Delayed Senescence. *Genes*, 11: 1026; doi:10.3390/genes11091026

Kusaka, M., Ohta, M. and Fujimura, T. (2005). Contribution of inorganic components to osmotic adjustment and leaf folding for drought tolerance in pearl millet. *Physiologia Plantarum*, 125: 474–489

Laxman, S.J. (1997). Plastic bag emasculation technique in sorghum (*Sorghum bicolor* L. (Moench)). *Journal of Research*, 25(2): 38-39

Li, Y., Song, H., Zhou, L., Xu, Z. and Zhou, G. (2019). Tracking chlorophyll fluorescence as an indicator of drought and rewatering across the entire leaf lifespan in a maize field. *Agricultural water management*, 2011: 190 – 201

Luche, H.D., da Silva, J.G. da Maia, L.C., de Oliveira, A.C. (2015). Stay-green: a potentiality in plant breeding. *Ciência Rural*, 45(10): 1755–1760

Lyimo, J.G. and Kangalawe, R.Y.M. (2010) Vulnerability and Adaptative Strategies to the Impact of Climate Change and Variability: The Case of Rural Households in Semi-Arid Tanzania. *Environmental Economics*, 1: 89-97

Lynch, J.P. (2013). Steep, cheap and deep: an ideotype to optimize water and N acquisition by maize root systems. *Annals of Botany*, 112(2): 347–357

Mace, E.S. and Jordan, D.R. (2011). Integrating sorghum whole genome sequence information with a compendium of sorghum QTL studies reveals non-random distribution of QTL and of gene rich regions with significant implications for crop improvement. *Theoretical and Applied Genetics*, doi:10.1007/s00122-011-1575-y

Mace, E.S., Singh, V., Van Oosterom, E.J., Hammer, G.L., Hunt, C.H. and Jordan, D.R. (2012). QTL for nodal root angle in sorghum (*Sorghum bicolor* [L]. Moench) co-locate with QTL for traits associated with drought adaptation TAG. *Theoretical and Applied Genetics*, 124(1): 97–109. https://doi.org/10.1007/s00122-011-1690-9

Mace, E.S., Xia, L., Jordan, D.L., Halloran, K., Parh, D.K., Huttner, E., Wenzl, P. and Kilian, A. (2008). DArT markers: diversity analysis and mapping in Sorghum bicolor. *BMC Genomics*, 9:26

Mafuru J.M., Norman D.W., and Fox J.S. (2007). Consumer Perception of Sorghum Variety Attributes in the Lake Zone Tanzania. *AAAE Conference Proceedings,* pp. 171-176

Mammadov, J., Aggarwal, R., Buyyarapu, R. and Kumpatla, S. (2012). SNP markers and their impact on plant breeding. *International Journal of Plant Genomics,* 2012 (ID 728398). doi:10.1155/2012/728398

Mamoudou, H.D., Hurry, G., Alfred, S., Alphons, G.J. and Van, B. (2006). Sorghum grain as human food in Africa: relevance of content of starch and amylase activities. *African Journal of Biotechnology*, 5(5): 384-395

Maqsood, M. and Ali, S.N.A. (2007). Effects of drought on growth, development, radiation use efficiency and yield of finger millet (Eleucine coracana). *Pakstan Journal of Botany*, 39(1): 123-134

Marenya, P.P., Barrett, C.B. (2009). Soil quality and fertilizer use rates among smallholder farmers in western Kenya. *Agricultural Ecomics*, 40(5): 561-572

Mavhura, E. Manatsa, D. and Mushore, T. (2015). Adaptation to drought in arid and semi-arid environments: Case of the Zambezi Valley, Zimbabwe. *Jamba*, 7(1): 144

Meyer, W., Mitchell, T.G., Freedman, E.Z. and Vilgays, R. (1993). Hybridization probes for conventional DNA fingerprinting used as single primers in the polymerase chain reaction to distinguish strains of *Cryptococcus neoformes*. *Journal of Clinical Microbiology*, 31: 2274–2280

Mc Guire S.J. (2008). Path-dependency in plant breeding: Challenges facing participatory breeding reforms in Ethiopian Sorghum Improvement Program. *Agricultural Systems* 96(1-3): 139-149. Doi: 10.1016/j. agsy. 2007.07.003

Meade, F., Byrne, S., Griffin, D., Kennedy, C., Mesiti, F. and Dan Milbourne, D. (2019). Rapid Development of KASP Markers for Disease Resistance Genes Using Pooled Whole-Genome Re-sequencing. *Potato Research*, https://doi.org/10.1007/s11540-019-09428-x

Meru, G.M. (2010). Genotyping BC_3F_2 populations of four Ethiopian sorghum varieties for Stay Green QTLs introgression through Marker assisted selection with SSRs. MSc. Thesis, Kenyata University, Nairobi- Kenya

Mgonja, M.A., Chandra, S., Gwata, E.T., Obilana, A.B., Monyo, E.S., Rohrbach, D.D., Chisi, M., Kudita, S. Saadan, H.M. (2005). Improving the efficiencies of national crop breeding programs through region-based approaches: the case of sorghum and pearl millet in southern Africa. *Journal of Food, Agriculture and Environment*, 3:124–129

Midega, C.A.O., Murage, A.W., Pittchar, J.O. and Khan, Z.R. (2016). Managing storage pests of maize: Farmers' knowledge, perceptions and practices in western Kenya. *Crop Protection*, 90: 142-149

Misra, A.N., Biswal, A.K. and Misra, M. (2002). Physiological, biochemical and molecular aspects of water stress responses in plants and the biotechnological applications. *Proceedings of the National Acadey of Sciences-India*, 72B: 115–134

Mkonda, M.Y. and He, X. (2017). Foodf Security and Policy in Tanzania's Semi-Arid Agro-Ecological Zone. *Sustainability*, 9: 1490; doi: 10.3390/su9081490

Mofokeng, M.A., Shimelis,H., Tongoona, P. and Laing, M.D. (2016). Constraints and varietal trait preferences of sorghum producers in South Africa. *Journal of Tropical Agriculture*, 54(1): 7-15

Monclus, R., Dreyer, E., Villar, M., Delmotte, F.M., Delay, D., Petit, J.M., Barbaroux C., Thiec, D.L., Bréchet, C. and Brignolas, F. (2006). Impactof drought on productivity and water use efficiency in 29 genotypes of Populus deltoids × Populus nigra. *New Phytologist*, 169: 765–777

Mongi, H., Majule, A.E. and Lyimo, J.G. (2010) Vulnerability and Adaptation of Rain Fed Agriculture to Climate Change and Variability in Semi-Arid Tanzania. *African Journal of Environmental Sciences and Technology*, 4: 371-381. https://doi.org/10.5897/AJEST09.207

Monyo, E.S., Mgonja, M.A. Ngereza, J. and Rohrbach, D.D. (2002). "Adoption of Improved Sorghum and Pearl Millet Varieties in Tanzania." *International Sorghum and Millets Newsletter*, 43: 12-14

Monyo, E.S., Mgonja, M.A., Rohrbach, D.D., Ngereza, J., Saadan, H.M and Ngowi, P. (2004). Adoption of improved sorghum and pearl millet technology in Tanzania. Bulawayo, Zimbabwe: *International Crops Research Institute for the Semi-Arid Tropics*, pp. 28

Morris, M., Dreher, K., Ribaut, J.M. and Khairallah, M. (2003). Money matters (II): costs of maize inbred line conversion schemes at CIMMYT using conventional and marker-assisted selection. *Molecular Breeding*, 11: 235–247

Morka, E.A. (2015). Physiological Indices for Drought Tolerance in Stay-green Sorghum (*Sorghum bicolor* L. Moench) Accessions. MSc. Thesis, Addis Ababa University

Mrema, E., Shimelis, H., Laing, M. and Bucheyeki, T. (2017). Farmers' perceptions of sorghum production constraints and *Striga* control practices in semi-arid areas of Tanzania. *International Journal of Pest Management*, 63(2): 146-156, DOI: 10.1080/09670874.2016.1238115

Mrema, E., Shimelis, H., Laing, M. and Bucheyeki, T. (2017). Screening of sorghum genotypes for resistance to *Striga hermonthica* and *S. asiatica* and compatibility with *Fusarium oxysporum* f.sp. *strigae*. *Acta Agriculturae Scandinavica, Section B — Soil & Plant Science*, 67(5): 395-404

Msongaleli, B., Tumbo, S., Rwehumbiza, F. and Kihupi, N. (2015). Determinants of farm-level decisions regarding cereal crops and varieties in semi-arid central Tanzania. *African Journal of Agricultural Research*, 10(30): 2968-2978

Murray, S.C., Rooney, W.L., Hamblin, M.T., Mitchell, S.E. and Kresovich, S. (2009). Sweet Sorghum Genetic Diversity and Association Mapping for Brix and Height. *The Plant Genome*, 2(1), 48–62. https://doi.org/10.3835/plantgenome2008.10.0011

Muui, C.W., Muasya, R.M. and Kirubi, D.T. (2013). Participatory identification and evaluation of sorghum (*Sorghum bicolor* (L.) Moench) landraces from lower eastern Kenya. *International Research Journal of Agricultural Science and Soil Science*, 3(8): 283–290

Mwamahonje, A. and Maseta, Z. (2018). Evaluation of yield performance of sorghum [*Sorghum bicolor* (L) Moench] varieties in Central Tanzania. *International Journal of Agronomy and Agricultural Research*, 13(2): 8-14

Mwamahonje, A., Eleblu, J.S.Y., Ofori, K., Deshpande, S., Feyissa, T. and Bakuza, W.E. (2021). Sorghum Production Constraints, Trait Preferences, and Strategies to Combat Drought in Tanzania. *Sustainability*, 13, 12942. https://doi.org/10.3390/su132312942

Mwanga J.N.W. (2002). Adoption of Improved Technologies for Sorghum and Pearl millet Production in Dodoma Region in Central Tanzania. Unpublished MSc. Dissertation, Sokoine University of Agriculture

Naoura, G., Sawadogo, N., Atchozou, E.A., Emendack, Y., Hassan, M.A., Reoungal, D., Amos, D.N., Djirabaye, N., Tabo, R. and Laza, H. (2019). Assessment of agro-morphological variability of dry-season sorghum cultivars in Chad as novel sources of drought tolerance. *Scientific Reports*, 9:19581. https://doi.org/10.1038/s41598-019-56192-6

National Research Council. (1996). Lost Crops of Africa: Volume I: Grains. Washington, DC: *The National Academies Press*. https://doi.org/10.17226/2305

Negarestani, M., Tohidi-Nejad, E., Khajoei-Nejad, G., Nakhoda, B. Mohammadi-Nejad, G. (2019). Comparison of Different Multivariate Statistical Methods for Screening the Drought Tolerant Genotypes of Pearl Millet (Pennisetum americanum L.) and Sorghum (Sorghum bicolor L.). *Agronomy*, 9: 645; doi: 10.3390/agronomy9100645

Nelimor, C., Badu-Apraku, B., Garcia-Oliveira, A.L., Tetteh, A., Paterne, A., N'guetta, A.S. and Gedil, M. (2020). Genomic Analysis of Selected Maize Landraces from Sahel and

Coastal West Africa Reveals Their Variability and Potential for Genetic Enhancement. *Genes*, 11:1054; doi: 10.3390/genes11091054

Ng, W.L. and Tan, S.G. (2015). Inter-Simple Sequence Repeat (ISSR) Markers: Are We Doing It Right? *ASM Science Journal*, 9(1): 30–39

Ngugi, K., Kimani, W., Kiambi, D., Mutitu, E.W. (2013). Improving Drought Tolerance in Sorghum bicolor L. Moench: Mar ker-Assisted transfer of the Stay-Green Quantitative Trait Loci (Q T L) from a Characterized Donor Source into a Local Farmer Variety. *International Journal of Scientific Research in Knowledge*, 1(6): 154-162. http://dx.doi.org/10.12983/ijsrk-2013-p154-162

Nhamo, L., Matchaya, G., Mabhaudhi, T., Nhlengethwa, S., Nhemachena, C. and Mpandeli, S. (2019). Cereal Production Trends under Climate Change: Impacts and Adaptation Strategies in Southern Africa. *Agriculture*, 9(30); doi:10.3390/agriculture902003

Nuijten, H., Temudo, M., Richards, P., Okry, F., Teeken, B., Mokuwa, G. and Struik, P. (2013).Towards a new approach for understanding interactions of technology with environment and society in small-scale rice farming. In Realizing Africa's Rice Promise; CABI: Wallingford, UK, pp. 355–36

Ochieng, L.A., Mathenge, P.W. and Muasya, R. (2011). A survey of on-farm seed production practices of sorghum (*Sorghum bicolor* L. Moench) in bomet district of Kenya. *African Journal of Food, Agriculture, Nutrition and Development*, 11(5): 5233-5253

Ogbaga, C.C., Bajhaiya, A.K. and Gupta S.K., (2019). Improvements in biomass production: Learning lessons from the bioenergy plants maize and sorghum. *Journal of Environmental Biology*, 40: 400-406

Ogbaga, C.C., Stepien, P., Dyson, B.C., Rattray, N.J.W., Ellis, D.I., Goodacre, R. and Johnson, G.N. (2016). Biochemical Analyses of Sorghum Varieties Reveal Differential Responses to Drought. *PLoS ONE*, 11(5): e0154423. https://doi.org/10.1371/journal.pone.0154423

Ogbaga, C.C., Stepien, P. and Johnson, G.N. (2014). Sorghum (Sorghum bicolor) varieties adopt strongly contrasting strategies in response to drought. *Physiologia Plantarum*, 152: 389–401. doi: 10.1111/ppl.12196 PMID:24666264

Ogeto, R.M., Cheruiyot, E., Mshenga, P. and Onyari, C.N. (2013). Sorghum production for food security: A socio-economic analysis of sorghum production in Nakuru County, Kenya. *African Journal of Agricultural Research*, 8(47): 6055-6067

Olsen, K.M. (2012). One Gene's Shattering Effects. *Nature Genetics*, 44: 616–617

Orr, A., Mwema, C., Gierend, A. and Nedumaran, S. (2016). Sorghum and Millets in Eastern and Southern Africa. Facts, Trends and Outlook. Working Paper Series No. 62. ICRISAT Research Program, Markets, Institutions and Policies. Patancheru 502 324, Telangana, India: International Crops Research Institute for the Semi-Arid Tropics. pp 76

Ouedraogo, N., Sanou, J., Kam, H., Traore, H., Adam, M., Gracen, V. and Danquah, E.Y. (2017). Farmers' perception on impact of drought and their preference for sorghum cultivars in Burkina Faso. *Agricultural Science Research Journal*, 7(9): 277 – 284

Pang, J., Turner, N.C., Du, Y., Colmer, T.D. and Siddique, K.H.M. (2017). Pattern of Water Use and Seed Yield under Terminal Drought in Chickpea Genotypes. *Frontiers in Plant Science*, 8:1375. https://doi.org/10.3389/fpls.2017.01375

Paterson, A.H., Bowers, J.E., Bruggmann, R., Dubchak, I., Grimwood, J., Gundlach, H. and Rokhsar, D.S. (2009). The *Sorghum bicolor* genome and the diversification of grasses. *Nature*, 457(7229): 551–556. https://doi.org/10.1038/nature07723

Paul, S., Wachira, F.N., Powell, W. and Waugh, R. (1997). Diversity and genetic differentiation among populations of Indian and Kenyan tea (*Camellia sinensis* (L.) O. Kuntze) revealed by AFLP markers. *Theoretical and Applied Genetics*, 94: 255-263

Peleg, Z., Reguera, M., Tumimbang, E., et al., (2011). Cytokinin-mediated source/sink modifications improve drought tolerance and increase grain yield in rice under water-stress. *Plant Biotechnol Journal*, 9(7): 747–58. 10.1111/j.1467-7652.2010.00584.x

Phuke, R.M., Anuradha, K., Radhika, K., Jabeen, F., Anuradha, G., Ramesh, T., Hariprasanna, K., Mehtre, S.P., Deshpande, S.P., Anil, G., Das, R.R., Rathore, A., Hash, T., Reddy, B.V.S. and Kumar, A.A. (2017). Genetic Variability, Genotype × Environment Interaction, Correlation, and GGE Biplot Analysis for Grain Iron and Zinc Concentration and Other Agronomic Traits in RIL Population of sorghum (Sorghum bicolor L. Moench). *Frontiers in Plant Science*, 8: 712 doi: 10.3389/fpls.2017.00712

Phuong, N., Afolayan, G., Stu¨tzel, H., Uptmoor, R. and El-Soda, M. (2019). Unraveling the genetic complexity underlying sorghum response to water availability. *PLoS ONE*, 14(4): e0215515. https://doi.org/10.1371/journal.pone.0215515

Platten, J.D., Cobb, J.N. and Zantua, R.E. (2019). Criteria for evaluating molecular markers: Comprehensive quality metrics to improve marker-assisted selection. *PLoS ONE*, 14(1):e0210529.https://doi.org/10.1371/journal.pone.0210529

Premachandra, G.S., Hahn, D.T. and Joly, R.J. (1994). "Leaf water relations and gas exchange in two grain sorghum genotypes differing in their pre- and post-flowering drought tolerance," *Journal of Plant Physiology*, 143(1): 96–101

Prohens, J. (2011). Plant breeding: a success story to be continued thanks to the advances in genomics. *Frontiers in Plant Science*, 2(51): 1-3.https://doi.org/10.3389/fpls.2011.00051

Rajkumar, B. F., Kavil, S.P., Girma, Y., Arun, S.S., Dadakhalandar, D., Gurusiddesh, B.H., Patil, A.M., Thudi, M., Bhairappanavar, S., Narayana, Y., Krishnaraj, P.U., Khadi, B.M. and Kamatar, M.Y. (2013). Molecular mapping of genomic regions harbouring QTLs for root and yield traits in sorghum (*Sorghum bicolor* L. Moench). *Physiology and Molecular Biology of Plants*, 19: 409–419. Doi: 10.1007/s12298-013-0188-0

Reddy, R.N.., Madhusudhana, R., Mohan, S.M., Chakravarthi, D.V., Mehtre, S.P., Seetharama, N. and Patil, J.V. (2013). [*Sorghum bicolor* (L.) Moench] Mapping QTL for grain yield and other agronomic traits in post-rainy sorghum. *Theoretical and Applied Genetics*, 126: 1921–1939. doi:10.1007/s00122-013-2107-8

Reddy, N.R., Ragimasalawada, M., Sabbavarapu, M.M., Nadoor, S. and Patil, J.V. (2014). Detection and va Perrierlidation of stay-green QTL in post-rainy sorghum involving widely adapted cultivar, M35-1 and a popular stay-green genotype B35. *BMC Genomics*, 15: 909. https://doi.org/10.1186/1471-2164-15-909

Reddy, M.P. Sarla, N. and Siddiq, E.A. (2002). Inter simple sequence repeat (ISSR) polymorphism and its application in plant breeding. *Euphytica*, **128:** 9–17

Ribaut, J.M. and Betran, J. (1999). Single large-scale marker assisted selection (SLS-MAS). *Molecular Breeding*, 5(6): 531-541

Ringo, J., Beatrice, W., Mary, M., Deshpande, S., Rathore, A., Mneney, E. and Gudu, S. (2015). Combining ability of some sorghum lines for dry lands and sub-humid environments of East Africa. *African Journal of Agricultural Research*, 10(19): 2048–2060

Rivero, R.M., Kojima, M., Gepstein, A., Sakakibara, H., Mittler, R., Gepstein, S. and Blumwald, E. (2007). Delayed leaf senescence induces extreme drought tolerance in a flowering plant. *Proceedings of the National Academy of Sciences of the United States of America*, 104(49): 19631–19636

Robertson, M.J., Fukai, S., Ludlow, M.M. and Hammer, G.L. (1993). Water extraction by grain sorghum in a sub-humid environment. II. Extraction in relation to root growth. *Field Crops Research*, 33(1-2): 99-112

Roling, N.G., Hounkonnou, D., Offei, S.K., Tossou, R. and Van Huis, A. 2004. Linking science and farmers' innovative capacity: diagnostic studies from Ghana and Benin. *NJAS - Wageningen Journal of Life Sciences* 52: 211-235

Rosenow, D.T and Clark, L.E. (1981). Drought tolerance in sorghum. In: Loden HD, Wilkinson D, eds. *Proceedings of the 36th annual corn and sorghum industry research conference*, pp. 18–31

Routley, R., Broad, I., McLean, G., Whish, J. and Hammer, G. (2003). The effect of row configuration on yield reliability in grain sorghum: I. Yield, water use efficiency and

soil water extraction. *Proceeding of the Eleventh Australian Agronomy Conference*, Geelong, 2-6 February, 2003. Geelong, Victoria: Australian Society of Agronomy

Sabadin, P.K., Malosetti, M., Boer, M.P., Tardin, F.D., Santos, F.G., Guimara˜es, C.T., Gomide, R.L., Andrade, C.L.T., Albuquerque, P.E.P., Caniato, F.F., Mollinari, M., Margarido, G.R.A., Oliveira, B.F., Schaffert, R.E., Garcia, A.A.F., van Eeuwijk, F.A. and Magalhaes, J.V. (2012). Studying the genetic basis of drought tolerance in sorghum by managed stress trials and adjustments for phonological and plant height differences. *Theoretical and Applied Genetics*, DOI 10.1007/s00122-012-1795-9

Saddam, S., Bibi, A., Sadaqat, H.A and Usman, B.F. (2014). Comparison of 10 sorghum (*Sorghum bicolor* L) genotypes under various water stress regimes. *The Journal of Animal & Plant Sciences*, 24(6): 1811-1820

Sadia, B., Awan, F.S., Saleem, F., Sadaqat,H.A., Arshad, F.S. and Shaukat, H. (2018).*Genetic Improvement of Sorghum for Biomass Traits Using Genomics Approaches*, pp. 23-37. http://dx.doi.org/10.5772/intechopen.73010

Sakiyama, N.S., Ramos, H.C.C., Caixeta, E.T. and Pereira, M.G. (2014). Plant breeding with marker-assisted selection in Brazil. *Crop Breeding and Applied Biotechnology*, 14(1): 54–60

Sanchez, A.C., Subudhi, P.K., Rosenow, D.T., and Nguyen, H.T. (2002). Mapping QTLs associated with drought resistance in sorghum (*Sorghum bicolor* L. Moench). *Plant Molecular Biology*, 48(5–6): 713–726

Sankar, A.A. and Moore, G.A. (2001). Evaluation of inter-simple sequence repeat analysis for mapping in *Citrus* and extension of genetic linkage map. *Theoretical and Applied Genetics*, 102: 206–214

Satish, l., Shilpha, J., Pandian, S., Rency, A.S., Rathinapriya, P., Ceasar, S.A., Largia, M.J.V., Kumar, A.A. and Ramesh, M. (2016). Analysis of genetic variation in sorghum (*Sorghum bicolor* (L.) Moench) genotypes with various agronomical traits using SPAR methods. *Genes*, 576:581-585

Saunders, M., Lewis, P. and Thornhill, A. (2007). Research Methods for Business Students. 4th Edition, Financial Times Prentice Hall, Edinburgh Gate, Harlow

Saxena, N.P., Toole,O. and John, C. (eds.). (2002). Field Screening for Drought Tolerance in Crop Plants with Emphasis on Rice: *Proceedings of an International Workshop on Field Screening for Drought Tolerance in Rice*, 11–14 Dec 2000, ICRISAT, Patancheru, India

Schechambo, F.C., Sosovele, H. and Kisanga, D. (1999). 'RethinkingNatural Resource Degradation inSemi-Arid Sub-Saharan Africa: The Case of Semi-Arid Tanzania'. Report for ODI. http://www.tanzaniagateway.org/docs/tzlit.pdf

Schymanski, S.J. and Or, D. (2016). Wind increases leaf water use efficiency. *Plant, Cell and Environment*, 39: 1448–1459

Semagn, K., Babu, R., Hearne, S. and Olsen, M. (2013). Single nucleotide polymorphism genotyping using Kompetitive Allele Specific PCR (KASP): overview of the technology and its application in crop improvement. *Molecular Breeding*, 33: 1–14

Serraj, R., Hash, T.C., Buhariwalla, H.K., Bidinger, F.R., Folkertsma, R.T., Chandra, S., Gaur, P., Kashiwagi, J., Nigam, S.N., Rupakula, A. and Crouch, J.H. (2003). Marker-assisted breeding for crop drought tolerance at ICRISAT: Achievements and prospects. Tuberosa R., Phillips R.L., Gale M. (eds.), *Proceedings of the International Congress "In the Wake of the Double Helix: From the Green Revolution to the Gene Revolution*", 27-31 May 2003, Bologna, Italy

Shehzad, T. and Okuno, K. (2015). QTL mapping for yield and yield-contributing traits in sorghum (*Sorghum bicolor* L. Moench) with genome-based SSR markers. *Euphytica*, 203: 17–31. doi:10.1007/s10681-014-1243-9

Sher, A., Barbanti, L. and Ansar, M. (2013). Growth response and plant water status in forage sorghum [Sorghum bicolor (L.) Moench] cultivars subjected to decreasing levels of soil moisture. *Australian Journal of Crop Science*, 7(6): 801-808

Siebers, M.H., Yendrek, C.R., Drag, D., Locke, A.M., Acosta, L.R., Leakey, A.D.B. and Ort, D.R. (2015). Heat waves imposed during early pod development in soybean (Glycine max) cause significant yield loss despite a rapid recovery from oxidative stress. *Global Change Biology*, 21(8): 3114–3125

Simova-Stoilova, L., Vassileva, V. and Feller, U. (2016). Selection and Breeding of Suitable Crop Genotypes for Drought and Heat Periods in a Changing Climate: Which Morphological and Physiological Properties Should Be Considered? *Agriculture*, 6(2): 26. https://doi.org/10.3390/agriculture6020026

Simtowe, F. and Mausch, K. (2018). "Who is quitting? An analysis of the dis-adoption of climate smart sorghum varieties in Tanzania", *International Journal of Climate Change Strategies and Management*, https://doi.org/10.1108/IJCCSM-01-2018-0007

Sinha, S. and Kumaravadivel, N. (2016).Understanding Genetic Diversity of Sorghum Using Quantitative Traits. *Scientifica*, 2016: 3075023. http://dx.doi.org/10.1155/2016/3075023

Slegers, M.F.W. (2008). Exploring farmers' perceptions of drought in Tanzania and Ethiopia. Doctoral Thesis, Wageningen University

Sory, S. (2015). Marker assisted selection for post-flowering drought tolerance in *Sorghum bicolor* [L.] Moench. PhD Thesis, University of Ghana

Su, X., Wei, F., Huo, Y., Xia, Z.J. and Fi, P.S. (2017). Comparative physiological and molecular analyses of two contrasting flue-cured tobacco genotypes under progressive drought stress. *Frontiers in Plant Science*, 8: 827

Sukumaran, S., Li, X., Xianran Li, X., Zhu, C., Bai, G., Perumal, R., Tuinstra, M.R., P.V. Prasad, P.V.V., Mitchell, S.E., Tesso, T.T. and Yu, J. (2016). QTL Mapping for Grain Yield, Flowering Time, and Stay-Green Traits in Sorghum with Genotyping-by-Sequencing Markers. *Crop Science*, 56: 1429–1442. doi: 10.2135/cropsci2015.02.0097

Suleiman, M. and Rugumamu, C.P. (2017). Management of insect pests of stored sorghum using botanicals in Nigerian traditional stores. *Journal of Stored Products and Postharvest Research*, 8(9): 93-102. Doi: 10.5897/JSPPR2017.0247

Swai, O.W., Mbwambo, J.S. and Magayane, F.T. (2012) Gender and Adaptation Practices to the Effects of Climate Change in Bahi and Kondoa Districts, Dodoma Region, Tanzania. *Journal of Sustainable Development*, 5: 65-77. https://doi.org/10.5539/jsd.v5n12p65

Tadesse, T., Tesso, T. and Ejeta, G. (2008). Combining ability of introduced sorghum parental lines for major morpho-agronomic traits. Journal of SAT Agricultural Research, Volume 6. An open access journal published by ICRISAT

Takeda, S. and Matsuoka, M. (2008). Genetic approaches to crop improvement: responding to environmental and population changes. *Nature*, 9: 444-457

Tao, Y.Z., Henzell, R.G., Jordan, D.R., Butler, D.G., Kelly, A.M. and McIntyre, C.L. (2000). Identification of genomic regions associated with stay green in sorghum by testing RILs in multiple environments. *Theoretical and Applied Genetics*, 100(8): 1225–1232. https://doi.org/10.1007/s001220051428

Tari, I., Laskay, G., Takacs, Z. and Poor, P. (2012). Responses of Sorghum to Abiotic Stresses: A Review. *The Journal of Agronomy and Crop Science*, 199(4): 264-274. doi:10.1111/jac.12017

Tenywa, M.M., Nyamwaro, S.O., Kalibwani, R., Mogabo, J., Buruchara, R. and Fatunbi, A.O. (2018). Innovation Opportunities in Sorghum Production in Uganda. *FARA Research Reports*, 2(18): 20

Tesfamichae, A., Githiri, S.M., Kasili, R., Woldeamlak, A. and Nyende, A.B. (2015). Genetic Variation among Sorghum (*Sorghum bicolor* L. Moench) Landraces from Eritrea under Post-Flowering Drought Stress Conditions. *American Journal of Plant Sciences*, 6: 1410-142

Teshome, A., Baum, B. R., Fahrig, L., Torrance, J.K., Arnason, T.J. and Lambert, J.D. (1997). Sorghum [*Sorghum bicolor* (L.) Moench] landrace variation and classification

in North Shewa and South Welo, Ethiopia. *Euphytica*, 97(3): 255–263 https://doi.org/10.1023/A:1003074008785

Thomas, H. and Howarth, C.J. (2000). Five ways to stay-green. *Journal of Experimental Botany*, 51: 329–337

Thomas, H. and Ougham, H. (2014). The stay-green trait. *Journal of Experimental Botany*, 65(14): 3889–3900

Thomson, M.J. (2014). High-throughput SNP genotyping to accelerate crop improvement. *Plant Breeding and Biotechnology*, 2: 195–212 https://doi.org/10.9787/PBB.2014.2.3.195

Timu, A.G., Mulwa, R., Okello, J. and Kamau, M. (2014). The role of varietal attributes on adoption of improved seed varieties: the case of sorghum in Kenya. *Agriculture & Food Security*, 3: 9. https://doi.org/10.1186/2048-7010-3-9

Tuinstra, M.R., Grote, E.M., Goldsbrough, P.B. and Ejeta, G. (1997). Genetic analysis of post-flowering drought tolerance and components of grain development in *Sorghum bicolor* (L.) Moench. *Molecular Breeding*, 3(6): 439–448. https://doi.org/10.1023/A:1009673126345

Tuinstra, M.R., Grote, E.M., Goldsborough, P.B. and Ejeta, G. (1996). Identification of quantitative trait loci associated with pre-flowering drought tolerance in sorghum. *Crop Science*, 36: 1337-1344

United States Department of Agriculture (USDA). (2019). World Agricultural Production; Circular Series WAP 3–19; USDA: Washington, DC, USA

Uphadyaya, H.D., Pundir, R.P.S., Dwivedi, S.L., Gowda, C.L.L., Reddy, V.G. and Singh, S. (2009).“Developing a Mini- core Collection of Sorghum for Diversified Utilization of Germplasm,” *Crop Science*, 49(5): 1769-1780

United Repubic of Tanzania (URT). (2007). ‘National AdaptationProgramme of Action (NAPA)’. Dar es Salaam: Vice President’s Office, Division of Environment

United Republic of Tanzania (URT). (1997). Singida Region Socio-Economic Profile. Regional Commissioner Singida

Vadez, V., Deshpande, S., Kholova, J., Ramu, P. and Hash, C.T. (2013). Molecular Breeding for Stay-Green: *Progress and Challenges in Sorghum*. In R. K. Varshney & R. Tuberosa (Eds.), Translational Genomics for Crop Breeding (pp. 125–141). John Wiley & Sons Ltd. https://doi.org/10.1002/9781118728482.ch8

Van Haeringen W.A., Den Bieman M., Gillissen G.F., Lankhorst A.E., Kuiper M.T., Van Zutphen L.F. and Van Lith H.A. (2001). Mapping of a QTL for serum HDL cholesterol in the rabbit using AFLP technology, *Journal of Heredity*, 92: 322-326

Velazco, J.G., Jordan, D.R., Mace, E.S., Hunt, C.H., Malosetti, M. and van Eeuwijk, F.A. (2019). Genomic Prediction of Grain Yield and Drought-Adaptation Capacity in Sorghum is Enhanced by Multi-Trait Analysis. *Frontiers in Plant Science*, 10: 997

Verma, R., Kumar, R. and Nath, A. (2018). Drought Resistance Mechanism and Adaptation to Water Stress in Sorghum [*Sorghum bicolor* (L.) Moench]. *International Journal of Bio-resource and Stress Management*, 9(1): 167-172. Doi: HTTPS://DOI.ORG/10.23910/IJBSM/2018.9.1.3C0472

Vieira, M.L.C., Santini, L., Diniz, A.L. and Munhoz, C.F. (2016). Microsatellite markers: what they mean and why they are so useful. *Genetics and Molecular Biology*, 39(3)

Vignal, A., Milan, D., Sancristobal, M. and Eggen, A. (2002). A review on SNP and other types of molecular markers and their use in animal genetics. *Genetics Selection Evolution*, 34: 275. https://doi.org/10.1186/1297-9686-34-3-275

Wang, G., Mahalingan, R. and Knap, H.T. (1998). (C-A) and (GA) anchored simple sequence repeats (ASSRs) generated polymorphism in soybean, *Glycine max* (L.) Merr. *Theoretical and Applied Genetics*, 96: 1086–1096

Waqar-Ul-Haq, M., Malik, F., Rashid, M., Munir, M. and Akram, Z. (2008). Evaluation and Estimation of Heritability and Genetic Advancement for Yield Related Attributes in Wheat Lines. *Pakistan Journal of Botany*, 40: 1699-1702

Wasaya, A., Zhang, X., Fang, Q. and Yan, Z. (2018). Root Phenotyping for Drought Tolerance: A Review. *Agronomy*, 8, 241

Williams, J.G.K., Kubelik, A.R., Livak, J.K., Rafalsk, J.A. and Tingey, S.V. (1990). DNA polymorphisms amplified by arbitrary primers are useful as genetic markers. *Nucleic Acids Research*, 18: 6531-6535

Vos, P., Hogers, R., Bleeker, M., Reijans, M., van de Lee, T., Hornes, M., Frijters, A., Pot, J., Peleman, J. and Kuiper, M. (1995). AFLP: A new technique for DNA finger printing. *Nucleic Acids Research*, 23: 4407–4414

Walulu, R.S., Rosenow, D.T., Wester, D.B. and Nguyen, H.T. (1994). Inheritance of the Stay Green Trait in Sorghum. *Crop Science*, 34(4): 970–972

Wani, S.P., Albrizio, R. and Vajja, N.R. (2012). Sorghum. In: Steduto, P, Hsiao, TC, Fereres E, Raes D (eds) *Crop yield response to water. FAO, Irrigation and drainage paper number*, 66: 144–151

Wanous, M.K., Miller, F.R. and Rosenow, D.T. (1991). Evaluationof visual rating scales for green leaf retention in sorghum. *Crop Science*, 31: 1691-1694

Weeden, N., Timmerman, G. and LU, J. (1993). Identifying and mapping genes of economic significance. *Euphytica*, 73(1-2): 191- 198

Welsh, J. and McCelland, M. (1990). Fingerprinting genomes using PCR with arbitrary primers. *Nucleic Acids Research*, 18: 7213-7218

Wenzel, W.G. (1999). Effect of moisture stress on sorghum yield and its components, *South African Journal of Plant and Soil*, 16(3): 153-157. DOI:10.1080/02571862.1999.10635002

Wu, K.S., Jones, R., Danneberger, L. and Scolnik, P.A. (1994). Detection of microsatellite polymorphisms without cloning. *Nucleic Acids Research*, 22: 3257–3258

Wuensch, K.L. (2020). Binary Logistic Regression with SPSS. Retrieved from http://core.ecu.edu/psyc/wuenschk/MV/Multreg/Logistic-SPSS.PDF on 22nd May, 2020

Wortmann, C.S., Abede, G., Mburu, C., Kayuki, K.C. and Xerinda, S. (2006). Atlas of Sorghum: Production in Five Countries in Eastern Africa. International Sorghum and Millet Collaborative Research Support Programm, Lincoln, Nebraska, USA, pp. 63

Xiong, L., Wang, R.G., Mao, G. and Koczan, J.M. (2006). Identification of Drought Tolerance Determinants by Genetic Analysis of Root Response to Drought Stress and Abscisic Acid. *Plant Physiology*, 142(3): 1065–1074

Xu, W., Subudhi, P.K, Crasta, O.R., Rosenow, D.T., Mullet, J.E. and Nguyen, H.T. (2000). Molecular mapping of QTLs conferring staygreen in grain sorghum *(Sorghum bicolor* L. Moench). *Genome*, 43: 461–469

Yambao, E.B., Ingram, K.T. and Real, J.G. (1992). Root xylem influence on the water relations and drought resistance of rice. *Journal of Experimental Botany*, 43: 925–932

Yang, W., de Oliveira, A.C., Godwin,I., Schertz, K. and Bennetzen, J.L. (1996). Comparison of DNA marker technologies in characterizing plant genome diversity: variability in Chinese sorghums. *Crop Science*, 36: 1669–1676

Ye, X., Avendano, S., Dekkers, J.C.M. and Lamont, S.J. (2006). Association of twelve immune-related genes with performance of three broiler lines in two different hygiene environments. *Poultry Science*, 85: 1555–1569

Yu, H., Chen, X., Hong, Y.Y., Wang, Y., Xu, P., Ke, S.D., Liu, H., Zhu, J., Oliver, D.J. and Xiang, C. (2008). Activated expression of an *Arabidopsis* HD-START protein confers drought tolerance with improved root system and reduced stomatal density. *Plant Cell*, 20(4): 1134–1151. doi: 10.1105/tpc.108.058263

Zaeifizade, M. and Goliov, R. (2009). The Effect of the Interaction between Genotypes and Drought Stress on the Superoxide Dismutase and Chlorophyll Content in Durum Wheat Landraces. *Turkish Journal of Biology*, 33: 1-7

Zhang, F., Zhu, K., Wang, Y.Q., Zhang, Z.P., Lu, F., Yu, H.Q. and Zou, J.Q. (2019). Changes in photosynthetic and chlorophyll fluorescence haracteristics of sorghum under drought and waterlogging stress. *Photosynthetica*, 57(4): 1156-1164

Zietkiewicz, E., Rafalski, A. and Labuda, D. (1994). Genome fingerprinting by simple sequence repeat (SSR)-anchored polymerase chain reaction amplification. *Genomics*, 20: 176–183

Zou, G., Zhai, G., Feng, Q., Yan, S., Wang, A., Zhao, Q., Shao, J., Zhang, Z., Zou, J., Han, B. and Tao, Y. (2012).Identification of QTLs for eight agronomically important traits using an ultra-high-density map based on SNPs generated from high-throughput sequencing in sorghum under contrasting photoperiods. *Journal of Experimental Botany*, 63(15): 5451–5462

Zhu-Salzman, K., Salzman, R.A., Ahn, J.E. and Koiwa, H. (2004).Transcriptional regulation of sorghum defense determinants against a phloem-feeding aphid. *Plant Physiology*, 134: 420–431

APPENDICES

Appendix 1: Semi-structured questionnaire for focus group discussions on constraints of sorghum production, coping mechanism for drought and trait preference by sorghum farmers

I. Identification

Name of facilitator----------Date of discussion: ---/----/201-----

Working station----------Designation----------Region----------

Location of discussion: Village----------Ward----------

District----------Region----------

List of farmers

S/N	Name	Age	Gender		Mobile number
1					
2					
3					
4					
5					
6					
7					
8					
9					
10					

1. Do you involve with agricultural activities in your village? Yes---No----. If yes, which crops contribute income in your households? Starting with the highest to the lowest

i.

ii.

iii.

iv.

v.

2. What other sources that contribute income rather than crops?

i.

ii.

iii.

iv.

3. Do you grow sorghum? Yes [ ], No [ ], If yes why? ----------

---

4. Have you ever heard improved sorghum varieties? Yes [ ], No [ ] If yes, who introduced to you? Agricultural researchers [ ], International Organization (ICRISAT) [ ], NGOs [ ], Private Companies [ ], Others ----------

5. Which varieties do you grow in your village? Local [ ], Improved [ ], both [ ], Why? -------

---

6. Where do you access seed? Own saving seed [ ], Agro-dealers [ ], Agro researchers [ ], NGOs [ ] Other ----------

7. Which sorghum varieties are grown in your village and their maturity period?

S/N	Improved varieties	Early maturity (80-90 days	Medium maturity (95-105days)	Late maturity (above 110 days)
1				
2				
3				
4				
S/N	Local varieties	Early maturity (80-90 days	Medium maturity (95-105days)	Late maturity (above 110 days)
1				
2				
3				
4				

8. Why do you prefer these/this varieties/variety? ----------

---

9. Do you apply fertilizers in sorghum? Yes [ ], No [ ]

If no, why?----------

---

10. If yes, which type of fertilizers do you apply?

Inorganic fertilizers [ ], Organic fertilizers [ ], Both fertilizers [ ]

11. Do you face the problem of pests and diseases on sorghum? Yes [ ], No[ ]. If yes, list.

		Impact of pest in sorghum		
S/N	**Common Pest**	**Low**	**Medium**	**High**
1				
2				
3				
4				
5				

		Impact of disease in sorghum		
S/N	**Disease**	**Low**	**Medium**	**High**
1				
2				
3				
4				

12. Do you control pests and diseases by applying pesticides? yes [ ], No [ ], if no why? ------

---

13. What is the average yield of improved varieties do you get (bags/acre ----------, Local varieties (bags/acre or hectare) ----------

14. Is drought stressing a problem on sorghum crop in your village? Yes [ ], No [ ]

If yes, which stage of growth is highly affected; early growth [ ], vegetative growth [ ], post flowering growth [ ]

15. Do you have drought tolerance sorghum varieties in your village? Yes [ ], No [ ],

If yes, mention 1----------2----------3----------4----------

16. Comparing local and improved sorghum varieties, which is frequently affected by terminal drought stress? ----------

17. What should be done to reduce terminal drought stress effect on sorghum?

1----------2----------3----------
----------4----------5----------6----------
----

18. Is there need to develop new drought tolerance sorghum? Yes [ ], No [ ] If yes or no, why? ----------

19. Which criteria do you use for selection of suitable sorghum varieties?

Criteria	1st	2nd	3rd	4th	5th
disease and pest resistant					
High yielding					
Drought tolerant					
Striga spp tolerant					
Grain colour					
Early Maturity					
Late Maturity					
Storability					
Market availability					
Plant height					
Taste					
Tolerant to bird attack					
Grain weight					

Shelling					

Others----------

20. What do you think are constraints of low yield in sorghum?

1----------2----------3----------
4----------5----------6----------

21. What should be done to improved sorghum production in your village?

1----------2----------
----------3----------4----------

22. What are burning issues regarding sorghum production out of what have been discussed?

1----------2----------3------
----------4----------5----------

23. What do you think should be done to address those issues?

1----------

2----------

3----------

4----------

Appendix 2: Semi-structure questionnaires for Individual interview to understand farmers' perception on sorghum production

Questionnaire number----------

I. Identification

Name of interviewer----------Date of interview: ---/----/201-----
Working station----------Designation----------Region----------

Location of interview: Village----------Ward----------

District----------Region----------

Name of respondent: ----------

Gender: Male [ ] Female [ ] Age: ----------.

Role: Head of the household [ ]; Household activities [ ]; others----------

Highest level of educational: Illiterate [ ]; primary [ ]; secondary [ ]; diploma [ ] university [ ]

i. Number of family members in the household: [ ]

ii. Number of females in the household: [ ]

iii. Number of men in the household: [ ]

iv. How many acres/hectares do you own in your household? Acres [ ] or hectares [ ]

v. How many acres used for sorghum production? Acres [ ] or hectares [ ]

2. Which crops (Starting with the highest rank) contribute income in your household?

Crop	**1st**	**2nd**	**3rd**	**4th**	**5th**	**6th**	**7th**
Maize							
Pearl millet							
Cassava							
Sorghum							
Sunflower							
Bambara nut							
Finger millet							
Grape							
Ground nut							
Sweet potato							
Rice							
Cow pea							

3. Do you grow sorghum? Yes [ ], No [ ], If yes why? ----------

---

4. How long have you been growing? 1 years [ ], 2 years [ ] 3 years [ ], above 3 years [ ]

5. What is the amount of sorghum used for food by household? ----------bags

6. What is the amount of sorghum sold ? ----------bags

7. Have you ever heard improved sorghum varieties? Yes [ ], No [ ] If yes, who introduced to you? Agricultural researchers [ ], International Organization (ICRISAT) [ ], NGOs [ ], Private Companies [ ], Others ----------

8. Which varieties do you grow? Local [ ], Improved [ ], both [ ], Why? ----------

---

9. Where do you get seed? Own saving seed [ ], Agro-dealers [ ], Agro researchers [ ], NGOs [ ] Other ----------

10. Which sorghum varieties are grown in your village and their maturity period?

S/N	**Improved varieties**	**Early maturity (80-90 days**	**Medium maturity (95-105days)**	**Late maturity (above 110 days)**
1				
2				
3				
4				
S/N	**Local varieties**	**Early maturity (80-90 days**	**Medium maturity (95-105days)**	**Late maturity (above 110 days)**
1				
2				
3				

4				

11. Why do you prefer these/this varieties/variety? ----------
---

12. Do you apply fertilizers in sorghum? Yes [ ], No [ ]

If no, why?----------
---

13. If yes, which type of fertilizers do you apply?

Inorganic fertilizers [ ], Organic fertilizers [ ], Both fertilizers [ ]

14. Do you face the problem of pests and diseases on sorghum? Yes [ ], No[ ]. If yes, list.

		Impact of pest in sorghum		
S/N	**Common Pest**	**Low**	**Medium**	**High**
1				
2				
3				
4				
5				

		Impact of disease in sorghum

S/N	Disease	Low	Medium	High
1				
2				
3				
4				

15. Do you control pests and diseases by applying pesticides? yes [ ], No [ ], if no why? ------

---

16. What is the average yield of improved varieties do you get (bags/acre ----------, Local varieties (bags/acre or hectare) ----------

17. Is drought stressing a problem on sorghum crop in your village? Yes [ ], No [ ]

If yes, which type of drought is highly affecting crop growth? Early drought (young seedlings) [ ], mid drought (vegetative growth) [ ], terminal drought (flowering/seed setting) [ ],

18. What is the trend of rainfall from the past 10 years ago to now? is increasing[ ], is reducing[ ], is constant [ ]

19. Do you think there is need to develop new drought tolerant sorghum varieties? Yes [ ], No [ ] If yes or no why? ----------

20. Do you have drought tolerance sorghum varieties in your village? Yes [ ], No [ ],

If yes, mention 1……2……..3……..4……

21. How do you overcome drought? 1----------2----------

----------3----------4----------

----------5----------6----------

22. Do you have access to market? Yes [ ], No [ ]

If yes what is the price of sorghum grain per bag (100kg) Tsh --------- or price per bucket (18-20kg) Tsh ----------If no, where do you sell? ----------At what price--------

23. Which criteria do you use for selection of suitable sorghum varieties?

Criteria	**1st**	**2nd**	**3rd**	**4th**	**5th**
disease and pest resistant					
High yielding					
Drought tolerant					
Striga spp tolerant					
Grain colour					
Early Maturity					
Late Maturity					
Storability					
Market availability					
Plant height					
Taste					
Tolerant to bird attack					
Grain weight					
Shelling					

Others----------

24. What do you think are constraints of low yield in sorghum?

1----------2----------3----------

4----------5----------6----------

25. What should be done to improved sorghum production in your village?

1----------2----------

3----------4----------

Appendix 3. Selection of 10% best performance genotypes from BC_2F_1 populations for traits of plant height, STG, total number green leaves at maturity, panicle length, panicle width, and grain weight per plant.

Table 6.1. Selection of 10% best performance genotypes from BC_2F_1 populations for traits PH, STG, GLM, PL, PW, EX and GWT with B35 donor parent and Wahi and Hakika recurrent parents background

Genotypes	PH	STG	NGLM	PL	PW	EX	GWT (kg)
B35*Wahi_33	96.5	5	3	23	5.5	2	41.8
B35*Wahi_77	114	5	1	30.5	7.5	3	90.7
B35*Wahi_78	109	4	3	33	6.3	2	71.7
B35*Wahi_80	111	4	2	34	8	3	106.4
B35*Wahi_82	123	5	2	34	8	2	101
B35*Wahi_83	114.5	5	2	32	7.5	3	83.2
B35*Wahi_85	111	5	1	30.5	6	2	67.4
B35*Wahi_89	121	5	3	31.4	7	2	97.5
B35*Wahi_90	112.2	6	1	31	6	3	79.3
B35*Wahi_93	120.5	5	3	34	5	2	60.2
B35*Wahi_103	107	5	2	27	6	2	62.7
B35*Wahi_105	121	4	3	31	4.8	2	60.8
S35*Hakika_118	130	6	2	30	8	2	77.8
S35*Hakika_130	121	5	1	31	6.6	2	76.5
S35*Hakika_131	140	5	2	30.5	6.5	2	82.8
S35*Hakika_133	128	6	1	25	6	2	72
S35*Hakika_138	119	6	2	25	6	3	54.1

PH- Plant height (cm), STG- Stay green, Total number of green at maturity, PL- Panicle length (cm), PW- Panicle width (cm), EX- Inflorescence exersion, GWT- Grain weight (g)

Table 6.2. Selection of 10% best performance genotypes from BC_2F_1 populations for traits PH, STG, GLM, PL, PW, EX and GWT with B35 donor parent, and Macia recurrent parents background

Genotypes	PH	STG	NGLM	PL	PW	EX	GWT (kg)
B35*NACO_241	146	5	2	24	8	2	113.4
B35*NACO_242	135	6	0	25.5	8	3	120.4
B35*NACO_246	140	5	2	26	8.2	3	119.6
B35*NACO_255	135	6	1	24.5	6	3	100
B35*NACO_260	123.8	6	0	21	8	2	96
B35*NACO_263	131	6	1	21.5	8	2	102.1
B35*NACO_273	158.8	6	2	21.5	6.5	3	103.4
B35*NACO_291	134	5	2	25.5	8	3	100
B35*NACO_305	143	5	2	23	8	4	106.1
B35*NACO_307	147.8	5	3	26	8.5	4	147.3
B35*NACO_309	141	4	3	26	8.8	3	122.2
B35*NACO_311	148.5	4	2	24	8.5	2	106
B35*NACO_312	144.5	4	3	23.5	7.5	3	104.6
B35*NACO_314	131	4	5	24	9	2	96.2
B35*NACO_316	145	5	5	24	8	3	154.2
B35*NACO_317	121	6	2	22	7.5	3	113.3
B35*Macia_336	92	9	0	20	3.8	3	26.1
B35*Macia_343	110	7	0	23	6	3	26.6
B35*Macia_359	125	5	1	22.3	4.4	3	24
B35*Macia_363	135.8	7	0	22	4	3	29.6
B35*Macia_381	112.5	8	0	22	4	3	24.7
B35*Macia_384	135.5	7	1	25	6	3	24.3
B35*Macia_391	122	7	1	24	4	3	28.6
B35*Macia_394	156	7	1	23	6	3	33.3
B35*Macia_399	123	8	0	20	5	2	24.8

PH- Plant height (cm), STG- Stay green, Total number of green at maturity, PL- Panicle length (cm), PW- Panicle width (cm), IEX- Inflorescence exersion, GWT- Grain weight (G), NACO- NACO Mtama 1

Table 6.3. Selection of 10% best performance genotypes of BC2F1 populations for PH,STG, NGLM, PL, PW, EX and GWTwith B35 donor parent and Seguifa and Pato recurrent parents background

Genotypes	PH	STG	NGLM	PL	PW	EX	GWT/P
B35*Seguifa_408	143.8	5	4	20	7	3	114.7
B35*Seguifa_415	127.7	5	3	19	5	3	141
B35*Seguifa_425	134.9	6	1	24	8	2	85
B35*Seguifa_428	126.8	5	1	23	7	3	86.6
B35*Seguifa_436	143	5	2	20	7	3	91.8
B35*Seguifa_438	147.3	5	3	21.5	7	3	103.4
B35*Seguifa_444	141.2	5	2	21	7	3	107.7
B35*Seguifa_456	151.8	6	1	23	6.5	3	88.2
B35*Seguifa_460	147	5	3	23	6.5	2	102.4
B35*Seguifa_462	154.5	5	3	20.5	7	3	84.6
B35*Seguifa_465	132.5	5	2	20	7	3	97.9
B35*Seguifa_467	160	6	1	20.5	6	3	90.2
B35*Seguifa_468	130.5	6	1	22	8.3	3	137
B35*Seguifa_469	127.5	6	1	22.5	7	3	98.7
B35*Seguifa_480	169.5	6	1	21	6	3	93
B35*Seguifa_486	130	6	1	22.5	6	2	100.6
B35*Seguifa_487	117.5	6	1	19.5	7	3	92.4
B35*Seguifa_488	150.1	6	1	23	7	3	112.8
B35*Seguifa_493	137	6	1	21	7	3	88
S35*Pato_623	135.5	7	1	19	8	3	91.5
S35*Pato_630	115	8	1	18	8	2	88.7
S35*Pato_661	165.5	6	1	20	8	3	123.9
S35*Pato_668	138	7	1	22	8	2	111.6
S35*Pato_685	148	8	0	21.5	8	3	81.2
S35*Pato_686	149	9	0	24	7	3	83.3
S35*Pato_691	134	8	0	22	8.6	2	88.3
S35*Pato_713	155	6	1	22	8	2	93.1
S35*Pato_715	142	6	1	23.5	8	3	108.8
S35*Pato_721	146	6	1	21.5	7.2	2	103.1
S35*Pato_723	150	6	1	21.5	9	2	116
S35*Pato_726	140	7	0	21.5	7	2	84.7
S35*Pato_730	144	6	1	23	8	2	89.4
S35*Pato_735	155	6	1	18	8	3	101.7
S35*Pato_736	155	7	1	22	8	2	99.8
S35*Pato_737	137	6	1	22	8.8	3	104.1

PH- Plant height (cm), STG- Stay green, Total number of green at maturity, PL- Panicle length (cm), PW- Panicle width (cm), IEX- Inflorescence exersion, GWT- Grain weight (g)