Genetic Diversity Analysis of Lowland Sorghum [Sorghum bicolor (L.) Moench] Landraces under Moisture Stress Conditions and Breeding for Drought Tolerance in North Eastern Ethiopia

Ву

Amelework Beyene Assefa

MSc Plant Science (Molecular Breeding and Genetics) (Waginingen University, the Netherlands)

BSc. Plant Science (Alemaya University of Agriculture, Ethiopia)

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African Center for Crop Improvement
School of Agricultural, Earth and Environmental Sciences
College of Agriculture, Engineering and Science
University of KwaZulu-Natal
Republic of South Africa
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Abstract

Sorghum [Sorghum bicolor (L.) Moench] is one of the most important cereal crops grown in arid and semi-arid regions of the world. The North Eastern regions of Ethiopia are known for its high sorghum production and genetic diversity, and proneness to moisture stress. Globally, moisture stress is one of the major sorghum production constraints limiting genetic gain through breeding. Although, the importance of Ethiopia's sorghum germpalsm has been widely recognized both nationally and internationally, the genetic potential of the germplasm has not yet been fully assessed and exploited in breeding programmes. Therefore, the objectives of this study were: (1) to evaluate sorghum production systems and patterns, major production constraints and cropping mechanisms, varietal diversification, farmers' criteria for choosing varieties over time and space, and adoption of improved varieties, (2) to assess the agro-morphological and molecular diversity and population structure of lowland sorghum landraces collected from different geographic origins using morphological and SSR markers, (3) to assess the performance of sorghum landraces under moisture stress conditions and identify promising lines, and (4) to determine heterosis and combining ability of lowland sorghum landraces for grain yield, yield components and drought tolerance and to identify suitable parents for future hybrid cultivar development for the North Eastern Ethiopia or similar environments.

A survey was conducted in the North Eastern Ethiopia sampling three Administrative Zones, six Districts and 12 Peasant Associations. Data was gathered from a total of 171 farmers and analyzed using SPSS statistical package. The results suggest that the performance of sorghum was generally poor mainly due to moisture stress, pests, diseases, weeds, farmland fragmentation due to demographic pressure, poor soil fertility, and poor performance of the local varieties. The productivity of sorghum was also largely hindered by the use of inherently poor yielding local landraces as farmers were forced to abandon their high yielding, big-headed and late maturing sorghum varieties due to the prevalence of recurrent moisture stress. The survey found that the region is as a rich source of genetic diversity and more than 70 distinct sorghum landraces were identified. The majority of the farmers grew their local landraces, despite the accessibility and availability of many improved sorghum varieties and production packages. Farmers were willing to adopt the improved varieties if they had farmers preferred attributes such as as red seed colour, tall with high biomass yield. To benefit the most from the available improved technologies, farmers have to be part of the breeding process right from the very beginning.

Lowland sorghum accessions which exhibited farmer-desired traits were selected from the entire landrace germplasm collection at the national gene bank of Ethiopia. Field evaluations of the selected 278 landraces together with checks were held at Sirinka and Kobo agricultural research stations for 12 qualitative and 10 quantitative traits under stress and non-stress conditions. Two hundrad landraces were selected on the basis of their morphological distinctiveness and drought tolerance, in terms of earliness and yield stability. Molecular level diversity assessment was conducted using 30 SSR markers. Considerable magnitude of variation was observed among landraces between and within geographic origin for most of the traits studied. The morphological variability was also complemented by high molecular markers diversity. Thirty two pure lines were selected for inclusion as parents in the sorghum breeding programme for yield and drought tolerance.

The selected lines were then crossed to four cytoplasmic male-sterile lines that had different cytoplasm systems (A₁ to A₄₎ using a line x tester mating design scheme. The 32 parents, together with the 128 hybrids and 4 check varieties were evaluated for grain yield, yield components traits and drought tolerance under stress and non-stress environments. Data were analysed using GenStat statistical package following a fixed effects model. Nonadditive gene action was predominant in controlling plant height, grain yield, above ground biomass, grain filling duration, 100-seed weight and panicle weight, whereas additive gene action was found more important in controlling days to 50% anthesis and panicle length. Novel landraces with high GCA effects were selected including 214838-A, 242039-B, 75454, 73056-B, and 242050-A which will serve as potential parents for cultivar development. Similarly, the study identified new experimental hybrids i.e. ICSA 749 x 242039-B, ICSA 756 x 242049-B, ICSA 756 x 75454, ICSA 756 x 73059 and ICSA 756 x 214855 with high SCA effects and heterosis for grain yield which will be forwarded for further stability analysis and farmers participatory selections at representative growing environments. In general, the study identified invaluable sorghum germplasm and candidate hybrids useful for further breeding and conservation strategies.

Declaration

- I, Amelework Beyene, declare that
 - 1. The research reported in this thesis, except where otherwise indicated, is my original research.
 - 2. This thesis has not been submitted for any degree or examination at any other university.
 - 3. This thesis does not contain other persons' data, pictures, graphs or other information, unless specifically acknowledged as being sourced from other persons.
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Signed
Amelework Beyene Assefa
As the candidate's supervisors, we agree to the submission of this thesis:
Prof. Shimelis Hussein (Supervisor)
Prof. Pangirayi Tongoona (Co-Supervisor)

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Dedication

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Introduction to Thesis

The importance of agriculture in North Eastern Ethiopia

Ethiopia situated in east Africa has an area of 1.104 million square kilometres and estimated human population of 83 million (CSA, 2011). The Amhara National Regional State (ANRS) is the second largest region among the nine regions of Ethiopia. It occupies much of the north western and north eastern parts of Ethiopia. The total area is about 0.16 million km² constituting about 14.4% of the total area of the country. According to the census estimates of 2011, the total population of the region is estimated to be 18.86 million from the total population of 84.3 and contributing 23.4% of the human resource (CSA, 2011). From the total population 88% depend on agriculture for their livelihoods, which is higher than the national average of 85%. From this one can derive that agriculture is the mainstay economic sector in the region. It accounts for about 55.8% of the GDP of the regional state. This figure is still high when compared to the national average of 45%. Agriculture is the major source of food for the people and also raw materials for local industries and export earnings. The total cultivated land covered with different crops, in the region in the year 2011 was about 4.3 million ha. However; this figure showed a 3.3% increase in 2012 (CSA, 2012a). In the agricultural sector, crop production and animal husbandry are the major activities undertaken in the region. With regard to crop production 36 different crop types are grown including cereals, pulses, oil crops, fibre crops, fruits and vegetables in different parts of the region. Among the cereal crops sorghum, tef and maize are the major grain crops for millions of people in the region (CSA, 2012a).

Rationale for sorghum improvement

Sorghum [Sorghum bicolor (L.) Moench] ranks fifth worldwide after wheat, rice, maize, and barley (FAO, 2012). The crop is produced for its grain which is used for food, and stalks used for fodder and building materials in developing countries. In developed countries, grain or sweet stem sorghum is used primarily as animal feed, making sugar, syrup, and molasses (Doggett and Rao, 1995; Duncan, 1996). Sorghum is a widely adaptable crop, though its production has been limited to water- and heat-stresses within subtropics and tropical regions of the world (Doggett, 1988; House, 1995). Under optimal conditions, sorghum's yield potential is comparable to other cereals such as rice and wheat.

In Ethiopia, sorghum is the main staple food crop, ranking third after tef, and maize in total production and it ranks second after maize in productivity per hectare and in area harvested (CSA, 2012a). It is grown in almost all regions covering a total land area of 1.9 million ha (CSA, 2012a). The major sorghum production areas of the country include: Oromia at 38.6%, Amhara (38.1%), Tigray (11.2%), and Southern Nations Nationalities and Peoples (SNNP) (6.4%) (Figure 1.1.1). From 1998 to 2002, there was a noticeable drastic decrease in the hectarage under sorghum cultivation in Ethiopia. This was attaributed to land redistribution policy and seasonal changes (Teshome *et al.*, 2007). In 2012 cropping season, 79% (9.6 million ha) of the total land area under field crop cultivation was for cereals, of which sorghum accounted for 20% (CSA, 2012a).

Cereals contribute 86% (about 18.8 million tons) of the total field crop production of Ethiopia of which sorghum contributes 21% (4 million tons) (CSA, 2012a). In the same cropping season, the area under sorghum cultivation increased by 1.4% while productivity decreased by 0.22% as compared to the previous cropping season (CSA, 2012a). In the Amhara National Regional State, where the current study was undertaken, sorghum is the second most important food crop after tef. In this region, sorghum accounts for 22.5% of the total area covered by cereals and 25% of the total cereal production (CSA, 2012a).

The ability of sorghum to withstand drought stress and give reasonable yields under adverse environmental conditions has crowned its importance as a food security crop in arid and semi-arid lowlands (House, 1995). In stress environments, sorghum is the dominant crop and receives less agricultural inputs than any other major cereals (McGuire, 2008). It is a good source of income for small scale farmers because of its wide range of uses (Teshome *et al.*, 1999; McGuire, 2000). Despite the importance of the crop, its productivity is very low. On average, 1.5 t/ha of grain yield is obtained in Ethiopia, which is much lower than 3.8 t/ha average yield obtained in the USA (FAOSTAT, 2012). The low yields are caused by various production constraints which include biotic stresses (insects, diseases, birds and weeds), abiotic factors (moisture stress and low soil fertility) and continued use of low yielding traditional cultivars (Wortmann *et al.*, 2006). For example, 99.6% of the total area under sorghum is covered by traditional cultivars, which are less productive (CSA, 2012 a and b).

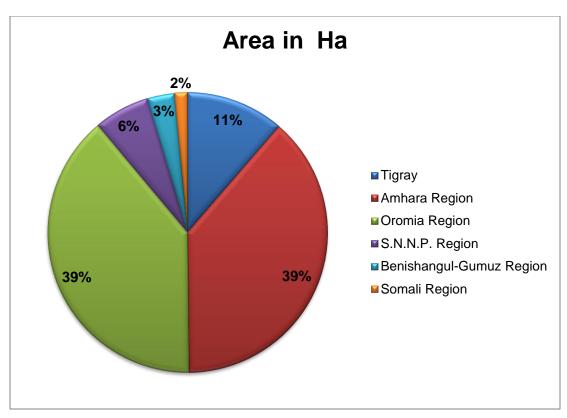


Figure 1.1.1 Area under sorghum in Ethiopia, 2011/12 cropping season (CSA, 2012a)

Rationale for working on moisture stresses

Moisture stress contributes to poor crop performance and low yield. Insufficient, unevenly distributed, and unpredictable rainfall are usually experienced in drier parts of Ethiopia. Typically, rain may be abundant and perhaps wasted through runoff or in some years much rain may fall completely outside the growing season. In other years, the amount of rain may be low and occur after the crops have germinated. Overall, soil moisture may be severely depleted under such conditions. Consequently, in almost all lowland areas, crops are prone to periodic moisture stress in one way or another because of such adverse effects (EARO, 2001). The effect of moisture stress on crop yield is dependent on the stage of plant development. Anthesis and grain filling stages appear to be most vulnerable. Occurrence of drought at these stages may result in reduced yield and/or complete crop failure (Khanna-Chopra and Sinha, 1988). Although drought stress at the beginning of the growing season (seedling stage) will severely affect plant establishment, sorghum has the capacity to recover soon after the onset of rain.

The North and North Eastern (NE) regions of Ethiopia are characterized by rugged topography, unreliable rainfall conditions and very diverse and complex farming systems. The performance

of the agriculture sector depends mainly on the quantity and distribution of rainfall, as it is exclusively rainfed agriculture. The crop growing periods in the NE regions is ranging from 60 to 120 days. According to FAO classification, climates with fewer than 120 days of growing period are described as dry lands (FAO, 1987). Crop productivity is the function of the genetic potential of the crops and of the total environment in which the crops are growing. However, in the dry land areas the environment is more yield limiting and hinders crops from expressing their full genetic potential (Ceccarelli *et al.*, 2004). Evaluation of germplasm in an area of adaption of a particular germplasm collection is vital to develop high yielding stable sorghum varieties (Frankel, 1970). Therefore assessing the performance of local germplasm that have been well adapted and co-evolved with the existing drought conditions, would enable to identify the best performing genotypes.

In Ethiopia, the prevalence of drought has increased from five to six years in the past two decades to every three years. In the past, when the rainfall situation was relatively better, farmers used to grow different late maturing sorghum landraces which could be planted in April/May. In times of delayed onset of rainfall, and shortened rainy seasons, farmers could switch to growing early maturing sorghum landraces. However, due to the prevalent shift in the rainfall patterns, farmers are increasingly growing less productive early maturing sorghum varieties, leading to marginalization or loss of highly productive late maturing sorghum landraces. This suggests an urgent need to conserve traditional sorghum varieties. Hence, knowledge of the diversity of sorghum landraces would be highly disrable.

Importance of genetic diversity for crop improvement

Genetic diversity is the basis for the success of any plant improvement programme (Hajjar and Hodgkin, 2007). Genetic diversity provides an extensive range of material fundamental for food, fiber, medicine, and industry (Teshome *et al.*, 1999). It is not only a necessary condition to improve yield and yield stability (Falco and Perrings, 2003; Tilman *et al.*, 2005), but also the raw material used by breeders to develop improved varieties (Buanec, 2005). It is also the basic condition for evolutionary success to species survival and adaptation (Rao and Hodgkin, 2002). Genetic diversity in specific crops includes landraces, primitive forms, cultivars, introductions, weedy and wild relatives of crop species (Harlan, 1992).

Ethiopia is one of the centers of crop diversity in the world (Vavilov, 1951). The country is is further believed to be the center of origin and domestication for sorghum (Doggett, 1988). The

importance of Ethiopian sorghum germplasm is also well recognized in sorghum improvement programmes both nationally (Gebrekidan, 1973; Gebrekidan and Kebede, 1978; Kebede, 1991) and internationally (Singh and Axtell, 1973; Maiti et al., 1984; Singh, 1985; Subramanian et al., 1987; Doggett, 1988). Previous reports indicated that Ethiopian sorghum germplasm is resistant to shoot fly (Maiti et al., 1984) and disease resistance (Tegegne et al., 1994), and have high grain quality (Singh and Axtell, 1973), high sugar content (Subramanian et al., 1987), high lysine and protein content (Gebrekidan and Kebede, 1978; Reddy et al., 2001), cold tolerance (Singh, 1985) and drought tolerance (Subudhi et al., 2000). The large variation that has been observed in Ethiopian sorghum germplasm is attributable to the diversity of environmental conditions in which it grows such as rainfall, temperature, altitude, growing period, and edaphic factors. In Ethiopia, the largest genetic diversity in sorghum is reported in the northeastern parts of the country. In addition, the advantages with landraces, however, are that they perform well under moisture stress because they are best adapted to the local conditions. Further landraces possess farmers' preferred attributes, despite their low productivity. The importance of sorghum landraces is also well recognized, particularly in the lowland areas where rainfall is unreliable and crop failures are common due to recurrent drought (Kebede and Menkir, 1987).

The International Plant Genetic Resources Institute (IPGRI) designated Ethiopian sorghum germplasm as one of the top research priorities. The institute has been supporting the Ethiopian Sorghum Improvement Programme (ESIP) in its effort to collect and characterize the crop genetic resources since its inception (IBPGR, 1981). In order to preserve the valuable genetic diversity, ESIP embarked on an extensive collection of landraces in 1973, with financial support from Canada's International Development Research Centre (IDRC) (Yemane and Lee-Smith, 1984). At the early stages, ESIP was mainly involved in exploring the local germplasm and collected more than 8000 farmers' varieties (Gebrekidan, 1975). After 1978, ESIP transferred its entire sorghum collection to the Plant Genetic Resource Centre of Ethiopia and since then breeders occasionally accessed the domestic collection in the genebank (Gebrekidan and Kebede, 1978).

In heterogeneous and marginal environments of Ethiopia where sorghum is mainly growing, farmers have used a wide range of selection criteria when selecting for adaptation to specific agroecology (Teshome *et al.*, 1999). Considerable efforts have been made by both the national and regional research institutions to improve the productivity of the sorghum in the country. However, farmers remained to be reluctant to use the improved varieties. This is because the improved varieties that have been developed so far were dwarf lines with low biomass yield

(Adugna, 2007). Additionally, in the event of good seasons the local landraces yield better than the improved varieties; the improved varieties are liable to bird attack due to their extreme earliness (Adugna, 2007). In sorghum growing belts of Ethiopia, red and brown grain color, tallness, high biomass yield, early maturity, drought resistance, *Striga* resistance and post harvest qualities such as *Injera* and malting quality are the most important farmers' preferred traits for selecting sorghum varieties (Wortmann *et al.*, 2006),

Although, Ethiopia is the centre of origin and domestication for sorghum and there is a wide sorghum genetic diversity available in the country, the sorghum improvement efforts are mainly focused on selection of genotypes from exotic sources (Adugna, 2007). As a result, the potential of the local landraces as sources of breeding material is not yet well known and exploited. Hence, knowledge of the diversity of sorghum landraces as a source of breeding material would be highly important. The proposed study, therefore, seeks to collect and gather indigenous knowledge about local sorghum landraces, characterize them for their response to drought, and assess their potential as a source of novel genes for sorghum breeding programmes.

Research objectives

The specific objectives of the study were as follows:

- to evaluate sorghum production systems and patterns, major production constraints and cropping mechanisms, varietal diversification, farmers' criteria for choosing varieties over time and space, breeding priorities and adoption of improved varieties in the North Eastern Ethiopia,
- to assess the agro-morphological and molecular diversity and population structure of lowland sorghum landraces collected from different geographic origins in Ethiopia using morphological and SSR markers,
- 3. to assess the performance of sorghum landraces under moisture stress conditions and identify promising lines in the North Eastern Ethiopia, and
- 4. to determine heterosis and combining ability of lowland landraces for grain yield, yield components and drought tolerance and to identify suitable parents for future sorghum hybrid cultivar development in the North Eastern Ethiopia or similar environments.

Research hypothesis

This study was conducted to test the following hypotheses:

- 1. In areas of sorghum growing belts of Ethiopia, farmers' preferences and perceptions of sorghum traits especially for drought tolerance are different due to different social, cultural, and economic conditions.
- 2. Considerable genetic diversity and high levels of variation for drought tolerance are available in locally adapted sorghum landraces of Ethiopia.
- 3. Different heterotic groups are represented in the Ethiopian sorghum landraces collection which can be exploited by hybrid breeding.

Outline of this thesis

This thesis consists of six distinct chapters in accordance with a number of activities related to the afore-mentioned objectives. Chapters 2-6 are written as discrete research papers intended for publication containing all the necessary information, some of which might have been presented in other chapters. Some overlap and unavoidable repetition may exist between the chapters and references.

Chapter	Title
-	Introduction to thesis
1	A review of the literature
2	Analysis of the sorghum production systems in the north Eastern Ethiopia: breeding
	priorities and implications on varietal adoption
3	Genetic diversity assessment of lowland sorghum [Sorghum bicolor (L.) Moench]
	landraces of Ethiopia using qualitative traits
4	Genetic diversity assessment of lowland sorghum [Sorghum bicolor (L.) Moench]
	landraces under moisture stress environments using quantitative traits
5	Molecular genetic variation in lowland sorghum [Sorghum bicolor (L.) Moench]
	landraces of Ethiopia assessed by simple sequence repeats (SSRs)
6	Combining ability and heterosis in lowland sorghum [Sorghum bicolor (L.) Moench]
	landraces under moisture stress condition
7	An overview of the research findings

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CHAPTER 1

A Review of the Literature

1.1 Introduction

Sorghum [Sorghum bicolor (L.) Moench] is an important source of food and feed in arid and semi-arid tropics. It is well adapted to drought prone environments; however, the productivity of sorghum is low owing to various biotic, abiotic and socio-economic constraints. Among the abiotic constraints, drought is considered to be the major one in the arid and semi-arid tropics preventing the full genetic expression of most crops. Drought tolerance is a complex quantitative trait controlled by many genetic and environmental factors. The responses of different plants, species and genotypes to drought are variable and are related to developmental stage, duration of drought and evolutionary adaptation of the crop. Drought tolerance involves the interaction of different morphological structures, physiological functions, and biochemical expressions. Improvement of crop performance under moisture stress condition is highly dependent on the proper choice of breeding materials, procedure and screening techniques. In this literature review, research findings from the 1960s up to present are included. Most of the basic researches on mechanism of drought tolerance were done in the early 1980s and the current researches focus on verification and fine-tuning of methodologies. This literature review is divided into five sections. The first section covers topics related to origin and domestication of sorghum. The second section focuses on the production constraints of sorghum in Ethiopia. The third section covers information on farmers' trait preferences in sorghum breeding. The fourth section focuses on the general significance of genetic diversity in plant breeding and the extent of sorghum genetic diversity in Ethiopia. The fifth section gives detailed information on mechanisms and genetics of drought in plants in general and sorghum in particular and information on breeding approaches for drought resistance.

1.2 Sorghum origin and domestication

It is believed that cultivated sorghum [Sorghum bicolor (L.) Moench] was first domesticated in north-eastern Africa. In 1951, Vavilov suggested Ethiopia as a center of origin of sorghum due to the wide variation of the crop (Vavilov, 1951). Some researchers argue for multiple centers of origin for the crop. Stemler et al. (1975) reported that any of the biogeographically, morphological, historical, and evolutionary evidences did not support that sorghum was neither domesticated nor originated in Ethiopia. Some authors suggest the origin of sorghum to be India

(Meadow, 1996), while others have proposed the origin and domestication of sorghum as southern China (Qiao and Zhenshan, 1970) and north China (Kimber, 2000). All theories concerning the origin and domestication of sorghum were based on archaeological evidences. However, the time and location are not necessarily accurate (Kimber, 2000).

Doggett (1988), on the other hand, reported that sorghum is originated in north eastern parts of Africa comprising Ethiopia, Sudan and East Africa. These regions contain the maximum diversity of both wild and cultivated species. Early domestication and selection of the crop in response to environmental factors and human need resulted in the wider variability. The environmental factors included day length, altitude, temperature, rainfall, and soil characteristic. Human needs usually reflect bigger panicle, non-shattering habit, large grain, tall plant height, and early crop duration. The greater diversity is, therefore, partly due to the diverse physical environments occurring in the region and partly due to the interaction of man with the environment (Rao *et al.*, 2002). As a result, new and stable sorghum biotypes were emerged attributed to selection, adaptation, intercrossing, and movement of plant material from place to place. Introduction of new biotypes evolved in other places and intercrossing with the native biotypes resulted in a development of new biotypes. This movement and evolution of biotypes gave rise to five sorghum races: bicolor, caudatum, guinea, kafir, and durra (Rooney, 2000).

1.3 Production constraints of sorghum

The productivity of sorghum in Ethiopia is low (1.5 t/ha) owing to various biotic and abiotic production constraints. Drought, low soil fertility (nutrient deficiency), stem borers, shoot fly, quelea birds *Striga hermonthica*, and other weeds are recognised as major production constraints in Eastern Africa (Wortmann *et al.*, 2006). Although, these constraints cause significant grain yield losses, the relative importance varies from region to region within and among the countries. For example, shoot fly is reported to cause significant yield loss in Ethiopia and Uganda, but is of relatively less important in Mozambique (Figure 1.3.1). Similarly, *Striga* has been cited to be the most important constraint in Kenya, and the second most important in Ethiopia and Uganda. In Ethiopia, drought and *Striga* were found to be very important in north and north eastern parts of the country, whereas quelea birds were seen as a major constraint in the Rift Valley and Southwest lowlands (Wortmann *et al.*, 2006). Research has also shown that moisture deficit during grain filling is most important for Ethiopia and Mozambique. Although mid-season stress has been shown to be relatively less important as compared to other growth stages in Ethiopia, it is said to be very important in other countries

(Simane *et al.*, 1998; Wortmann *et al.*, 2006; Mengistu, 2009). The north-eastern parts of Ethiopia, where this research has been undertaken represents one of the sorghum growing belts of the country. It is worth-noting that drought and *Striga* are equally important in the target region, though for the purpose of this study priority will be given to the former.

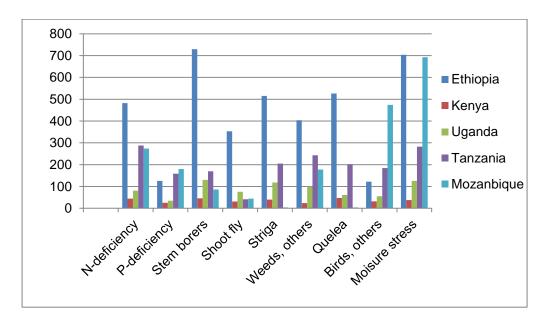


Figure 1.3.1 Estimated losses of grain sorghum (tones/year) due to nine most important constraints in five countries of eastern Africa (Wortmann *et al.*, 2006)

Moisture stress contributes to poor crop performance and yield. In Ethiopia, where more than 50% of the total area is semi-arid (Gamachu, 1977), insufficient, unevenly distributed, and unpredictable rainfall is usually experienced in drier parts of the country. At one point rain may be abundant and perhaps wasted through runoff; in some years much rain may fall completely outside the growing season. In other years the amount of rain may be low after the crops have germinated, soil moisture may be severely depleted. Consequently, in almost all lowland areas crops are prone to periodic moisture stress in one way or another because of the aforementioned realities (EARO, 2001). The effect of moisture stress on crop yield is dependent on the stage of plant development. Anthesis and grain filling stages appear to be more vulnerable; occurrence of drought at these stages may result in reduced yield and/or complete crop failure (Khanna-Chopra and Sinha, 1988; Younesi and Moradi, 2009). Although drought stress at the beginning of the growing season severely affect plant establishment, plants tend to recover soon when the rain falls late (Ramu *et al.*, 2008).

1.4 Farmers' preferences in sorghum breeding

Even if considerable efforts have been made by ESIP to improve the productivity of the crop, farmers have remained reluctant to use the improved varieties. This is due to the reason that in areas of sorghum growing belts of Ethiopia, the improved varieties did not meet the requirements of the small-holder farmers (Adugna, 2007). Additionally, farmers have employed a wide range of selection criteria when selecting for adaptation to specific agroecologies, particularly in heterogeneous and marginal environments (Teshome *et al.*, 1999). Moreover, the specialized exotic semi-dwarf lines were developed to the mechanized and intensive production systems elsewhere (McGuire, 2008). In the event of good seasons the local landraces yield better than the improved varieties; the improved varieties are liable to bird attack due to their extreme earliness, and they have reduced biomass yield (Adugna, 2007).

Sorghum stalk is an important product and is used as cooking fuel, fodder, and construction material. Overall, the value of the stalk is about 40% of the value of grain produced and it is highly valued in Ethiopia (Wortmann *et al.*, 2006). Grain colour is also recognized as an important trait with great preferences for red and brown. Although it is not scientifically proven, red and brown grain types are often associated with high tannin content, and are less preferred by birds and less affected by mold (Wortmann *et al.*, 2006). The process of technology development should include the smallholder farmers and explore their knowledge. The main reason for poor adoption rate and low impact of the improved varieties is lack of breeders' awareness of the traits that farmers desire (McGuire, 2008).

1.5 Genetic diversity and breeding for drought tolerance

Genetic variability is defined as the variability observed in a given crop plant that can be attributed to genes that encode specific traits, and can be transmitted from one generation to the next (Acquaah, 2007). The degree of expression of genes is influenced by the environment. Genetic variability can be created in nature through hybridization and recombination, mutation, and modification of chromosome number and structure. Some variability can be easily recognized and classified into distinct non-overlapping classes, while some other kinds of variability occur in a continuum, and cannot be classified into discrete groups (Acquaah, 2007).

Assessment of genetic variability in crops has strong impact on crop improvement programmes, and conservation of genetic resources (Assar *et al.*, 2005). Genetic variability can be detected at morphological, biochemical or molecular levels. Some genetic variations are manifested as

visible morphological traits (Ayana and Bekele, 1999). The use of qualitative and quantitative morphological traits as techniques for characterization and evaluation of genetic diversity has long been documented and most widely practiced in many crops in general and sorghum in particular (Haussmann *et al.*, 1999; Grenier *et al.*, 2004). The importance of such traits has been influenced by G x E interaction (Newbury and Ford-Lloyd, 1997). However, the application of morphological traits in the analysis of variation continues to be important since data collection does not require expensive technology and such traits are vital in formulation and understanding of ideotypes (Banziger *et al.*, 2006).

Some other genetic variations are compositional or chemical that require various tests for evaluation (Shechter, 1975). Isozymes (Dje et al., 1998; Dje et al., 2000; Ayana et al., 2001; Gregova et al., 2004; Zong et al., 2005) and seed storage proteins (Gepts, 1990) were the most widely used biochemical markers in genetic diversity assessment. Often, the importance of these types of markers is inherently impeded by low polymorphism. The availability of DNA based molecular tools, on the other hand, enables breeders to examine genetic diversity at molecular level (Ayana et al., 2000; Dje et al., 2000, Grenier et al., 2004; Ghebru et al., 2002; Uptmoor et al., 2003; Assar et al., 2005). The application of DNA molecular markers as compared to morphological and biochemical markers overcomes the problem of polymorphism and they are highly informative. Moreover, DNA marker application has facilitated the identification of agronomic traits in wild, traditional and improved germplasm through the dissection of quantitative traits (Tanksely and McCouch, 1997).

Breeding for drought tolerance requires novel sources of resistance (Terán and Singh, 2002). Wild species, traditional varieties, commercial cultivars, and breeding lines are used as sources of genes for drought resistance in most breeding programmes. Bansal and Sinha (1991) suggested exploiting the potential of different species within a crop as a source of resistance. However, Dar *et al.* (2006) advocated the importance of traditional germplasm, as a source of resistance genes for drought in the semi-arid tropics, where moisture stress is the greatest challenge for crop improvement. Blum (2004) also indicated that sorghum is a warm-season and photoperiod sensitive grass that is characterized more by diversity than homogeneity. Similarly, Habyarimana *et al.* (2004) pointed out that the effect of heterogeneity and heterozygosity of tropical landraces of sorghum enables to display high adaptation to drought stress. Although sorghum has originated in Africa, it spread across wide geographic areas, covering a wide range of latitude, longitude, altitude, day length, rainfall, and temperature

regimes, and hence, it adapted to a range of biotic and abiotic stress factors. This resulted in the evolution of several landraces cultivated in their respective sub-regions (Rao *et al.*, 2002).

The early agricultural practices, such as crop domestication and artificial selection resulted in the loss of genetic diversity in cultivated crop species, as compared to their wild progenitors (Doebley et al., 2006; Yamasaki et al., 2007). The extent of the loss depends on the magnitude of the bottleneck of domestication, the population size, and the duration of the bottleneck (Doebley et al., 2006). The loss of diversity was not experienced equally by all genes in the genome during domestication (Doebley et al., 2006; Yamasaki et al., 2007). For genes that do not control favorable phenotypes, the loss is basically a function of the magnitude of the bottleneck of the environment. However, the loss is more severe for genes that determine desirable phenotypes. Case et al. (2005) agreed with the above theory and reported that the wild sorghum genotypes exhibited significantly higher gene diversity than the landraces. Although the landraces were significantly less diverse than the wild type of sorghums, they exhibited 86% of the gene diversity observed in the wild types (Case et al., 2005). Various studies have reported the presence of high genetic diversity in sorghum landraces (Ayana et al., 2000; Ghebru et al., 2002; Geleta et al., 2006; Manzelli et al., 2007; Deu et al., 2010; Nguni et al., 2011). Therefore, the landraces of sorghum are important sources of genes, when breeding for drought resistance. In recent years, researchers have exploited genetic resources with attributes related to drought resistance in different crop plants owing to gene synteny (Sanchez et al., 2002; Shao et al., 2005; Sivaramakrishnan et al., 2006). Conventional breeding through well-designed crosses or biotechnological methods are employed to transfer genes from one crop plant to the other (Mitra, 2001).

1.6 Definition of drought resistance/tolerance

Drought is one of the major global problems affecting crop production worldwide (Jie *et al.*, 2002). In the semi-arid tropics, drought is often the main production factor causing a significant yield loss (Matthews *et al.*, 1990). It is defined as a meteorological event during which precipitation is inadequate to meet crop water requirements that results in a loss of yield below that expected under optimal water supply (Kramer, 1983; Thomas, 1997). It is a normal recurrent feature of climate that can occur in virtually all climatic zones; however, its feature varies significantly from region to region. In the semi-arid tropics where dryland farming is practiced, drought is a common phenomenon that occurs at different periods during the growing season (Blum, 1988). There is also a high season-to-season variability of rainfall, temperature,

and radiation in the tropics. Besides, locations are greatly variable in topographic, soil, existing agricultural practices, and other associated biotic stress factors (Chapman *et al.*, 2000b).

Drought is a combination of temperature (Prasad *et al.*, 2008) and water (Campos *et al.*, 2004) stress effects, in which evapo-transpiration is the major driving force that affects the soil, plant, and atmospheric continuum of the hydrologic cycle (Kramer, 1983). In earlier studies, predictions of drought were mainly based on the amount and distribution of precipitation (Blum, 2011). However, in recent studies soil moisture balance and soil characteristics were introduced in the assessment of drought. Lack of adequate soil moisture or water deficit, affects the ability of plants to grow and complete a normal life cycle (Moussa and Abdel-Aziz, 2008). Drought can have major consequences on growth, development and yield of plants by affecting several physiological, morphological and biochemical processes (Simpson, 1981). It is the major cause of poor crop performance and low yield, and sometimes it causes total crop failure. In the tropics, the probability of drought is highest at the start and end of the growing season.

Drought can occur at both seedling, pre-flowering and post-flowering stages of development, and has the most adverse effect on yield (Tuinstra *et al.*, 1997; Kebede *et al.*, 2001). Drought stress at the seedling stage of development will severely affect plant establishment (Baalbaki *et al.*, 1999). If it occurs at pre-flowering, flowering, or grain filling stages, it may result in reduced yield, or complete crop failure (Blum, 1996). Researchers have analyzed drought tolerance as pre-and post-flowering stresses and the reaction of genotypes to these stresses are variable and controlled by different genetic mechanisms (Rosenow *et al.*, 1996). Pre-anthesis moisture stress has effects on yield components such as stand count, tillering capacity, number of heads and number of seeds per head, while post-anthesis moisture stress has influences on transpiration efficiency, CO₂ fixation and carbohydrate translocation. The latter, in turn, results in low yield and premature plant senescence (Rosenow *et al.*, 1996; Thomas and Howarth, 2000; Vajrabhaya *et al.*, 2001; Xin *et al.*, 2008).

Levitt (1980) defined drought resistance as mechanisms of drought avoidance, recovery, survival and tolerance. These drought tolerance mechanisms are associated with plant survival and production. Survival is the ability of the crop to survive drought irrespective of the yield it produces, while production is the ability of the crop to grow and yield under water stress conditions. Drought resistance can affect biomass or economic yield indirectly via water transpired, water-use efficiency, and harvest index (Solomon and Labuschagne, 2003; Manavalan et al., 2009). Passioura (1996) defined biomass as the product of the amount of

water transpired and water-use efficiency; whereas economic yield is the product of water transpired, water-use efficiency and harvest index.

Drought tolerance is a complex quantitative trait influenced by many genetic and environmental factors (Ceccarelli *et al.*, 2004). The environmental factors include: rainfall, temperature, solar radiation, wind, humidity, and soil characters, such as hardiness, water holding capacity, and nutrient availability (Thomas, 1997). The responses of different plants, species, and genotypes to drought are variable in relation to developmental stage, duration of drought, and evolutionary adaptation of the crop (Sanchez *et al.*, 2002). In sorghum, for example, plants that are adapted to arid and semi-arid environments showed higher drought tolerance than plants of humid origin (Blum and Sullivan, 1986).

1.7 Mechanism of drought resistance in plants

Sorghum being the crop of the tropics, is known for its ability to withstand drought better than any cereal crops. Several studies have been conducted in understanding the mechanism of drought resistance in sorghum and in identifying essential traits for drought tolerance (Blum, 2011). Drought resistance, therefore, involves the interaction of different morphological structures, physiological functions, and biochemical expressions (Mitra 2001; Borrell *et al.* 2006). Sorghum genotypes were found to differ for nearly all recognized drought resistance/tolerance mechanisms, such as reduced plant size, short growth duration, leaf rolling, stomatal conductance, and stay-green (Blum *et al.*, 1989).

1.7.1 Reduced plant size and growth durations

Reduced leaf area index, small and narrow leaf structure, reduced plant stature, and low tillering ability are reported as drought adaptive mechanisms in plants (Richards, 2000; Richards *et al.*, 2002). Mortlock and Hammer (1999) observed that larger plants have larger leaf area and transpire more water than smaller plants. In water limited conditions, transpiration is first restricted by reduction of leaf expansion (Borrell *et al.*, 2001). In addition, genetic dwarfing of tall genotypes improves the grain yield potential of sorghum in arid and semi-arid environments. Most drought tolerant cultivars that have been developed through breeding so far are dwarf in stature. Dwarf cultivars are efficient in balancing assimilate translocation between the developing grain and other vegetative organs as compared to tall genotypes (Kouressy *et al.*, 2008b).

Late flowering and continuous increase in height is not a desirable trait in drought prone areas (Blum *et al.*, 1989; Rai *et al.*, 1999). A comparative genetic analysis of plant height and time of flowering across the Poaceae family revealed that genes affecting these two plant characters are found linked in sorghum (Lin *et al.*, 1995). This largely explained the positive correlation between the two traits. Breeders should always consider the physiological interaction of plant height and flowering, as flowering inhibits apical growth (Smith and Frederiksen, 2000). In dry land environments, high tillering is not also a preferred character in sorghum (Ishikawa *et al.*, 2005). All the dwarf sorghum genotypes have low tillering ability. In rice, however, dwarf genotypes are also characterized by their high tillering ability in contrast with sorghum genotypes (Ishikawa *et al.*, 2005).

Short growing duration is considered as an important trait of drought escape (Blum *et al.*, 1989). The advantage of early maturing genotypes in drought affected areas has long been realized by breeders. On the other hand, most studies showed that high yield potential and late maturity are positively correlated under favorable conditions depending on rate of grain filling potential of the genotypes (Pantuwan *et al.*, 2002; van Oosterom *et al.*, 2006). Hence, drought escape by shortening the growing period is made at the expense of the crops yield potential (Blum, 1988). The traditional tall varieties are characterized by extended crop duration (late maturity), moderately high biomass yield, and low grain yield under conditions of drought. The dwarf cultivars, on the other hand, have reduced biomass yield, and relatively high grain yield. Although dwarfing genes contribute to yield improvement, further increase in yield can be achieved through increasing sink capacity by improving assimilate availability through early expression of stay-green traits and delaying leaf senescence (Kouressy *et al.*, 2008a).

Blum *et al.* (1989) reported an apparent variation in phenology among sorghum genotypes. It was observed that early maturity had an obvious advantage in grain yield and harvest index under drought conditions. In this particular experiment, on average the most resistant cultivar were 11 days earlier than the most susceptible cultivars. However, this author suggested that phenology may not be an exclusive selection criterion *per se*, since other factors have also been involved in affecting a genotype's performance under water stress.

1.7.2 Leaf rolling and stomatal conductance

In plants, stomatal conductance and leaf rolling are found to be reliable physiological indicators of drought tolerance (Kadioglu and Terzi, 2007). Bittman and Simpson (1989) reported that

stomatal conductance and leaf rolling are strongly associated with leaf water potential. Dingkuhan *et al.* (1999), on the other hand, indicated that these two mechanisms are controlled by different factors; as stomatal conductance is controlled by soil moisture dependent root signal, while leaf rolling is controlled by leaf water potential.

Hsiao *et al.* (1984) reported the strong correlation of leaf rolling with leaf water potential, and hence, leaf rolling is used as a visual scoring criterion for selecting drought resistance in plants. The rolling of leaves, usually, occurs following the reduction in leaf water potential. However, the degree of leaf rolling depends on the ability of the plant to adjust osmotically at low leaf water potential (McCree *et al.*, 1984; Flower *et al.*, 1990). Plants with high osmotic adjustment revealed less degree of leaf rolling, and hence, less degree of leaf rolling is considered as an indicator of a greater degree of desiccation avoidance, through a deep root system (Hsiao *et al.*, 1984). Khan *et al.* (2007) found that drought tolerant genotypes in faba bean exhibited lower stomatal conductance associated with increased leaf temperature, which gives rise to high transpiration efficiency and lower carbon isotope discrimination. It was also suggested that the increased leaf temperature and transpiration rate was due to controlled transpirational cooling system induced by stomatal closure. The drought susceptible genotypes, on the other hand, showed higher stomatal conductance and lower leaf temperature that resulted in lower transpiration rate.

There is a high degree of controversy on the importance of the two traits such as leaf rolling and stomatal conductance as a drought resistance mechanism. Redmann (1985) and Begg (1980) indicated that leaf rolling has detrimental effect on transpiration rate through changes in leaf stomatal conductance, and reduction in effective leaf area. They further argued that leaf rolling enhances stomatal closure by increasing leaf resistance to water loss. However, Heckathorn and DeLucia (1991) argued that leaf rolling had positive effects on reducing leaf temperature and loss of water by decreasing the incident irradiation. These authors explained that stomatal closure alone caused 70-80% decrease in transpiration rate in both species, however, leaf rolling caused a decrease of only 2% extra transpiration rate in both species. Therefore, leaf rolling has less value in reducing water loss compared to stomatal closure in these species. Although leaf rolling has insignificant effect on transpiration and leaf temperature, it may increase the survival of plants by enhancing stomatal closure in extreme drought conditions (Heckathorn and DeLucia, 1991). The significance of using these traits as a physiological indicator of plant drought adaptive mechanisms depends on the crop species and the environment. In conditions where there are no sofesticated instruments to measure transpiration

efficiency and stomatal conductance, leaf rolling is good indicator of drought resistance/tolerance.

1.7.3 Stay-green or non-senescence

Leaf senescence is a programmed cell death resulting from drought and other environmental stress factors. It is characterized by loss of chlorophyll and progressive decline in photosynthetic capacity (Tuinstra *et al.*, 1997; Crasta *et al.*, 1999; Tao *et al.*, 2000). Stay-green, on the other hand, is a post-anthesis drought resistance trait in plants that provides resistance to pre-mature leaf senescence to the plant under severe moisture stress condition during grain filling stage. It contributes to an improved yield and yield stability under moisture stress condition.

Pre-mature plant tissue death usually occurs when plants are subjected to water stress during the grain filling period in sorghum (Rosenow and Clark, 1981). Stay-green is associated with a higher level of chlorophyll content, cytokinin, and leaf nitrogen concentration under moisture stress conditions. Xu et al. (2000) reported that all the stay-green lines showed higher levels of chlorophyll content than the normal lines. Hence, the visual scoring of leaf and plant senescence for stay-green response suggested by Wanous et al (1991) is also validated by Xu et al. (2000). McBee (1984), Thomas and Smart (1993) and Thomas and Howarth (2000) found that stay-green sorghum lines exhibited high levels of cytokinin, suggesting that the reduced senescence rate of the stay-green lines may be due to a higher level of cytokinin. Furthermore, stay-green genotypes are also associated with higher leaf nitrogen concentration, particularly at flowering (Borrell and Hammer, 2000; Borrell et al., 2000a), and basal stem sugars (Duncan, 1984), than senescent genotypes. This suggests that the stay-green trait may possibly contribute to higher transpiration efficiency of non-senescent genotypes. To the contrary, leaf senescence is characterized by loss of chlorophyll and progressive decline in photosynthetic capacity (Tuinstra et al., 1997; Crasta et al., 1999; Tao et al., 2000).

Greater green-leaf area duration is observed to occur during grain filling stage and, therefore, van Oosterom *et al.* (1996) described that the stay-green as post-flowering green leaf area duration (GLAD). The stay-green sorghum lines appear to be the combined effect of three distinct factors namely, green leaf area at flowering, time of onset of senescence, and subsequent rate of senescence (van Oosterom *et al.*, 1996; Borrell *et al.*, 2000a). Large variations have been reported in the proportions of green-leaf area among different genotypes as a result of combined effects of differences in onset and rate of senescence (Borrell *et al.*,

2000a; Mahalakshmi and Bidinger, 2002). An increase in biomass yield of about 47% over and above that obtained from senescent genotypes has been reported in genotypes that express the stay-green trait under post-anthesis moisture deficit (Borrell *et al.*, 2000b). Lack of grain yield differences observed by these authors among genotypes grown under irrigated conditions, further suggests that the stay-green trait does not hamper yield under no moisture deficit conditions.

Stay-green improves resistance to diseases and lodging (Tenkouano *et al.*, 1993). In sorghum, genotypes with the stay-green trait continue to fill their grain generally under moisture stress conditions (Rosenow and Clark, 1981), exhibit improved resistance to charcoal rot (*Macrophomina phaseolina*) and induced lodging (Henzell *et al.*, 1984; Woodfin *et al.*, 1988). However, Tenkouano *et al.* (1993) reported that non-senescence and charcoal rot reaction are genetically independent.

1.8 Genetics of drought tolerance in sorghum

Several genes are involved in drought stress tolerance in various plant species. The function of these genes is either protecting the cell from water deficit by the production of important metabolic proteins or regulation of genes for signal transduction. The expression of a dehydrin, *dhn1* gene in sorghum as a response to water deficit was reported by Wood and Goldsbrough (1997). Expression and accumulation of *dhn1* gene in seedlings and pre-flowering sorghum was identical among genotypes, but genotypes showed variation in timing of expression of the gene. This suggested that the expression of dehydrins is possibly an important drought adaptation mechanism in sorghum.

The expression of genes related to water deficit in plants is found to be induced by water stress, desiccation, and abscisic acid (ABA). Yamaguchi-Shinozaki *et al.* (2002) also observed wide variation in the timing of induction and expression of drought related genes. The authors classified these genes into two groups; the first group are responsible for proteins which function directly in stress tolerance, and the second group give protein factors involved in the regulation of signal transduction and gene expression under drought. Most of these drought inducible gene expressions are induced by ABA. However, various researchers reported the existence of ABA-dependent, and ABA-independent signal transduction cascades between the initial signal of drought stress and the expression of the genes. Furthermore, Shinozaki and

Yamaguchi-Shinozaki (1997, 2000) suggested that at least two independent pathways exist in plants.

The purpose of studying the genetics of drought resistance in plants is to identify genetic factors that determine the productivity of crops under drought stress conditions. Advances in crop improvement under water-limited conditions are only possible, if drought resistance traits are identified and selected in addition to yield (Borrell *et al.*, 2000a; Sanchez *et al.*, 2002). The quantitative trait loci (QTLs) that have been mapped on the 10 linkage groups of sorghum so far are involved in controlling traits related to yield and yield components, root systems, stay-green, plant height, flowering, and maturity.

Tuinstra *et al.* (1997) identified 13 genomic regions associated with post-anthesis drought tolerance in sorghum. Four QTLs were identified for yield and yield stability, seven for duration of grain development and seed weight, and two for the stay-green trait. A number of traits related to drought resistance have been identified and mapped; however, the stay-green trait is recognized as the most crucial drought resistance trait in sorghum. There are three stay-green gene sources (B 35, SC 56 and E 36-1) from which all QTLs that have been mapped so far on 10 linkage groups on sorghum (Kebede *et al.*, 2001; Haussmann *et al.*, 2002).

Tao *et al.* (2000) identified two stay-green QTLs located on linkage group B and I. Likewise, Crasta *et al.* (1999) and Xu *et al.* (2000) identified four stay-green QTLs and mapped two of the QTLs (*Stg1* and *Stg2*) on linkage group A, and the other two, *Stg3* and *Stg4* on linkage group D and J, respectively. The stay-green QTLs were ranked based on their contribution to the stay-green phenotype as *Stg2*, *Stg1*, *Stg3*, and *Stg4* in their order of merit. Furthermore, Xu *et al.* (2000) also mapped three QTLs (*Chl1*, *Chl2* and *Chl3*) for chlorophyll content, and the map position coincides with the stay-green QTLs. The phenotypic association of the stay-green trait and chlorophyll content may be explained by the map position of these QTLs on the genome.

Many secondary factors, such as differences in flowering time, reproductive sink strength together with variation in the environmental factors alter the expression of the stay-green trait (Harris *et al.*, 2007; Tao *et al.*, 2000). Six maturity genes (Ma_1 - Ma_6) were identified, and mapped on the sorghum genome. The dominant form of these genes causes extreme lateness (Childs *et al.*, 1997) cited by Morgan *et al.* (2002). Two maturity QTLs were positioned near a stay-green QTL linkage group and the major independent maturity QTLs were highly correlated with stay-green rating (Subudhi *et al.*, 2000; Xu *et al.*, 2000). Tropical genotypes are found to be

dominant for all four loci (Ma_1 - Ma_4) that control the time of flowering (Quinby, 1974). However, substituting the dominant maturity gene, Ma_1 , to recessive ma_1 converts a tropical sorghum to temperate one that will flower in high latitude (Major *et al.*, 1990). Tuinstra *et al.* (1998) identified two QTLs that conditioned the expression of the stay-green trait. The physiological association of the maturity and stay-green traits are not well understood. The indistinct association between the two traits suggests that the earliness trait may work against reproductive sink strength during post-anthesis drought stress.

Walulu *et al.* (1994) indicated that the stay-green trait in sorghum is controlled by a major gene that exhibits different levels of dominant gene action depending on the environment. However, van Oosterom *et al.* (1996) further studied the stay-green trait as a function of GLAD which is affected by green leaf area at flowering, time of onset of senescence, and subsequent rate of senescence. It was reported that the three stay-green components appearred to be inherited independently. The inheritance of the onset of leaf senescence was additive and senescence rate was dominant. Consequently, GLAD was found to be partially dominant. Besides, the expression of these three factors is also affected by many environmental factors, and hence, the combined genetic effects of the three factors and the environmental factors should be considered when designing breeding programmes for drought resistance (Borrell *et al.*, 2000b; Mahalakshmi and Bidinger, 2002). Delayed senescence in sorghum, therefore, is a valuable trait in which it improves genotypes adaptation to drought stress, grain filling and grain yield under stress.

1.9 Breeding for drought resistance/tolerance

Generation and selection of new combinations of genes to produce genotypes with superior trait performance than that of the existing genotypes within the target environment is the major objective of plant breeding (Chapman *et al.*, 2003). In any breeding programme, defining the critical traits to improve grain yield under a given target environment is critical (Fernandez, 1992). Identification of important traits depends on the degree of influence of a trait on yield, expression of the trait at whole plant level, the nature of the target environment (rainfall amount, distribution, onset and cessation, available soil water, nutrient status of the soil, and diseases), and economic environment (the necessity of grain quality and quantity). In maize, for example, it was found that early flowering, crop water use efficiency and early vigour are important traits to improve yield under drought condition (Richards, 1996).

The higher flexibility of sorghum in adapting to diverse climatic conditions resulted in evolution of tropical and temperate sorghum varieties. The tropical varieties are characterized by being tall, late maturity, low harvest indices, photoperiod sensitivity and poor population performance. They are generally adapted to low population levels and exhibit low response for improved agricultural practices (fertilizer and mechanized harvesting). The temperate sorghum varieties, on the other hand, are characterized by dwarf stems, early maturity, and high yielding and produce less dry matter per plant (Rao *et al.*, 2002). In the early sorghum improvement programmes, conversions of tropical varieties to temperate variety were made by substituting two recessive alleles for their dominant height alleles in the tropical varieties and three recessive alleles for dominant maturity counterparts. The conversion programme started with hybridization of tropical and temperate varieties followed by successive backcrossing (Acquaah, 2007).

In breeding for drought tolerance pure line selection method has been used in many national and regional sorghum improvement research programmes in Africa and Asia (Acquaah, 2007). However, pedigree and bulk selection methods are commonly used in most international and national breeding institutions. Pedigree selection in segregating populations derived from planned crosses is the dominant breeding strategy to develop pure line varieties and hybrid parents in sorghum (Dar *et al.*, 2006). If the transfer of few traits relating to drought resistance to a high yielding cultivar is required, backcrossing is the appropriate breeding method (Mitra, 2001).

Exploitation of heterosis has become part of the routine in most sorghum breeding programmes after the discovery of stable and heritable cytoplasm-nuclear male sterility systems in the crop. This discovery further enables large-scale commercial hybrid seed production feasible (Dar *et al.*, 2006). The study of the expression of hybrid vigour in grain sorghum revealed that 84% increase in number of seed per plant, 82% in grain weight, and 12% stover weight were observed in hybrids over the better parent (Doggett, 1988).

In the past drought resistance screening was done under optimal conditions, because the maximum genetic potential of yield can only be realised under optimum conditions. Additionally, it is believed that high positive correlation exists between performance in optimum and stress conditions (Tuinstra *et al.*, 1997; Habyarimana *et al.*, 2004). However, the high genotype by environment interaction may restrict the expression of the yield potential under drought condition (Chapman *et al.*, 2000 a and b). Although there is a yield penalty when selecting

plants under drought condition in contrast to optimal environmental conditions, Richards (1996) and Tuinstra *et al.* (1997) suggested that selection under both optimal and drought conditions represents the ideal condition to select for yield and yield stability, drought tolerance and expression of drought related traits. Hence, drought resistance and its impact on yield involve interaction between plant water relations and plant physiological functions. The interactions are further complicated by the frequency and duration of drought, plant development stage and other stress factors such as low soil fertility and biotic stress factors.

1.10 Conclusion

Drought is a normal recurrent feature of climate that occurs in virtually all climatic zones. Its intensity varies significantly with climatic factors (rainfall, temperature, and radiation) and other demographic factors (topography, altitude, and soil factors) (Thomas, 1997). Drought resistance is a complex quantitative trait affected by many genetic and environmental factors. It involves the interaction of different morphological, physiological and biochemical functions (Mitra, 2001). Moreover, the responses of different plants, species, and genotypes to drought are variable in relation to developmental stage, evolutionary adaptation of the crop, intensity and duration of drought (HongBo *et al.*, 2006). Understanding the genetic and physiological basis of drought resistance in plants is the first priority when breeding for drought resistance.

Lin et al. (1995) and Kouressy et al. (2008b) recognized the importance of reduced plant size in terms of small and narrow leave structure and genetic dwarfing of the plant for drought resistance. Reduction of leaf area index, through reducing the number of leaf and narrowing the leaf structure, resulted in the reduction of the effective photosynthesis area. This, in turn, reduced the amount of assimilates produced and the grain yield. However, this reduces the amount of water loss through transpiration. Genetic dwarfing, on the other hand, increase efficiency of plants in balancing assimilates translocation between the developing grain and other vegetative organs (Kouressy et al., 2008b).

An early maturing genotype yields less compared to a long maturing genotype under favourable environment. This is because drought escape by shortening the growing period is made at the expense of the crops potential yield. As a result Blum *et al.* (1989) suggested that short duration may not be an exclusive selection criterion *per se* since other factors have also been involved in affecting genotype performance under water stress. Stay-green (delayed senescence), however, is a valuable trait in which it improves genotype adaptation to drought stress, grain

filling and grain yield under stress (Borrell *et al.*, 2000b; Mahalakshmi and Bidinger, 2002). No grain yield difference was observed among genotypes grown under stressed and irrigated conditions, suggesting that stay-green trait does not hamper yield under moisture deficit conditions as compared to osmotic adjustment and early maturity (Borrell *et al.*, 2000a). The balance among these characters maintains adequate productivity by providing effective resistance mechanism.

Many researchers have proposed various characteristics related to drought resistance that could be used in selection and breeding programmes. However, comprehensive understanding of the physiological and genetic basis of adaptation in moisture stress condition is still lacking. Moreover, the interaction between the different characteristics within a plant and the environment makes drought resistance breeding very complicated. Hence, the chance of obtaining a drought resistant cultivar with all the important characters is low. The choice of specific traits as selection criteria depend on the crop species, the heritability, and ease of transfer of the traits. Moreover, since drought resistance is the interaction of different morphological, physiological, and biochemical traits, a combination of different resistance traits, rather than a single trait, should be used as a selection criteria.

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CHAPTER 2

Analysis of the Sorghum Production Systems in the North Eastern Ethiopia: Breeding Priorities and Implications on Varietal Adoption

2.1 Abstract

Sorghum is one of the most important crops worldwide after wheat, rice, maize, and barley. Examining the present socio-economic conditions of sorghum producing farmers will have a paramount importance in designing possible improvement strategies based on farmers' priorities. The objectives of this study were to evaluate the sorghum production system and pattern, major production constraints and coping mechanisms, varietal diversification, farmers' criteria for choosing varieties over seasons, and adoption of improved varieties, the effects of moisture stress and coping strategies. Purposive sampling was used to identify three Administrative Zones, 6 Districts and 12 Peasant Associations. Random sampling was then applied to sample 171 households. Group discussions, individual interviews and review of secondary dada were employed for data collection. The results suggested that the performance of sorghum is generally poor which is mainly accounted for by moisture stress, pests, diseases, weeds, farmland fragmentation due to demographic pressure, poor soil fertility, and poor performance of the local varieties. Among the constraints, drought at grain filling stage was identified by farmers as the most important production problem in the target region. The productivity of sorghum was also largely hindered by the use of inherently poor yielding local landraces as farmers were forced to abandon their high yielding, big-headed and late maturing sorghum varieties due to the prevalence of recurrent moisture stress. Although 50% of the interviewed farmers had information about improved varieties of sorghum, more than 91% of the respondents grew local varieties. The importance of addressing the missing link in the researchextension-farmer linkages for better diffusion and impact of improved varieties cannot be underestimated. However, despite the greater availability of improved sorghum varieties from the research system they have not yet been adequately transferred and adopted by farmers. To benefit the most from the available improved technologies, farmers have to be part of the breeding process right from the very beginning. Emphasis should be given to the conservation and utilization of farmers' varieties which have evolved and adapted over the years to local agro-climatic conditions. Therefore, assessing the level of genetic variation maintained by traditional farmers should enhance efficient utilization of genetic resources.

Keywords: genetic diversity, moisture stress, participatory rural appraisal, sorghum

2.2 Introduction

Globally sorghum [Sorghum bicolor (L.) Moench] is the fifth most important cereal crop after wheat, rice, maize, and barley (FAO, 2006). In Ethiopia, sorghum is the main staple food crop, ranking second after maize in total production. It ranks third after wheat and maize in productivity per hectare, and after tef and maize in area cultivated (CSA, 2011). It is grown in almost all regions covering a total land area of 1.9 million ha (CSA, 2011). The major sorghum production areas of the country include Oromia at 39.1%, Amhara (37.6%), Tigray (11.5%), and Southern Nations Nationalities and Peoples (SNNP) (5.7%). In 2010/11 cropping season, 82% (9.7 million ha) of the total land area under field crop cultivation was allocated for cereals, of which sorghum accounted for 20% (CSA, 2011).

The Amhara National Regional State (ANRS) of Ethiopia occupies much of North Western and North Eastern region of Ethiopia. The total area is about 0.16 million km² constituting about 20.9% of the total area of the country (CSA, 2011). Agriculture is the major source of food for the people and also raw materials for local industries and export earnings. Thus, agriculture is the mainstay economic sector in the region. The total cultivated land covered with different crops, in the Amhara region in the year 2007 was about 3.7 million ha. However; this figure showed a 10.5% increase in 2011 (CSA, 2011). In the agriculture sector, crop production and animal husbandry are the major activities undertaken in the region. With regard to crop production, cereals, pulses, oil crops, fibre crops, fruits and vegetables are grown in different parts of the region. Crop production in the region is dominated by cereals which include tef, sorghum, wheat, maize and barley (Figure 2.2.1). Sorghum ranks first (constituting 32 and 46% of the total area and production allotted for cereals, respectively) in the region followed by tef (29 and 22% in the same order) (CSA, 2011).

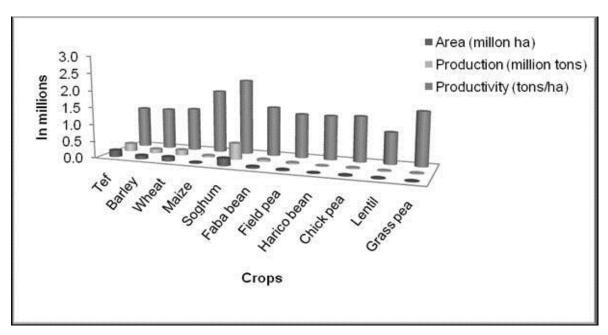


Figure 2.2.1 Cropped area, production and productivity of major crops in the main season in the three Zones of the Amahara Region (CSA, 2011)

The State's diverse biophysical and socio-cultural environments have resulted in a great diversity of plant species. The variability in environmental factors together with farmers' active selection, contributes to the extensive level of sorghum diversity in the Region (Gebrekidan, 1973; Ayana and Bekele, 2000). The presence of many landraces and wild relatives of sorghum make the region a primary/secondary centre of diversity (Flora of Ethiopia and Eritrea, 1999). Sorghum shows high morphological and genetic diversity for several traits. These traits include drought tolerance, disease and pest resistance, grain quality, grain size, plant height etc. However, this huge potential of the plant genetic resource base has not yet been explored and utilized to improve agricultural production and productivity. Proper assessment and documentation of farmers' knowledge of sorghum varieties, sources of seed, specific characteristics and use is vital for wise utilization and conservation of available sorghum genetic resources.

Wortmann *et al.* (2006) reported that drought, low soil fertility (nutrient deficiency), stem borers, shoot fly, quelea birds, *Striga*, and weeds are recognised as major production constraints in Eastern Africa. Although these constraints cause a significant grain yield loss, the relative importance varies from region to region, within and among the countries. For example, shoot fly is reported to cause significant grain loss in Ethiopia and Uganda, but is of relatively less

importance in Mozambique. Similarly, *Striga* has been cited to be the most important constraint in Kenya, and the second most important in Ethiopia and Uganda. In Ethiopia, drought and *Striga* were found to be the most important in North and North Eastern parts of the country whereas quelea birds were seen as the major constraints in the Rift Valley and Southwest lowlands (Wortmann *et al.*, 2006). Research has also shown that moisture deficit during grain filling is the most important for Ethiopia and Mozambique (Simane *et al.*, 1998; Wortmann *et al.*, 2006; Mengistu, 2009). It is worth noting that drought and *Striga* are equally important production constraints in the target region although priority has been given to the former for the purpose of this study.

In the North Eastern Ethiopia, the traditional farming practice heavily relies on rainfed crop production system which is characterized by poor crop performance and low yield. The major factors responsible for poor crop performance and reduced yield include moisture stress, low soil fertility, parasitic weed (*Striga hermonthica*), and lack of improved and suitable production technologies. The most important factor influencing the production and productivity of crops in the region is rainfall (Tadesse and Tesfahun, 2001). Rainfall is insufficient in amount, uneven in its distribution and unpredictable in its inception, especially in the lowlands (EARO, 2001). In almost all lowland areas, crops are prone to periodic moisture stress. Overall, the rainfall patterns, frequency of moisture stress and farmers' understanding of moisture stress and indigenous cultural practices used by farmers to cope with moisture stress has a paramount importance for designing future moisture stress coping strategies.

Drought is one of the major challenges affecting crop production worldwide. Climate changes will increase the frequency of droughts, particularly in many countries in Africa. For instance, by 2050, water shortages are expected to affect 67% of the world's population (Ceccarelli *et al.*, 2004). Drought can occur at any stages of the crop development. However, in arid and semi-arid tropics, the probability of drought is highest at the start and end of the growing season. Drought stress at the beginning of the growing season will severely affect plant establishment. If drought occurs at the seedling, flowering, or grain filling stages, it may result in reduced establishment, reduced yield, or complete crop failure (Tumwesigye and Musiitwa, 2002). Assessment of sorghum growth stages where moisture stress is more serious, varietal choice of farmers across seasons and the response sorghum varieties to moisture stress has a significant impact in designing future breeding programmes.

Farmers' technology/variety preferences, socio-economic constraints and agro-ecological factors play important role to their technology adoption, productivity and production. This applies well to sorghum technologies that have been developed so far and future technology generation programmes. Therefore, assessing the present socio-economic conditions of sorghum producing farmers will have a paramount importance in designing possible improvement strategies based on farmers' priorities. Therefore, this study was carried out to assess sorghum production system and pattern, to identify farmers' major sorghum production constraints (such as moisture stress) and coping mechanisms, to evaluate varietal diversification, farmers' criteria for choosing varieties over seasons, technology generation and adoption of improved varieties and the role of different stakeholders in technology promotion and dissemination.

2.3 Material and methods

2.3.1 Description of the study area

The study was conducted in the Amhara National Regional State (ANRS) in the North-Eastern region of the Federal Democratic Republic of Ethiopia. This region is well known for its proneness to recurrent drought. The study encompasses three Administrative Zones¹ that stretch from Waghemra in the north to Kalu in the south. This area is known as tef-sorghum production belt and it is one of the micro-centres of sorghum diversity in Ethiopia (Gebrekidan, 1979; Benor *et al.*, 1999).

The study covered a wide geographic area and with varying farming systems. The rainfall pattern is most often bi-modal and sometimes mono-modal. Table 2.3.1 presents the total population, ranges of annual rainfall and temperature of the surveyed districts. Annual rainfall averages ranged from 550 mm (Sekota) to 1250 mm (Tehulederie). The rainfall amount and pattern are modified by altitude; areas in higher elevation are receiving relatively more rainfall than at the lower elevations. Temperature varies between 7°C to 32°C. The annual average rainfall in the Districts ranged from 683 to 935 mm, the maximum being recorded from

¹ A zone is an administrative unit below a Regional State (or Province) and it is composed of a number of Districts. A District (locally called "*Wereda*") is an administrative level which is composed of a number of Kebeles (15 to 45 Kebeles in a Wereda). A "Kebele" (or a Peasant Association in the rural setup) is the smallest unit of administrative structure and it often has 500 to 1500 households.

Tehulederie (1250 mm) and the minimum from Kobo and Sekota (500 mm). The yearly average temperature ranges between 17 – 31°C. From the above, one can assume that the study areas receive sufficient amount of rainfall to support crop production. However, the precarious rainfall, usually manifested by heavy down-pours and frequent dry spells, makes rainfed agriculture a risky venture. In some areas like Sekota and Kobo, the effective rainy days in a given growing season are less than 15. Consequently, in almost all lowland areas, crops are prone to periodic moisture stress. In terms of total population, Kalu District is highly populated while Sekota is sparse. However, pertaining to population density, Tehulederie is highly populated (319 persons per Km²) followed by Kalu (309) and Gubalafto (212) (Table 2.3.1).

The average agricultural density of the study area, considering both crop and grazing lands regardless of terrain, is 203 persons per Km². This shows that there is high population density which has a negative impact on agricultural development due to high subsistence requirement and limited source of off-farm earnings. The highest percentage of cultivated land is found in Tehulederie (52%) followed by Habru (23%), with the average of 33% for all the Districts (Table 2.3.1). The farming community practices a mixed farming system (crop-livestock) and grows tef and sorghum as the main crop enterprises.

Table 2.3.1 Total area, population, annual rainfall and temperature ranges of the Districts

District	No.	Total	No.	Total area	Cultivated	% of	Annual	Temp.
	PAs	population	people/	(km²)	land (km²)	cultivated	rainfall	range
			km²			land	(mm)	(°C)
Kalu	34	304,336	309.4	983.69	281.60	28.63	750 - 900	25 - 35
Tehulederie	19	129,173	318.7	405.37	212.57	52.44	1000 - 1250	10 - 32
Habru	30	210,830	170.1	1239.79	286.74	23.13	750 - 1000	15 - 29
Gubalafto	34	208,004	212.4	979.35	361.73	36.94	600 - 1050	15 - 25
Kobo	40	244,046	121.9	2001.57	595.37	29.75	500 - 800	20 - 33
Sekota	33	148,489	86.2	1722.43	422.48	24.53	500 - 600	17 - 29

Source: District office of Agriculture and Rural Development (DOARD) and CSA (2011)

2.3.2 Sampling and data collection procedures

Table 2.3.2 shows the selected study sites and the number of farmers who participated in this study. The study was carried out in three administrative sorghum growing Zones from which six districts were chosen i.e. Waghemra (Sekota district), North Welo (Kobo, Gubalafto and Habru)

and South Welo (Tehulederie and Kalu). Purposive sampling procedure was followed to select Zones and Woredas based on the relative importance of sorghum in the area. A total of 12 Peasant Associations (PAs) (2 from each district) were selected from low (<1600 masl) and midaltitude (1600-1900 masl) agro-ecologies based on the relative importance of sorghum in the area. Sampled farmers (9-20 from each PA) were randomly selected from PA household list. Random sampling was used to avoid systematic biases with regard to the distribution of the population sampled. The interviews were held with a total of 171 farmers and of which 29 (17%) of the participants were women. At the beginning, the plan was to at least increase the woman participants to 30%, however, due to many socio-economic problems; the majority of women either rented or shared out their plots. As a result, they could not respond to most of the questions in the questionnaire.

Both secondary and primary data were collected for the study. Secondary data were collected mainly from Zonal and District Agriculture offices and agricultural research centers. Primary data were generated from the sampled PAs and households using structured interviews. The data collection instrument had questions related to demographic features, access to land, input use and crop production, capital assets, sorghum diversity as defined by farmers, crop production constraints and coping mechanisms. The farmers were asked to list sorghum varieties they grew, their seed sources, and specific characteristics of the varieties they cultivated. Focus group discussions were held with about 15 - 20 farmers in each PA. The discussion was focused on topics related to genetic diversity, sorghum trait preferences, main sorghum production constraints, and coping mechanisms.

Both primary and secondary data were analysed using SPSS (SPSS, 2005). Descriptive statistics were used to summarize sorghum production constraints, the existing genetic diversity and its specific characteristics and use, farmers' selection criteria, moisture stress and coping mechanisms. Significance tests on household characteristics were conducted based on standard error derived using the formula SD = $\sqrt{\frac{STD_1^2}{N_1} + \frac{STD_2^2}{N_2}}$. Matrix ranking was employed to select the most important crops across Districts.

Table 2.3.2 Study sites and number of participant farmers

Zones	District	PAs	Agroecology	No. of farmers
South Wello	Kalu	Wodajo	Low-altitude	12
		Chorisa	Mid-altitude	14
	Tehulederie	Seglele	Low-altitude	13
		Gobeya	Mid-altitude	13
North Wello	Habru	Girana	Low-altitude	15
		Humo	Mid-altitude	20
	Gubalafto	Hara	Low-altitude	15
		Gedober	Mid-altitude	15
	Kobo	Jarota	Low-altitude	15
		Gobye	Mid-altitude	15
Wag-Hemera	Sekota	Debre Birhan	Low-altitude	9
		Deble Mariam	Mid-altitude	15

2.4 Results and Discussion

2.4.1 Socio economic conditions

In the surveyed areas, livelihood was mainly dependent on agriculture. Farmers who owned farmland were privileged, in relative terms, than those who did not have land. Land redistribution was made in 1991 based on criteria like family size and fertility status of the land. Due to population pressure and new claimants, most farmers possessed fragmented pieces, generally ranging between one to five plots of land at different localities. According to the survey data, the average land size per household was 1 hectare (Table 2.4.1), which was larger than the South and North Wello average land holding of 0.81 ha per household (CSA, 2011). Not only was land miniaturization a challenge that farmers faced but also land fragmentation was a very crucial land tenure issue. Farmers often rented or shared land to address the land shortage. Land shared-in is gaining access to land in return for paying the owner a percentage of the production, whereas rented-in is gaining access to land by paying rent to the owner. Although it was not statistically significant, the average amount of land rented-in was higher (0.56 ha) in the midlands than in the lowlands (0.53 ha). However, the average land shared-in within the lowland was much higher (0.87 ha) than the midlands (0.67 ha). Oxen were the major source of draught power for crop production. Most of the women and those farmers who did not have a pair of

oxen were forced to share out their plots. According to the survey data, plots which were rented out were mainly irrigable.

Table 2.4.1 Land holding in the surveyed areas

Land holding	All	Low altitude	Mid-altitude	Difference	T - value
	(N=171)	(N=79)	(N=92)	in means	
	Mean (SD)	Mean (SD)	Mean (SD)	Mean (SD)	-
Total land owned	1.00 (0.65)	0.97 (0.62)	1.04 (0.68)	-0.07(1.57)	-0.69
Land rented-in	0.54 (0.25)	0.53 (0.28)	0.56 (0.24)	-0.04(0.03)	-0.21
Land shared-in	0.89 (0.85)	0.87 (0.60)	0.67 (0.39)	-0.04(0.20)	-0.19
Land shared-out	0.67 (0.39)	0.75(0.45)	0.64 (0.39)	0.11(0.09)	0.38
Land rented-out	0.01 (0.01)	0.02 (0.01)	0.01 (0.01)	0.01 (0.02)	0.40

Note: Figures in parentheses are standard deviations

The area and yield estimates were determined by first, assessing the total land area (includes both land owned, rented-in and shared-in) planted by the respondent farmer in 2009/2010 cropping season; second, dissecting the land by the type of crop grown and the area allocated for each crop, then assessing the total amount of harvest for each crop (this includes intercropping). The average area of land allocated per household for sorghum in 2009/10 was 0.87 hectares which shows how important this crop is to the farming system studied. Having allocated such significant amount of land, farmers could harvest an average yield of only 0.82 t/ha with the average productivity being 1.1 t/ha. This figure is significantly low compared to the national and regional average yield of 2.1 t/ha and 2.2 t/ha, respectively (CSA, 2011). The data in this study is only for one production season and comparing production and productivity temporally would require more time series data.

The average family size in the mid altitude areas was 5.6 which is significantly higher than in the lowlands (5.1) (Table 2.4.2). For the rural communities, access to basic infrastructures such as schools, health centers, roads, markets and extension centers remains a challenge. Farmers in the mid altitude areas had better access to transportation than those in the lowlands. However, the farmers in the lowlands had more market access than farmers in the mid altitudes. Farmers had equal access to extension services in all areas (Table 2.4.2).

Table 2.4.2 Household characteristics and access to different facilities

	All	Low altitude	Mid-altitude	Difference	T -
	(N=171)	(N=79)	(N=92)	in means	value
	Mean(SD)	Mean(SD)	Mean(SD)	Mean(SD)	<u>-</u>
Characteristics					
Age of household head (years)	42.1 (11.1)	42.2 (11.5)	42.0 (10.9)	0.18 (1.6)	0.12
Family size (number)	5.4 (1.8)	5.1 (1.7)	5.6 (1.8)	-0.5 (0.8)	-1.78*
Distance to all weather road (hrs)	47.2 (53.1)	55.3 (59.1)	40.2 (47.0)	15.0 (0.03)	1.82**
Distance to market (hrs)	76.9 (56.2)	52.5 (54.7)	97.9 (48.8)	-5.7 (0.2)	-5.7**
Distance to DA center (hrs)	28.0 (30.2)	25.6 (22.8)	30.2 (35.3)	-4.6 (0.09)	-1.03

Notes: Distance is measured in walking minutes and DA is the development agent located at the extension center. Figures in parentheses represent standard deviations. * and ** denote significant differences at the 5 and 10% probability levels, respectively.

2.4.1.1 Food availability at household level

Sorghum in Wello is harvested in November (37% households) and first half of December (63%). The majority of farmers reported that they experienced severe food shortage in the months of August, September and October. The severity of food shortage during the year is determined by the number of months that the households experience food shortages. In the lowlands, the food insecurity was higher than the midlands. This may be explained by the fact that apart from insufficient rainfall, lowland areas are highly affected by extremely depleted soil fertility, land degradation, deforestation, plant and animal pests and diseases. The food shortage was beyond three months in each district. However, in Sekota, the insecurity level increased to six months. In all the districts, food was available from December to May. The number of months of plenty of food depends on the family size and the attitude of farmers towards saving and managing to survive with little food. In Ethiopia, in general and in Wello specifically, farmers have many cultural and religious functions such as "Zikir", "Senbetie", "Sedeka", "Serg", etc. These are the social activities that lead to extravagant consumption of the available food. The government extension programmes have made some positive contribution in changing farmers' attitudes towards saving and wise use of available food resources. Only 11% of the respondent farmers achieved food security at the household level (Figure 2.4.1).

Although there were some encouraging improvements in food security overall in the region, a lot still remains to be done in this area.

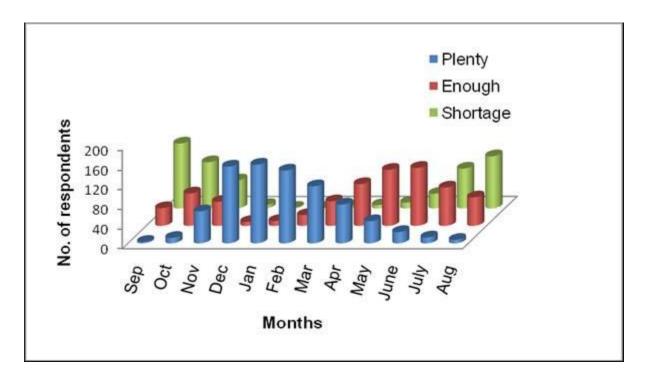


Figure 2.4.1 Availability of food at the household level during the months of the year

2.4.1.2 Farmers' coping strategies during months of food shortage

Respondents mentioned that factors such as moisture stress, low soil fertility, insect pests, and shortage of farmland coupled with burgeoning human population, were the prime causes of food shortage. Farmers are well experienced with uncertainties and risks with regard to food shortage and they have developed dynamic and evolving coping mechanisms. The different coping strategies employed by the farmers to grapple food shortage are shown in Table 2.4.3. Households relied on selling animals, getting credit, off-farm wage employment, migration, food aid, selling horticultural crops, petty trading, safety net programmes and remittances. Although crop production was the main occupation of the farmers in all the districts studied, a substantial proportion (92%) can be categorized as mixed farmers who practised both crop and livestock production. Livestock, especially small ruminants and poultry production was practiced as a food security strategy. Although type and quantity of livestock ownership varied among Districts,

about 37% of the farmers mentioned that they were keeping animals for selling them to supplement their food needs during the times of food shortages.

In areas where irrigation facilities were available, production and marketing of horticultural crops was an important source of income for 10% of farmers in the lowlands and 16% in the midlands. Additionally, off-farm activities were mentioned as the main sources of supplementary income for 18% of farmers in lowland areas compared to 23% in midlands. Remittance (sent from non-resident family members and relatives) was a more important coping strategy for farmers in the lowlands. Farmers in the mid-altitudes benefited more from the safety net programmes than those from the lowlands. Safety net programme is a component of the government's food security strategy, in which the rural communities in the food insecure areas have been provided with either food or cash to lessen short-term consumption shortage and encourage households to engage in production and investment activities. About 25% of the respondents were generating income by means of petty trading.

Table 2.4.3 Farmers' coping strategies during months of food shortage

Strategy	All (N = 171)	Low-altitude (N = 79)	Mid-altitude (N = 92)
Selling of animals	73 (24.5)	29 (22.1)	44 (26.3)
Credit	66 (22.1)	30 (22.9)	36 (21.6)
Off-farm wage employment	41 (13.8)	18 (13.7)	23 (13.8)
Migration	28 (9.4)	14 (10.7)	14 (8.4)
Food aid	28 (9.4)	14 (10.7)	14 (8.4)
Selling horticultural crops	26 (8.7)	10 (7.6)	16 (9.6)
Petty trading	25 (8.4)	11 (8.4)	14 (8.4)
Safety net	9 (3.0)	3 (2.3)	6 (3.6)
Remittance	2 (0.7)	2 (1.5)	0 (0.0)

Note: The sum of the frequencies does not add to the total number of sampled farmers because some farmers have given multiple strategies and the strategies have been grouped. Figures in parentheses represent percentages.

2.4.2 Sorghum production and variety use

2.4.2.1 Sorghum production trends

Farmers were asked to describe the trend in sorghum production in the past five years. According to 30% of the farmers sorghum cultivation showed an increasing trend over the years. Sorghum was the most sought after crop because of its tolerance to moisture stress, relatively high yield and suitability for livestock feed (78%). However, 22% of the respondents were of the opinion that the increase in the cultivation of sorghum was rather due to area expansion into grazing and marginal lands in response to population pressure.

Conversely, 71% of the farmers thought that sorghum production had been decreasing because of the climate i.e. erratic rainfall, delayed onset and early cessation and sometimes the rain falling outside the growing season; poor soil fertility, *Striga* and stalk borer infestations. They further justified that in areas where small irrigation schemes were established, farmers shifted from growing sorghum to more market oriented crops. In addition, during rainy seasons flooded rivers over-flowed into the farmlands that turned into waste land further reducing sorghum production. According to 49% of the farmers, the area under sorghum had remained relatively unchanged and crop rotation was being practiced. Farmers said in those areas only tef and sorghum are the only crops grown.

2.4.2.2 The relative importance of sorghum

In ARNS, sorghum is the second most important food crop after tef. It accounts for 22% of the area covered under cereals and 27% of the total cereal production (CSA, 2011). To get a good grasp of the relative importance of different crops in the study area, farmers were asked about the crops they had cultivated in 2009/10. The sampled farmers on average cultivated 2.4 crops in the year 2009/10. Crop diversification in the mid-altitudes was higher (3 crops) than the low altitudes (2 crops). The mid-altitudes faced relatively less moisture stress and the agro-ecology allowed farmers to grow more crops such as wheat, barley, faba bean, field pea and lentil than the low altitudes. Accordingly, sorghum and tef were predominantly grown in both altitudinal zones (mentioned by 41% and 28% of the farmers). Maize, chickpea, barley, wheat, horticultural crops, faba bean, finger millet and linseed were the other crops mentioned in order of their importance. Barley and wheat were also important in Sekota and Tehulederie, respectively. In areas where irrigation facilities were available (Kobo, Gubalafto and Habru), maize and horticultural crops such as tomato, pepper and onion were grown (Table 2.4.4). Barley and

haricot bean were typically important in Kobo and Habru, respectively. Maize and haricot bean were important in Kobo as wheat and faba bean were in Tehulederie.

About 98, 97, 68, 43% of the respondents used sorghum for home consumption, animal feed (the stalk), cash generation, and crop rotation, respectively. According to 97% of the farmers interviewed, sorghum was best suited to the prevailing cropping system and adapted to the climate. Sorghum was produced for various purposes. The grain was used for preparing "injera" (the staple food), brewing "tella" (local beer), baking bread, making porridge, preparing "kollo" (roasted grain), and "nifro" (boiled grain). Farmers have popping sorghum varieties which were used instead of popcorn during cultural functions. The majority of the farmers (96.5%) used the stalk from sorghum for various purposes: livestock feed, fuel wood, construction material, sale, and payment in-kind. The stalk of sweet stem varieties was chewed.

Table 2.4.4 The ranking of major crops in the sampled areas

			S	cores			Mean
Crops	Sekota	Kobo	Gubalafto	Habru	Tehulederie	Kalu	Score
Sorghum	5.0	5.0	5.0	5.0	4.5	5.0	4.9
Tef	4.3	5.0	4.8	4.5	5.0	4.0	4.6
Faba bean	2.2	3.0	3.0	2.4	4.0	2.5	2.9
Horticultural crops	1.4	4.0	3.5	3.9	2.1	2.5	2.9
Maize	2.3	4.0	2.9	2.8	3.3	2.0	2.9
Wheat	2.8	2.1	2.0	2.3	4.0	3.0	2.7
Barley	4.1	2.3	1.9	2.7	2.3	2.0	2.6
Chick pea	2.0	2.0	3.0	2.7	2.0	3.0	2.5
Oil crops	2.9	2.4	2.3	3.0	2.0	2.3	2.5
Finger millet	2.2	2.7	2.6	3.0	2.1	1.9	2.4
Field pea	2.7	2.7	1.9	2.3	2.0	2.1	2.3

Note: The mean scores are computed per site and for all sites. Farmers rank each crop on 1-5 scale, 5=very important; 4=important; 3=intermediate; 2=less important; 1 = not important.

Most farmers (81%) grew sorghum as a sole crop and a small minority intercropped it with sesame, safflower, haricot bean and cowpea. Farmers indicated that crop choice for intercropping and crop rotation was made considering either the root system or nitrogen fixing

ability of the other crop. Sesame and safflower have short root systems and do not compete with the deep rooted sorghum so that these crops extract nutrients from different soil niches. However, farmers grew sorghum with pulse crops for their nitrogen fixing ability to enhance soil fertility. It was also mentioned that they used tef, wheat, barley and a number of pulse crops for crop rotation purpose owing to the above mentioned reasons.

2.4.2.3 Sources of seeds and seed processing

In the study areas, markets, farmers' cooperatives, local seed producers, and Agriculture Departments were identified as the major sources of seed. However, 69% of seed for planting was obtained by reserving own seed from the previous harvests and 55% from the market (Table 2.4.5). About 70% of the respondents from the mid-altitudes and 69% from the lowlands reserved seed from their own produce. However, more farmers in the lowlands (23%) purchased seed from markets than in the midlands areas (17%). This could be due to relatively higher incidences of crop failer in the lowlands than in the midlands.

Farmers had access to seeds of some of the improved sorghum varieties through extension programmes of the Agriculture offices. On the other hand, if the farmers wanted to plant a new sorghum variety, seed could be obtained through exchange among farmers. The farmers indicated that the quality of seeds obtained from the market and cooperatives was inferior whereas the seeds from government extension programme and research centers were superior in quality. The seeds provided by the research or government extension programme are basic or pre-basic seed which is uniform and best in quality. Moreover, farmers indicated that seed exchange among farmers could be carried out either based on close relationships (neighbors or relatives) or randomly from any seed producer (farmer to farmer seed exchange), as the seed quality varied accordingly. Farmers exercised growing of a mixture of varieties following pragmatic method by considering different maturity groups, different uses and market value to have good harvests. The ratio of each varietal mixture varies from year to year depending on constraint encountered and the reaction of the varieties to the bottlenecks. Well-matured, large seeded, big and healthy panicles are selected in the field and harvested first. The intact panicles are tied together and hanged up on a wall for 2 - 3 weeks till the seeds are dried to optimal moisture content. Then they were threshed and seeds stored in a covered pot with a preservative agent against weevil attack. This is done either by mixing the seeds with ash, or powdered pepper or by mixing with tef seed or applying Actellic or Sevin. All the cultural practices hindered the movement and reproduction of weevils in the storage device and they do

not have any negative impact on the quality of the seeds. However, the efficacy of controlling the weevil damage is relatively lower than the chemical pesticide.

Table 2.4.5 Seed sources of farmers

Source	All (N = 171)	Low-altitude (N = 79)	Mid-altitude (N = 92)
Own saved	189 (68.7)	83 (66.9)	106 (70.2)
Market	55 (20.0)	29 (23.4)	26 (17.2)
Farmer-to-farmer seed	17 (6.2)	6 (4.8)	11 (7.3)
exchange			
Neighbor farmer/relatives	7 (2.5)	2 (1.6)	5 (3.3)
Government extension	7 (2.5)	4 (3.2)	3 (2.0)

Figures in parentheses represent percentages.

2.4.2.4 Diversification of sorghum varieties

Sorghum is one of the most diverse crops distributed over a wide range of agro-ecologies in Ethiopia. It is produced mainly in the lowlands and to a considerable extent in the mid-altitude parts of Wello. Only low and mid altitudes are considered in this study because most of the diversities are found in these altitudinal zones. "Zengada", red-grained, long stalked and late maturing sorghum variety was the most widely grown and well established in the highlands. About 93% of the respondents predominantly grew local varieties while only 7% grew improved varieties, often in addition to the local varieties. In this study, it was found that farmers maintained a number of varieties (1 – 5) in a given farm at a time based on their different needs. However, Teshome *et al.* (2007) reported that farmers in South Welo grew over 24 sorghum landraces in a single farm. In this study, the level of sorghum diversity was found to be relatively low. This is because, unlike the study by Teshome *et al.* (2007) that took plant samples on a hectare basis that enabled them to identify more landrace diversity, the present study was conducted mainly by asking farmers to list and identify the varieties they grew by their specific characteristics and they probably considered only those varieties which grew in relatively higher proportions in their fields.

The number of uses to which the crop is put was an important factor affecting the nature and types of varieties that farmers used. This, in turn, affects crop diversity. Since farmers could not get all the various desirable features of varieties from a single variety, farmers using sorghum for many purposes ended up growing many varieties to benefit from the features of the different

varieties i.e. they needed to diversify. In this case, farmers on average grew 1.62 varieties of sorghum. Some varieties were grown for their stems (for chewing especially the sweet stems and construction materials), others were grown for their good brewing quality and yet others were grown for their food quality for "injera" and good biomass for animal feed. This practice allowed farmers to have harvest security in time of unpredictable environmental stress and derive multiple nutritional values (Teshome *et al.*, 2007). However, there were also very few sorghum varieties which were used for multiple uses.

In the low and mid-altitudes of the six districts of Wello, where the survey was undertaken, a total of 70 (11 improved and 59 landraces) sorghum cultivars, which are agro-morphologically distinct, were identified by farmers. The varieties as described by the farmers did vary in a number of morphological and socio-economic characters such as grain colour, head morphology, plant height, growth cycle, earliness, drought resistance, total plant biomass, market value and grain yield. Farmers in different localities often name the same variety differently. To avoid multiple counting of the varieties, experts and Development Agents (DAs) were consulted to rectify and ensure that the varieties were really different. However, there was still confusion in identifying the improved varieties since farmers either named the same variety differently or different varieties got the same name.

Out of the 70 varieties identified, only 33 (47%) varieties (8 from Sekota, 4 = Kobo, 5 = Gubalafto, 9 = Habru, 12 = Tehulederie and 11 = Kalu) were planted in 2009/2010 growing season (Table 2.4.6). Only four varieties (Degalet, Jamiyo, Jigurty and Zengada) were found to be the most common varieties in Wello. These landraces can be considered as common and widely adapted to diverse agro-ecological and socio-economic conditions of the farming communities. However, most of the landraces were unique to a specific locality and this may be the result of natural and artificial selection within locality. Improved varieties which were recently released and popularized, such as Birhan, Gobye, Abshir, Teshale, and Girana-1 were known by their given names. However, varieties which were released a long time ago had been given "local" names by the farmers. For example, 76 T, #23 was named as Wediakir. Similarly, Malkamu, Esmaeal, Ajaebe, Red America and Subihan were the different names given for a number of the improved varieties. Farmers indicated that due to the prevalence of recurrent drought and low soil fertility, most (53%) of the long cycle and late maturing sorghum landraces were either lost or highly marginalized. Shewayrga et al. (2006) also reported that the rate of genetic erosion is enormously increasing due to recurrent drought, low soil fertility and biotic stress factors.

2.4.2.5 Farmers' sorghum variety choices

Farmers had different sets of varieties for different seasons. Most of the surveyed areas received a bi-modal rainfall pattern with a short rainy season (Belg) from March to May and long rains (Meher) from June to September. Consequently, the planting was done twice per year: mid-April to mid-May and mid-June to mid-July. Long cycle and late maturing sorghum varieties were mainly sown in April. If the rainfall comes late, the short cycle varieties were planted. Similarly, after planting, farmers were usually forced to replant several times if the rain became erratic. Jigurty and Amsela were identified as short cycle, early maturing and perceived to be drought tolerant local varieties. These varieties were grown by most households in June/July. All improved varieties, known to be short cycle, early maturing and drought tolerant were grown in the main season (June to September) as a security crop by some of the farmers. Among the long cycle varieties, Jamiyo (takes 5 months to mature) and it is perceived as an early maturing sorghum landraces and it is preferred by most farmers.

Table 2.4.6 List of sorghum varieties grown in 2009/10 cropping season

District	Name of variety
Sekota	Kerkimie (18), Amsela (13), Kuancha (5), Alkua (5), Shula (3), Bulie (1),
	Gobye (1), Birhan (1)
Kobo	Degalet (14), Jamiy, (14), Jigurty (12), Wediakir (1)
Gubalafto	Degalet (25), Jamiyo (9), Jigurty (8), Red America (1), Miltie (1)
Habru	Jigurty (18), Degalet (16), Jamiyo (14), Teshale (3), Abshir (2), Girana-1
	(2), Wediakir (2), Meko (1), Melkamu (1)
Tehulederie	Jamiyo (10), Zengada (8), Kolobo (8), Wediakir (4), Abshir (3), Humera (3),
	Lefo (2), Degalet (2), Timja (1), Seid Musa (1), Ahiyo (1), Aynanbo (1)
Kalu	Tengelie (13), Cherekit (11), Ajaebe (5), Bakelo (5), Jiru (4), Gorad (2),
	Subihan (2), Zengada (2), Esmaeal (1), Mokakie (1), Mira (1)

Figures in parentheses represent frequencies of farmers who grew the variety.

In all the surveyed areas, farmers used intricate combinations of traits in selecting sorghum varieties. Most farmers (86%) were willing to pay for improved varieties if they fulfilled at least three of the traits listed in Table 2.4.7. Higher yield (25%), early maturity (17%), drought resistance (17%), good baking quality (10%), and adaptability (10%), resistance to insects (6%), and biomass yield and stalk palatability (6%) were the traits mainly preferred by farmers (Table

2.4.7). In addition to early maturity, resistance to insects and tolerance to *Striga*; yield potential, drought resistance and adaptability were cited to be the most important traits in the lowlands, whereas 'injera' quality, market price and biomass yield and stalk palatability were perceived as the most important selection criteria in the midlands. Yielding potential, early maturity and drought tolerance were considered as indispensable selection criteria as they allowed the crop to escape drought stress and ensure good harvest under the prevailing moisture stress conditions. Sorghum is mainly grown for household consumption and women are often responsible for ranking varieties for their post-harvest desirable traits. Women ranked good baking ("injera") and brewing ("tella") qualities as top criteria for varietal choice while men did not consider these traits to be important. Women also indicated that most of the improved varieties were not desirable due to their poor baking and brewing qualities. This indicates that varietal development should consider gender aspects.

Table 2.4.7 Farmers'-preferred traits for sorghum variety choice

Trait	All (N = 171)	Low-altitude	Mid-altitude
		(N = 79)	(N = 92)
Yielding capacity	140 (24.8)	68 (25.8)	72 (24.0)
Early maturity	97 (17.2)	45 (17.0)	52 (17.3)
Drought resistance	97 (17.2)	48 (18.2)	49 (16.3)
Good baking quality	57 (10.1)	24 (9.1)	33 (11.0)
Best adaptability	56 (9.9)	30 (11.4)	26 (8.7)
Resistance to insects	33 (5.9)	16 (6.1)	17 (5.7)
Good biomass and stalk palatability	33 (5.9)	13 (4.9)	20 (6.7)
Tolerance to Striga	18 (3.2)	8 (3.0)	10 (3.3)
Good market price	15 (2.7)	4 (1.5)	11 (3.7)
Resistance to birds and storage pests	7 (1.2)	3 (1.1)	4 (1.3)
Good malting ability	6 (1.1)	3 (1.1)	3 (1.0)
Resistance to diseases	5 (0.9)	2 (0.8)	3 (1.0)

Note: The sum of the frequencies does not add to the total number of sampled farmers because farmers have given multiple traits and traits have been grouped and their frequencies are summed up

2.4.2.6 The use of improved varieties

As the country of origin and domestication for sorghum, there is tremendous amount of variability of the crop in Ethiopia. The Ethiopian Sorghum Improvement Programme (ESIP) was started in the 1970s. At the early stages, ESIP was mainly involved in exploring the local germplasm and collected 8000 farmers' varieties in collaboration with Plant Genetic Resource Centre of Ethiopia (Gebrekidan, 1975). The author elucidated the importance of the Ethiopian sorghum germplasm in supplementing the genetic base of the world collection and serving as a gene pool for a number of traits.

ESIP mainly focused on exotic materials which were imported and screened in three agroecologies (lowland, intermediate and highland). The exotic materials were poorly adapted to the highlands; thus, ESIP focused on local materials for the higher elevations. However, the lowland programme exclusively used exotic germplasm since the landraces lacked the early maturing trait dwarfing genes (Kebede, 1991; Gebrekidan and Kebede, 1978). ESIP have released more than forty improved varieties since 1973, out of which about 16 varieties were released for the lowlands and the rest were released for intermediate and high lands (Table 2.4.8). All the sorghum varieties that have been released by the national and regional sorghum research programmes in drought prone areas were from exotic sources. The varieties were mainly released for their early maturity, drought resistance, resistance to *Striga hermonthica* and stalk borer, and relatively high yield potential. Despite the huge yield potential (2 – 6 tons per ha), these varieties which provide six fold more yield than farmers varieties are not appealing to the smallholder farmers owing to various reasons.

Even if considerable efforts have been made to improve the productivity of the crop, only 9.5% of the sampled farmers have experience to use improved varieties. By and large, the majority of the interviewed farmers in the study area (91%) grew local varieties. Comparatively more farmers in the lowlands (13%) had more experience to grow improved varieties than the midaltitudes (6%). This may be explained by the fact that the more risk-prone situations in the lowlands forced farmers to experiment new varieties.

Table 2.4.8 Introduced sorghum varieties released for the lowlands of Ethiopia

Variety	Pedigree	Year of	Plant	Maturity	Seed	Yield	Specific
		release	height	date	color	(t/ha)	characters
76T#23	76T#23	1976			white		Early maturing
Kobomash 76	NES-830x705	1976			white		
Melkamash 79	Diallel Pop 7-682	1979			white		
Meko	M 36-121	2000	110-170	120	white	4.0	Good food
							making quality
Yeju	ICSV 111	2000	150-210	108	white	5.0	Early maturing
							and drought
							tolerant
Teshale	3443-2-OP	2000	169-271	123	white	6.1	Drought tolerant
Gobye	P-9401	2001	110-163	103	white	4.0	Striga resistant
							and early
							maturing
Abshire	P-9403	2001	113-164	101	white	3.0	Early maturing
							and Striga
							resistant
Birhan	# 8566	2001	106-167	101	reddish	4.0	Striga resistant
					brown		
Abuare	90 MW 5335	2003	134-156	120	white	4.0	Drought tolerant
Hormat	ICSV 1112 BF	2005	161-171	121	white	2.3	Striga resistant
Girana-1	CR:35 x DJ1195	2007	135-305	122	white	4.0	Drought tolerant
	x N-13						
Misikir	PGRC/E69441 x	2007	123-191	126	white	4.0	Drought tolerant
	P-9401						and resistant to
							stalk borer
Raya	IS234453xP-	2007	114-220	126	white	2.5	Drought tolerant
	9403						
Gedo	Gambella	2007	116-138	122	white	3.4	Drought tolerant
	1107xP-9401						
Mesay	Meko x Gobye	2011	138-231	119	white	3.9	Early maturing

Source: Sirinka Agricultural Research Center (SARC)

The released exotic early maturing dwarf sorghum varieties were not well assimilated into the production system. About 32% of the respondents indicated that they were willing to use

improved varieties but they had no access to information. Farmers required field demonstrations of the new varieties to observe the superiorities of the varieties to those varieties that they were growing. However, 24% of the farmers were reluctant to use improved introduced varieties because the specialized semi-dwarf lines were not attractive to them due to their reduced biomass, making them less valuable for livestock feed. The stalk is an important product and is used as cooking fuel, fodder, and simple construction material. Wortmann *et al.* (2006) indicated that in Ethiopia, the value of the stalk is about 40% of the value of the grain produced.

On the other hand, 14% of the respondents indicated that in good seasons the local cultivars yielded better than the improved varieties. Most of the farmers lack financial resources to pay for improved seed and commercial fertilizer. About 8% and 6% of the households were not willing to use improved varieties due to high input prices and the varieties were highly susceptible to bird damage, respectively (Table 2.4.9). Un-affordability is one of the reasons for the low level of use of improved seeds. It is not just the seed that is expensive but its requirements for other inputs (fertilizer and pesticides) make it un-affordable. Fertilizer use was not that common in the study area; only 20% of the interviewed farmers applied fertilizer on sorghum during the 2009/10 cropping season. The reason for not using fertilizer may be related to the risk of applying it in erratic rainfall conditions.

Table 2.4.9 Farmers' reasons for not using improved varieties

Reason	All (N = 171)	Low-altitude	Mid-altitude	Percent
		(N = 79)	(N = 92)	
Lack of information	16	7	9	32.0
Low biomass/dwarf varieties	12	4	8	24.0
Low yielding varieties	7	0	7	14.0
Susceptibility to birds	4	1	3	8.0
High input price	3	1	2	6.0
Shortage of land and sharing out land	3	1	2	6.0
Susceptibility to diseases, insects and	2	1	1	4.0
Striga				
Susceptibility to moisture stress	2	0	2	4.0
Poor food quality	1	0	1	2.0

The farmers indicated that, due to the recurrent moisture stress, the impact of fertilizer was insignificant. Sorghum is a self-pollinated crop and culturally farmers maintained their own seed for centuries. To encourage adoption of sorghum technologies, the price of improved seeds is subsidized by the government. Improved seeds were distributed with the price range of 5 – 15 Birr/kg while the average price of local seeds is 10.67 Birr/kg. Increasing production and productivity of the agricultural sector by encouraging farmers to use improved technologies is given high priority by the government. As a result, the price of most crops' improved varieties seeds is highly subsidized

Farmers indicated that the improved varieties were very early maturing with white seed color that made them liable to bird attack. The white varieties are very appealing to birds while red and brown sorghum varieties are less attractive. Although it is not scientifically proven, red and brown grain types are often associated with high tannin content, and are less preferred by birds and less affected by mold (Wortmann *et al.*, 2006). Traditionally, farmers grow different varieties of various seed colors (white, red, yellow) and head types (loose and compact) in their plots to minimize bird damage. Interestingly, while characterizing the farmers' varieties, it was found that there were white sorghum cultivars which were not eaten by birds. This may enable breeders to introgress bird resistant gene(s) into otherwise better yielding locally adapted sorghum varieties.

Access to information is one of the key factors for technology transfer. About 65 and 19% of the respondent farmers obtain information from the development agents and neighboring farmers, respectively (Table 2.4.10). However, in the midlands farmers got information from the nearby research centers. It is not surprising that farmers in the lowlands did not access information from research centers, since two of the research centers in the North Eastern Ethiopia are situated in the midlands. Government extension programmes play a vital role in building the knowledge and transferring information to the farming community. Almost all the interviewed farmers (96.5%) had access to extension services. Most of the farmers (82%) consulted the extension agent at least once a month during the growing season. Only 25% of the interviewed farmers listened to agricultural programmes on TV or radio.

Table 2.4.10 Sources of information for farmers

Source	All (N = 171)	Low-altitude (N = 79)	Mid-altitude (N = 92)	Percent
Development agent	62	33	29	65.3
Neighbors	18	7	11	18.9
Nearby research center	10	0	10	10.5
Radio	2	1	1	2.1
TV	2	2	0	2.1
On farm trials/Field days	1	0	1	1.1

Only about half of the interviewed farmers had information about improved varieties of sorghum. It is rather surprising that 71% of the farmers have never used any improved varieties. Farmers in many places do not know about the available sorghum technologies, mainly due to the loose research-extension-farmer linkage. Only 16% of the interviewed farmers had participated in technology evaluation and transfer activities such as field days, Farmers Training Centers (FTCs), seminars and on-farm trials. In the ARNS, 18,083 households were participating in sorghum extension package programmes, mainly on Striga control practices and fertilizer use. However, most of the areas under the extension programme concentrated on tef, maize and wheat production. Surprisingly, despite its importance to the national agricultural production, there is no much extension activity in sorghum improved seed production. However, there are many early maturing and highly productive sorghum varieties available in the region (CSA, 2012). The effect of improved technology on the livelihoods of subsistence farmers may be noticeable if more farmers apply improved sorghum technologies. Low adoption of improved varieties and high preference for local varieties are explained by such variety attribute factors. The main reason for poor adoption rate and low impact of the improved varieties is breeders' failure to integrate the traits farmers' desire. McGuire (2008) described that the process of technology development should include farmers preferences and explore their knowledge.

2.4.3 Constraints to sorghum production and coping mechanisms

2.4.3.1 Constraints

Although the farmers in Wello are rich in sorghum genetic diversity, they could not produce sufficient yield to feed their families. As they relied on rainfed sorghum production moisture

stress, insect pests, low soil fertility, the parasitic weed *Striga hermonthica* and birds are among the major constraints contributing to poor crop performance and low yield (Table 2.4.11). Ceccarelli *et al.* (1991) also reported that resource poor farmers in marginal areas sufferred more from such production constraints than others. Though the importance of these constraints varied from District to District, moisture stress was ranked first in all the areas. The second important problem was insect pests, of which stalk borer, armyworm and ball worm were ranked as the major ones. In mid-altitudes, farmland shortage, poor soil fertility and weeds were found to be more important than in the lowlands. On the contrary, low yielding local varieties, birds, limited and poor quality of improved varieties were seen as major constraints in the lowland areas. About 14% of the respondents indicated that they had shortage of land. Poor soil fertility was also a significant problem which was associated with soil erosion and continuous cultivation of the land with no fallowing and nutrient replenishment due to high population density.

Generally, moisture stress, insect pests, land shortage and poor soil fertility are the most important problems that farmers face in agricultural production. As sorghum is an indigenous and highly diversified crop in the North Eastern Ethiopia, analyzing the extent of genetic variation maintained by farmers and assessing the genetic potential of landraces for moisture stress and identifying moisture-stress tolerant varieties will provide a sustainable solution. Considering the tremendous availability of sorghum diversity and chronic problem of recurrent drought, further studies need to be conducted on improving the yielding potential of local cultivars through breeding for drought resistance.

Table 2.4.11 Major constraints for sorghum production

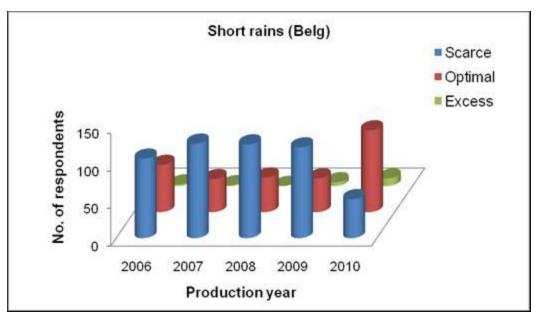
Constraint	All (N = 171)	Low-altitude	Mid-altitude (N	Percent
		(N = 79)	= 92)	
Moisture stress	170	78	92	28.6
Insect pests	143	66	77	24.0
Limited land availability	88	38	50	14.8
Poor soil fertility	82	33	49	13.8
Weeds	24	8	16	4.0
Low yielding local varieties	22	12	10	3.7
Other vertebrate pests (birds and rodents)	17	10	7	2.9
Limited availability of improved varieties	16	10	6	2.7
Diseases	10	4	6	1.7
Poor quality of improved varieties	9	7	2	1.5
Flood	6	5	1	1.0
High cost of commercial fertilizer	6	1	5	1.0
Poor supply of seeds	2	1	1	0.3

Note: The sum of the frequencies does not add to the total number of sampled farmers because some farmers have given multiple constraints and the constraints have been grouped.

2.4.3.2 Moisture stress

A. Rainfall pattern

Since the North Eastern part of Ethiopia is characterized by erratic rainfall (both in amount and distribution), farmers were requested to describe the rainfall patterns of the area as scarce, optimal or excess. Although the percentage of farmers rating the different seasons varied across Districts, the majority of the farmers agreed that *Belg* rain in the years from 2006 – 2009 was scarce, while 2010 was described as the year of adequate rain. However, the Meher rains were optimal from 2006 – 2009 and excess in 2010 (Figure 2.4.2). Although this needs further investigation, there is probably a tradeoff between *Belg* and *Meher* rain as low rain in *Belg* relates to relatively enough rain in *Meher* and adequate rain in *Belg* associates with excess rain in Meher. In the years from 2006 to 2009, mainly short cycle varieties were planted and there was low level of sorghum production. Conversely, in 2010 there was huge production of high yielding and long cycle sorghum varieties such as *Kerkimie, Degalet, Jamiyo, Tengelie*, and *Cherekit*.



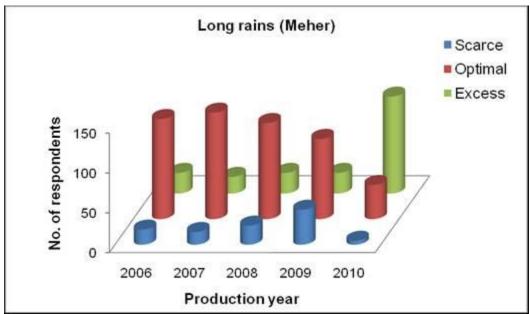


Figure 2.4.2 Rainfall patterns for short (Belg: March to May) and long rainy season (Meher: June to September) from 2006 - 2010

B. Frequency of moisture stress

Farmers often mentioned that their strong dependence on rainfall for agriculture hardly satisfied food needs of their households. Moisture stress within and between seasons was a common phenomenon in all the surveyed areas. About 46, 29 and 13% of the interviewed farmers replied that they experienced moisture stress every year, every two years and every three years,

respectively. Farmers were facing a severe rain shortage during *Meher* (84% households) than *Belg* season (10% households). Only 6% of the respondents mentioned that moisture stress was a problem in both seasons.

The effect of moisture stress on crop yield is dependent on the stage of plant development. In this study, most of the farmers (64%) indicated that they encountered moisture stress at grain filling stage and 27% at flowering stage (Table 2.4.12). This result is in agreement with Wortmann *et al.* (2006) who reported that terminal stress is a major occurrence in Ethiopia. Anthesis and grain filling stages appear to be the most vulnerable growing stages that may result in reduced yield and/or complete crop failure (Khanna-Chopra and Sinha, 1988).

Table 2.4.12 Farmers' experience of moisture stress among the different growing stages

Stages of plant growth	Frequency	Percent
Grain filling stage	110	64.3
Flowering stage	46	26.9
Flowering and grain filling stages	8	4.7
Seedling and grain filling stages	5	2.9
Seedling stage	1	0.6
Seedling and flowering stages	1	0.6

2.4.3.3 Moisture stress coping mechanisms

Moisture stress was a very common production problem in the study areas. Most of the farmers (88%) reported moisture stress problem occurring at least every three years. Table 2.4.13 presents the most important coping strategies to deal with this production problem. Farmers had some indicators to predict impending moisture stress. For example, very cold weather with little rain showers in the months of October and November was taken as a sign of moisture stress in April. In such cases, farmers suspended growing late maturing and high yielding sorghum varieties in favor of planting early maturing and low yielding varieties from mid-June to mid-July. If they expected sufficient rain in the *Belg* season, they planted late maturing sorghum varieties. When they anticipated different levels of moisture, farmers planted a range of sorghum varieties from different maturity groups to increase the probability of good harvest and/or to avoid a complete crop failure.

About 27% of the interviewed farmers indicated that at the time of moisture stress they grew early maturing sorghum varieties to cope with the stress conditions and 22% replaced sorghum with other crops such as tef and chickpea. Quite a large number of farmers (38%) pointed out that they either planted early maturing varieties or replaced sorghum with other crops depending on the availability of seeds and the severity of the stress conditions. Other practices included stone mulching (4%), planting early maturing varieties using stone mulch (2%), planting stress tolerant sorghum varieties (2%), and plowing the land using a tie ridger (1%) as major moisture stress coping strategies (Table 2.4.13). And yet some 4% of the households believed that the problem came as a curse from God and there was no mechanism to alleviate moisture stress at all. Stone mulching is one moisture conservation mechanisms, which is practiced through covering the land with stones to prevent evaporation from the soil. In general, farmers in North Eastern Ethiopia have long experiences to cope with unreliable rainfall by either growing a number of varieties from different maturity types or shifting from late maturing to early maturing varieties or replacing sorghum with tef or chickpea.

Table 2.4.13 Moisture stress coping mechanism and their relative importance

Mechanism	All (N = 171)	Low-altitude	Mid-altitude	Percent
		(N = 79)	(N = 92)	
Planting early maturing sorghum	115	57	58	46.4
varieties				
Replacing the crop with other crop	111	39	72	44.8
Stone mulching	11	5	6	4.4
Planting moisture stress tolerant	3	2	1	1.2
sorghum varieties				
Use tie ridger	1	0	1	0.4
Applying irrigation	1	0	1	0.4

Note: The sum of the frequencies does not add to the total number of sampled farmers because some farmers have given multiple mechanism and these have been grouped.

2.4.3.4 Other production constraints

Farmers indicated that most of the insect pests co-existed with their crops all the time. However, the insect pest incidence and distribution varied from year to year depending on the climatic conditions and cropping systems. The major sorghum pests were stalk borer, army worm, boll

worm, grasshoppers, sorghum chaffer, shoot fly and storage weevils (Table 16). Stalk borer was considered as the most important sorghum pest followed by army worm in all the surveyed areas. Farmers indicated that they conspicuously observed the symptoms of stalk borer as it produces bored and broken stems, chaffy heads, dead hearts and stunted growth. Farmers very well recognized the impact of factors such as drought and crop rotation on the incidence of stalk borers. In the study areas, the use of pesticides and botanicals and rouging out of infected plants were the most cited pest control practices. Generally, the occurrence of diseases was not as frequent and severe as insects; occasionally, diseases such as Anthracnose, smut, leaf blight, and rust were mentioned as production constraints. The incidence and severity of diseases may be related with drought as disease severity has a positive association with moisture.

Table 2.4.14 Important insect pests and diseases of sorghum in the study area

Local name of the	Common name	All (N =	Low-altitude	Mid-altitude	Percent
insect pest	(English name)	171)	(N = 79)	(N = 92)	
Ageda korkur	Stalk borer	130	60	70	55.3
Temch	Army worm	39	20	19	16.6
Guay til	Ball worm	21	8	13	8.9
Fentara/Anbeta	Grasshopper	16	6	10	6.8
Zinzina	Sorghum chaffer	11	4	7	4.7
Nekez	Storage weevils	8	3	5	3.4
Degeza	Welo bush cricket	7	3	4	3.0
Bileh	Shoot fly	3	0	3	1.3
Local name of the	Common name	All (N =	Low-altitude	Mid-altitude	Percent
disease	(English name)	171)	(N = 79)	(N = 92)	
Lamba/Adrik	Anthracnose	20	11	9	27.4
Aregoso/Chores	Smut	19	12	7	26.0
Mich	Leaf blight	9	3	6	12.3
Magid/Wag	Rust	9	5	4	12.3
Marazinib	Honeydew (ergot)	5	5	0	6.8
Fengle	Root rot	4	4	0	5.5
Kolim		4	2	2	5.5
Yerkum anget		3	2	1	4.1

Note: The sum of the frequencies does not add to the total number of sampled farmers because some farmers have given multiple diseases/insect pests and these have been grouped.

2.5 Conclusions and implications for sorghum breeding priority setting

In North Eastern Ethiopia, crop production is the most important livelihood activity supplemented by livestock production to a smaller extent. Sorghum and tef are the two most important crop enterprises on which millions of farmers depend, especially in drought prone and complex farming systems of the region.

Sorghum has a variety of uses *i.e.* the grain is used for food and the stalk as livestock feed, fuel wood and construction material. It is grown in a wide range of agro-ecologies; most importantly in highly moisture stressed environments where other crops can least survive. The North Eastern Ethiopia region is known for its rich sorghum genetic diversity, mainly due to the agro-ecological and cultural diversity of the growers and consumers. Despite its huge potential in the region, sorghum productivity is well below the national and global average. The results suggest that the main bottlenecks for this poor performance include moisture stress (29%), insect pests and diseases (24%), farm land fragmentation due to demographic pressure (15%), poor soil fertility (14%), weeds (4%) and poor performance of the local varieties (4%). The productivity of sorghum is largely hindered by the use of inherently poor yielding local landraces as farmers are forced to abandon their high yielding, big-headed and late-maturing sorghum varieties due to the prevalence of recurrent moisture stress.

In the target region, moisture stress during grain filling stage was one important constraint followed by insect pests particularly shoot fly. Although drought is largely unpredictable, farmers in North Eastern Ethiopia were dealing with it by either growing a diverse set of traditional varieties from different maturity types or shifting from late maturing to early maturing varieties or replacing sorghum with tef or chickpea. The use of moisture stress tolerant varieties was considered to be less important due to the fact that there were very few alternatives and many of the traditional varieties were long cycle and susceptible to moisture stress. The high yielding and drought tolerant improved varieties commercially available were less appealing to the typical small scale farmers due to many socio-cultural and post-harvest quality preferences.

The importance of effective research-extension-farmer linkage for better diffusion and impact of improved technologies cannot be under-estimated. Despite the greater availability of improved sorghum technologies from the research system they have not yet been adequately transferred

and adopted by farmers. Less coordinated and fragmented efforts of the research and extension activities have by and large affected both the process and the outcome. To benefit from the available improved technologies, farmers have to be part of the breeding process right from the very beginning. This can be done through participatory plant breeding in identification of priority traits, on-farm demonstrations, popularization and re-evaluation of the technologies. The different stakeholders need to be involved in identifying farmers' needs and priorities and re-orienting the research agenda for the development of demand-driven client oriented technologies and feedback assessment of the performance of the technologies.

Farmers distinguish and describe the landraces using many agro-morphological characters. They take into account a number of varietal attributes (such as grain yield potential, total plant biomass, insect/disease resistance, post-harvest food quality, earliness, drought resistance, and bird and *Striga* resistance). Despite the crop's potential to improve food security and agricultural sustainability, the genetic resource base has not yet been exploited to its full potential. The sorghum improvement efforts of the country mainly focused on selection of genotypes from exotic sources. Farmers keep crop evolution alive through shaping and maintaining genetic diversity based on their diverse needs and requirements. Farmers grew a mixture of sorghum landraces in a single plot and the relative proportion of the varietal mix depended on the prevailing environmental conditions and diverse farmers' needs and requirements. Emphasis should be therefore be given to conservation and utilization of farmers' varieties which have evolved and adapted over the years to local agro-climatic conditions and make use of their merits in breeding programmes to develop varieties.

As indicated by many authors, sorghum is believed to have originated from Africa. As a result, enormous amount of genetic variability exists. As a crop that is grown in a wide range of geographic areas covering a range of latitude and longitude, altitude, day length, rainfall and temperature regimes, sorghum is adapted to a range of biotic and abiotic stress factors. The crop shows high morphological and genetic diversity for several traits. In the traditional farming systems, both natural and artificial selection pressures are involved in generation and diversification of sorghum genetic resources over time and space. The availability of adequate genetic variability among the traditional sorghum varieties is an opportunity that enables breeders to breed for high productivity and yield stability. To improve the productivity and yield stability under drought stress conditions, landraces are important sources of genes as they are well adapted and maintained in a wide range of environments. The potential of the traditional varieties as sources of breeding material should be well studied and documented.

The current direction of agricultural research in Ethiopia is towards importing and testing the adaptation of finished and market oriented technologies. The research system has so far released more than 16 lowland improved sorghum varieties. However, farmers still continue to grow local varieties that are low yielding and incompatible with the changing environment. One of the reasons that the new exotic varieties failed to assimilate well into the production system is that farmers are operating in complex and risk prone environments. The conventional plant breeders use very few selection criteria which revolve around yield, uniformity and stability while traditional farmers employ more diverse and complex selection criteria revolving around stable crop performance over seasons and grow a range of landraces which meet their needs in a very complex and heterogeneous environments.

Farmers employ several selection criteria often influenced by socio-cultural, economic, and environmental conditions. The selection criteria include yielding capacity, early maturity, drought resistance and adaptability, baking quality, resistance/tolerance to insects and weeds, biomass yield and stalk palatability. The relative importance of selection criteria vary from District to District and among altitudinal zones. For example, yield potential, drought resistance and adaptability are reported to be the most important criteria in the lowlands but of relatively less importance in the midlands. Similarly, baking quality, market price, biomass yield and stalk palatability are very important selection criteria in the midlands.

It is the author's belief that assessing the level of genetic variation maintained by traditional farmers should enhance efficient utilization of genetic resources. Moreover, to improve the productivity of the crop through breeding for drought resistance, landraces are important sources of genes. The heterogeneity of tropical landraces of sorghum enables it to display high adaptation to drought stress. Farmer participation in setting breeding goals and varietal evaluation will remain critical to enhance adoption and genetic diversity. Breeders and farmers should work together in elucidating selection criteria so as to overcome crop loss in the heterogeneous and marginal environments. Genetic improvement should, however, be complemented by efficient moisture conservation and agronomic practices.

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CHAPTER 3

Genetic Diversity Assessment among Lowland Sorghum [Sorghum bicolor (L.) Moench] Landraces using Qualitative Traits

3.1 Abstract

A total of 262 sorghum [Sorghum bicolor (L.) Moench] landraces collected from the lowlands of Wello, Ethiopia, were assessed for 12 qualitative characters to determine the level of genetic diversity among the accession, within and between districts of collection. Field characterizations were conducted at two localities using an unbalanced-incomplete block design with two replications. For all accessions and districts of collection phenotypic frequencies were determined per class to calculate phenotypic diversity indices using the Shannon-Weaver diversity index (H'). Multivariate methods, principal component, cluster and discriminant analyses, were performed to examine the patterns of variation with respect to regions of collection. Results revealed that all characters observed showed high phenotypic diversity ranging from 0.98 to 1.00 and an average diversity index of 0.91. Four principal components (PC) contributed to 82% of the total variation among the landraces. Thirty four percent of the total variance was accounted for PC1 which was highly correlated with stay-green, inflorescence compactness, grain colour and panicle exsertion. The discriminant analysis identified five functions that explained 92% of the variation among the accessions and districts of collection. The DA depicted 59% correct regional base classification of the landraces. Overall, the accessions displayed high polymorphism for leaf rolling and stay-green traits which are important for selection under drought stress conditions. Intra-district phenotypic diversity index was significantly larger than Inter-districts diversity. The study demonstrated the existence of considerable level of diversity that could be exploited for future sorghum breeding and for conservation of the germplasm.

Keywords: diversity index, Ethiopia, phenotype, Sorghum bicolor, Wello

3.2 Introduction

Sorghum (*Sorghum bicolor* [L.] Moench) is the fifth most important cereal crop worldwide after wheat, rice, maize, and barley (FAO, 2006). In developing countries, the grain is used for food and the stalk for fodder and building materials, whereas, in developed countries it is primarily used as animal feed. Sorghum is a widely adaptable crop though its production has been limited to water- and heat-stress areas within subtropics and tropical regions of the world. It has high yield potential comparable to other cereals such as rice and wheat under optimal conditions. In 2005, the total annual worldwide sorghum production was 58.6 million tons from nearly 44.7 million ha, with an average yield of 1.31 t/ha (FAO, 2006). It is the main staple food in Ethiopia, ranking fourth after tef (*Eragrostis tef*), maize (*Zea mays*), and wheat (*Triticum aestivum*), both in area coverage, and production (CSA, 2012). In Ethiopia the crop is mainly used as a source of food, feed, production of local beverages and construction material at the household level (Teshome *et al.*, 1999; McGuire, 2000).

Ethiopia has diverse geophysical and climatic environments that result in a great diversity of tropical, subtropical and temperate climates, soil and vegetation. The variability in environmental factors has an important implication on the diversity of crop plants in the country. Thus, Ethiopia is considered as one of the centers of crop origin or diversity (Vavilov, 1951; Flora of Ethiopia and Eritrea, 1999). Further the presence of several landraces and wild type crop species places the country as a primary or secondary centre of diversity. The country is a primary or secondary diversity centre for crops including barley, sorghum, tef, tetraploid wheats, finger millet, gomenzer, noug, linseed, safflower, faba bean, fieldpea, chickpea, lentil, fenugreek, grass pea, cotton, kenaf and castor bean (IBC, 1995). Harlan (1969) described Ethiopia as a land of crop diversity.

Sorghum is one of the most diverse crops distributed over a wide range of agro-ecologies in Ethiopia. Several wild, weedy, and intermediate forms of sorghum are reported (IBC, 1995). Assessment of genetic variability in crops is useful for crop improvement programmes and conservation of genetic resources (Assar *et al.*, 2005). Genetic variability can be detected at morphological, biochemical or molecular level. Considerable genetic variations are manifested as visible morphological traits (Ayana and Bekele, 1999). Ayana and Bekele (1998, 1999, 2000), based on 415 sorghum accessions collected from different regions of Ethiopia and Eritrea, reported high phenotypic variations that existed between the regions of collections. Teshome *et al.* (1997) and Abdi *et al.* (2002) also studied morphological variation among

sorghum landraces collected from North Shewa and South Wello regions of Ethiopia. Geleta and Labuschagne (2005) estimated the genetic diversity of 64 sorghum accessions collected from the eastern highlands of Ethiopia. The authors indicated that there was a wide morphoagronomical variation in the Ethiopia sorghum landrace collections.

A major collection of land races was made in 1973 by Ethiopian Sorghum Improvement Programme (ESIP) in the north eastern region of the country where the largest diversity is found. In this region drought stress is a major sorghum production constraint. Its adaptation to drought stress environments makes sorghum an important cereal crop in Ethiopia and worldwide (Doggett, 1988; House, 1995). Kebede and Menkir (1987) contend that sorghum is an important crop in the lowland areas of Ethiopia where rainfall is unreliable and recurrent drought is common. Although this region is where the largest diversity of sorghum is found, assessment of the level of genetic diversity within and between localities is limited. In this study, 262 local landraces of sorghum collected from the lowlands of north eastern Ethiopia were evaluated for 12 qualitative characters and the levels of genetic diversity were estimated. To this effect, the objectives of this research were: (i) to assess the extent of phenotypic variability among sorghum landraces using 12 phenotypic characters, and (ii) to estimate the level of diversity within and between districts of collection.

When a large number of accessions are to be evaluated for several morphological and agronomic characters, multivariate methods are useful statistical tools often used for characterization, evaluation and classification of genetic resources (Peeters and Martinelli, 1989; Ayana and Bekele, 1999). These methods are also useful in identifying groups of accessions that have desirable characters useful for crossing (Ayana *et al.*, 2003; Grenier *et al.*, 2004; Geleta and Labuschagne, 2005; Ali *et al.*, 2009), for effective planning of germplasm collection (Ayana *et al.*, 2003; Geleta and Labuschagne, 2005), for establishing core collections (Frankel, 1984; Frankel and Brown, 1984; Brown, 1989 a and b; Kouame and Quesenberry, 1993), for examining the patterns of variation in germplasm collection (Brown, 1991; Perry and McIntosh, 1991) and studying some aspects of crop evolution (Camussi *et al.*, 1985; Cowen and Frey, 1987).

3.3 Materials and Methods

3.3.1 Plant material

Out of the total landraces collection of Ethiopia, 262 sorghum accessions (*Sorghum bicolor* (L.) Moench) of lowland origin collected from the northeastern parts of Ethiopia were used for this study. The landraces were obtained mainly from the Ethiopian Institute of Biodiversity Conservation (IBC) along with their passport data. The landraces were collected from three Administrative Zones (Waghimra, North and South Wello) represented by nine Districts: Ambasel (63 entries), Kalu (68) [South Wello]; Kobo (9), Bugna (14), Gubalafto (63), Habru (8) [North Wello]; Dahina (14) Zikuala (9), and Sekota (14) [Waghimra].

3.3.2 Field experiments and data collection

Field experiments were conducted at Sirinka Agricultural Research Center and Kobo sub center under rainfed conditions. Sirinka and Kobo research sites are situated at 11°45' and 12°9' N latitude and 39°36' and 39°38' E longitude with an altitude of 1850 and 1400 masl, respectively. The 262 accessions were grown in single row plots of 3m at a seeding rate of 10kg ha⁻¹ and inter and intra spacing of 0.75m and 0.3m, respectively. The experiment was laid out in an unbalanced incomplete block design of 10 blocks and 32 plots within each block in two replications. Fertilizer was applied at the rate of 100kg ha⁻¹ diammonium phosphate (DAP) and 50kg ha⁻¹ urea as recommended for sorghum in the lowland of Ethiopia. All the DAP was applied at the time of planting, while urea was applied in a split application. Other standard agronomic practices were followed as required.

Twelve qualitative characters, which were obtained from the standard sorghum descriptor lists for characterization, were used for the sorghum phenotypic characterization (IBPGR/ICRISAT, 1993) (Table 3.3.1). Scoring for stay-green character and leaf rolling were done since sorghum accessions grown under rainfed conditions, at both sites, normally experience post-anthesis moisture stress.

Table 3.3.1 Sorghum morphological descriptors and their keys used in the study

Key	Descriptors and code
PE	Slightly exserted <2cm (1), exserted 2-10cm (2), well exserted
	>10cm (3), peduncle re-curved (4)
LC	Dark green (1), light green (2)
LO	Erect (1), dropping (2)
IC	Very loose erect (1), very loose dropping (2), loose erect (3),
	loose dropping (4), semi loose erect (5), semi loose dropping
	(6), semi compact elliptic (7), Compact elliptic (8), Compact oval
	(9), half broom corn (10), broom corn (11)
GRC	Red (1), yellow (2), brown (3), white (4), Light orange (5), white
	with orange (6), white with red (7)
AW	Absent (1), present (2)
GLC	White (1), red (2) Purple (3), black (4), gray (5), brown (6), dark
	brown (7)
GLV	25 % grain covered (1), 50% grain covered (2), 75% grain
	covered (3), 100% grain covered (4), glume longer than grain (5)
HS	Elliptical (1), oblong (2), round (3), semi loose (4) loose (5)
MDC	White (1), dull green (2), yellow (3), brown (4), purple (5)
LR	Non rolled leaf (1), 25% leaves rolled (2), 50% leaves rolled (3)
	75% leaves rolled (4), all leaves rolled (5)
SG	Very slight senescent (1), 25% leaves senescent (2), 50%
	leaves senescent (3), 75% leaves senescent (4), complete
	senescent (5)
	PE LC LO IC GRC AW GLC GLV HS MDC LR

3.3.3 Data analysis

The raw data of 262 genotypes, grown at the two locations, were subjected to statistical analysis using GenStat (Payne *et al.*, 2008) and Agrobase (2007). Frequency distributions for the twelve qualitative characters were determined for all the accessions and grouping observations according to district of collection. Shannon-Weaver index of diversity (H') was computed for each character from frequency distributions observed in the different classes

(Perry and McIntosh, 1991) as $H'=1-\sum_{i=1}^n pi\log_e pi$; where, H'= Shannon Diversity Index; $p_i=$

the proportion of accessions in the ith class of an n-class character; n= the number of phenotypic classes of traits. Each diversity index value was divided by its maximum value (log_en) and normalized to keep the values between 0 and 1. This method is commonly used to determine diversity from phenotypic frequencies among germplasm (Ayana and Bekele, 1999; Grenier *et. al.*, 2000; Kefyalew *et al.*, 2000; Grenier *et. al.*, 2004; Geleta and Labuschagne, 2005). The diversity index for each character was computed from the complete data set while the average diversity index was computed for each character for the collection regions. Besides, proportions of diversity within localities and between localities were estimated in relation to the total variation.

Standardization of the data to a mean of zero and a variance of unity was made to avoid differences in scales used for recording data on the different characters. The data was also subjected to Principal Component (PC) analysis procedure using SAS statistical software (SAS Institution, 2004) and Agrobase (2007). PC analysis was conducted on average values measured from 262 accessions and nine districts of collection over three provinces. This method is used to identify the traits contributing to a larger part of the total variation among the landraces (Sneath and Sokal, 1973).

Principal component analysis is a useful statistical technique often used to identify patterns in the data, and expressing the data in a way that number of prevailing components and associated predictor variables are identified (Kim and Muller, 1978; Johnson and Wichern, 1988; Heberger *et al.*, 2003). This method helps to classify our data set as a combination of the contributions from each of those predictor variables and describes the linear relationship of the predictor variables as a component that accounts for most of the variation in the observed variables. This method is also used to avoid multicollinearity in the data set that might adversily affect the selection response in crop improvement programmes.

Discriminant analysis was used to verify the accessions membership to their regions of collection. Hierarchical cluster analysis was performed to examine the grouping patterns of the nine regions of collection based on their similarity with respect to the corresponding means of all the twelve qualitative characters. The measure of similarity was Euclidian distance and clustering method was Un-weighted Pair-Group Method using arithmetic mean (UPGMA)

(Sneath and Sokal, 1973) For the PCA and DA analyses entry values were used as rows of the input matrix and the 12 qualitative characters as column variables of the matrix.

3.4 Results and discussion

3.4.1 Descriptive statistics

The frequency distribution of the accessions by their district of collection of the qualitative characters is presented in Table 3.4.1. Fifty one percent and 24% of the accession samples were found to have compact elliptical and compact oval heads, respectively. The frequency of semi loose dropping, semi compact elliptical and half broom corn phenotypic classes were comparatively low among the genotypes studied. Only two and four individual samples were observed with loose dropping and semi loose erect phenotypic head classes from the entire sample, respectively. However phenotypic classes of very loose erect, very loose dropping, loose erect, and broom corn heads were not observed in the samples studied. The result complies with Geleta and Labuschagne (2005) and Ayana and Bekele (1998), who reported the predominance of compact elliptical and oval heads, respectively. It was also indicated that very compact panicles were a distinctive characteristic of extremely dry conditions where compact durra sorghum races are frequent in such areas (Rao et al., 1996, Grenier et al., 2004).

The majority (58%) of the accessions were found to be awned. Although awned types were predominant, awnless types were also observed in all the districts. This result agrees with the findings of Geleta and Labuschagne (2005) where the prevalence of awned types was reported in the Hararghe collections. In the present study, only lowland origin sorghum accessions were used and the presence of awns may have some positive impact on drought tolerance. In small cereals, the presence of ear awns, had positive and significant correlation with grain yield and suggested to be used as morphological marker while selecting lines for good photosynthetic activity and high yield under moisture stress condition (Tambussi *et al.*, 2007; Khaliq *et al.*, 2008; Samarah *et al.*, 2009; Taketa *et al.*, 2011). Stay-green was scored for five phenotypic classes as described by Xu *et al.* (2000) and 52% of the landraces showed ½ of the leaves senescent followed by 28% where ¼ of the leaves were senescent. No accession sample was recorded showing complete plant death while 5% of the germplasms were found to be very slightly senescent. This suggests that, in these landrace samples, there might be stay-green genes which can be used as possible phenotypic markers for future breeding purposes.

Of the five phenotypic classes observed for midrib colour, white colour was predominant followed by dull green and yellow. The proportion of landraces with white (39%), dull green (28%) and yellow (31.7%) midrib colours were higher than the proportions of landraces with brown (0.3%) and purple (1%) midrib colours which were significantly lower. Midrib colour, which is not apparently important to growth and survival, is one of the most important traits used by the farmers to differentiate sweet stem and grain sorghum varieties. A wide range of variability was recorded in grain colour, glume cover, glume colour and head shape. These characters are reported to be very important in naming the sorghum landraces in Ethiopia by farmers (Teshome *et al.*, 1997).

Table 3.4.1 Summary statistics of frequency distribution among 12 qualitative traits among 262 sorghum accessions collected from nine Districts

_				Inflo	esc	ence	cor	npact	ion			Aw	/ns		Sta	y-gre	en			(Grair	n colc	ur				Glun	ne co	/er			He	ad sh	ape	
District ^{a, b}	1	2	3	4	5	6	7	8	9	10	11	1	2	1	2	3	4	5	1	2	3	4	5	6	7	1	2	3	4	5	1	2	3	4	5
Amb	0	0	0	0	1	2	2	34	16	8	0	23	40	5	14	35	9	0	15	15	7	20	3	1	2	25	16	18	4	0	20	16	15	10	2
Bug	0	0	0	0	0	0	0	9	2	3	0	7	7	0	4	5	5	0	2	3	1	7	1	0	0	5	1	7	1	0	2	8	1	1	2
Dah	0	0	0	0	0	2	0	10	2	0	0	8	6	0	2	10	2	0	6	3	0	4	0	0	1	2	1	8	3	0	9	2	1	2	0
Gub	0	0	0	2	0	3	8	34	13	3	0	28	35	4	23	33	3	0	21	10	5	22	5	0	0	15	21	19	4	4	21	15	13	11	3
Hab	0	0	0	0	0	2	0	3	2	1	0	3	5	1	1	4	2	0	4	3	1	0	0	0	0	0	4	3	1	0	2	2	1	2	1
Kal	0	0	0	0	3	1	2	31	19	12	0	25	43	2	22	35	9	0	14	17	6	26	3	1	1	19	22	19	6	2	27	6	19	12	4
Kob	0	0	0	0	0	3	2	2	2	0	0	6	3	0	5	4	0	0	5	1	0	2	1	0	0	0	4	3	2	0	1	0	3	5	0
Sek	0	0	0	0	1	0	0	7	4	2	0	7	7	1	4	8	1	0	5	2	3	3	0	1	0	5	2	6	1	0	5	2	4	3	0
Ziq	0	0	0	0	0	1	0	2	3	3	0	3	6	0	0	4	5	0	1	3	1	4	0	0	0	3	3	2	1	0	4	1	0	4	0
X ² -value (p=0.05)					-	75.48	3 (64)					6.34	4 (8)		16	:.35* (8	3)				82.5	9** (4	3)				37.	35 (32	()			50).97* (32)	

		Midrib	colc	ur		Pa	anicle	exser	tion	Le orient		Leaf	f colour Leaf rolling					Glume colour							
	1	2	3	4	5	1	2	3	4	1	2	1	2	1	2	3	4	5	1	2	3	4	5	6	7
Amb	29	14	20	0	0	2	29	23	9	15	48	31	32	48	15	0	0	0	5	0	14	7	29	4	4
Bug	8	4	2	0	0	0	10	1	3	4	10	5	9	8	6	0	0	0	5	1	4	1	1	0	2
Dah	5	3	6	0	0	0	9	4	1	6	8	5	9	13	1	0	0	0	1	0	5	1	4	1	2
Gub	19	23	19	1	1	3	26	30	4	19	44	29	34	56	7	0	0	0	6	3	20	9	15	5	5
Hab	2	2	4	0	0	0	4	1	3	3	6	1	7	4	4	0	0	0	2	1	1	0	2	0	2
Kal	24	18	24	0	2	4	37	20	7	52	16	29	39	51	17	0	0	0	10	0	21	5	22	9	1
Kob	2	4	3	0	0	0	3	5	1	3	11	4	5	8	1	0	0	0	0	0	4	4	0	1	0
Sek	8	3	3	0	0	0	4	7	3	9	5	6	8	9	5	0	0	0	2	0	0	5	4	3	0
Ziq	5	2	2	0	0	0	6	3	0	2	7	5	4	6	3	0	0	0	3	0	1	0	4	1	0
X ² -value (p=0.05)		20.7	6 (32)				29.1	4 (24)		7.75	(8)	5.35	5 (8)		30.0)8* (2	24)				39	9.13 (4	18)		

^a Amb=Ambasel, Bug=Bugna, Dah=Dahina, Gub=Gubalfto, Hab=Habru, Kal=Kalu, Kob=Kobo, Sek=Sekota, Ziq=Zequal

 $^{^{\}rm b}$ Numbers in parenthesis after the $\varkappa^{\rm c}$ -values are degrees of freedom, * and ** denote significant differences at p=0.05 and p=0.01 levels, respectively

Out of the seven phenotypic classes of grain colour used in this study, white, red and yellow grain colours were predominant with 33%, 27% and 21% frequencies respectively. This finding is consistent with the results of Geleta and Labuschagne (2005) where white, yellow and red colours were frequently encountered in the Hararghe collection. This is mainly because sorghum in Ethiopia is grown for food as Injera and making local beverages for which white colour grains are preferred for food and yellow and red grains for the beverages (Ayana and Bekele, 2000). It is also reported that grain colour is recognized as an important trait with a great preference for red and brown seed grains (Wortmann et al., 2006). Red and brown grain types are often associated with high tannin content which is less preferred by birds and less affected by mold (Wortmann et al., 2006). Assessment of glume cover revealed in 29% and 28% of the entries in the landraces collection characterized with ¼ and ½ of the grain covered by glumes, respectively. The proportion of accession with ¾ of the grain covered with glumes (32%) was predominant while the proportion of landraces with grain fully covered with glumes (9%) and glumes longer than grain (1%) was very low. In general, open glume types are predominant than closed glumes suggesting farmers preference towards open types which is highly associated with theshability.

Panicle exsertion was scored into four phenotypic classes, 48% of the accessions were found to have exserted characters followed by well-exserted (35%) (Table 3.4.1). This result partially concurs with Geleta and Labuschagne, (2005) where well-exserted characters were the predominant ones in the Hararghe collection. Rao *et al.* (1996), in their morphological diversity analysis of the Indian germplasm, reported that landraces have poor panicle exsertion as compared to improved varieties. However, the present study found that there are also landraces with well-exserted and exserted panicles.

3.4.2 Shannon-Weaver index of diversity

Understanding of the genetic diversity of germplasm is fundamental to effectively use these genetic resources and to design proper strategies for their conservation (Grenier *et al.*, 2004). The extent of phenotypic diversity which is computed based on Shannon-Weaver diversity index and its partitioning within and between districts is presented in Table 3.4.2. The study showed that all the phenotypic characters considered were highly polymorphic, with the maximum and minimum diversity index scores of 1 and 0.97 being for inflorescence compactness and grain colour, respectively. The overall average phenotypic diversity index of accessions was 0.99 showing high variability with respect to all phenotypic character classes. The accessions also showed very high polymorphism for leaf rolling and stay-green (0.99 each). Stay-green (Xu *et al.*, 2000) and leaf rolling (Hsiao *et al.* 1984) are generally recognized as important traits for plant productivity under post-flowering drought stress

conditions. Hence, the two characters are used as visual scoring criteria for selecting drought resistance in plants. The Shannon-Weaver diversity index values obtained in the present study appear larger than those reported by Geleta and Labuschagne (2005). Unlike this study, they only used 45 genotypes and the low diversity index values may be the result of a small sample size.

Table 3.4.2 Diversity index (H') estimates for 12 phenotypic characters in 262 sorghum accessions, partitioning between and within collection districts

	Diversity index ^a										
Characters	H'	H _{cl}	H _{cl} /H'	(H'-H _{cl})/H'							
Awns	0.99	0.96	0.97	0.03							
Inflorescent compactness	1.00	0.82	0.82	0.19							
Glume colour	0.98	0.89	0.91	0.09							
Glume cover	0.98	0.88	0.90	0.11							
Head shape	0.98	0.88	0.89	0.11							
Leaf colour	0.99	0.93	0.94	0.06							
Leaf orientation	0.99	0.85	0.86	0.14							
Midrib colour	0.98	0.91	0.93	0.08							
Panicle exsertion	0.99	0.82	0.83	0.17							
Grain colour	0.97	0.86	0.89	0.12							
Leaf rolling	0.99	0.76	0.77	0.24							
Stay-green	0.99	0.86	0.87	0.13							
Average	0.99	0.87	0.88	0.12							

^a H'= Diversity index, H_{cl} = Average diversity index of each character for the collection districts, H_{cl}/H' = Proportion of diversity within district, $(H'-H_{cl})/H'$ = Proportion of diversity between districts

The average diversity index (H_{cl}) pooled over the collection districts also showed a range of variation from 0.76 for leaf rolling to 0.96 for awns (Table 3.4.2). A high proportion of diversity was also observed within districts. This is in agreement with the findings of Ayana and Bekele (1998, 1999, 2000) where sorghum accessions collected from different regions of Ethiopia and Eritrea were reported to have high level of variation within the region of origin. However, in the present study, very low diversity was observed between districts (Table 3.4.2) and this may be attributed to the fact that all the accessions were collected from lowland areas, with an altitude of less than 1600 masl, in all the collection districts. The environment might have exerted similar influence on the development of phenotypic characters. Sorghum is grown in a wide range of environmental conditions which include rainfall, temperature, altitude, growing period, and edaphic factors (Stemler *et al.* 1977).

The diversity indices differed among the districts and all districts showed from high to average scores for all the phenotypic characters except Dahina that scored a relatively lower diversity (0.37) for leaf rolling (Table 3.4.3). In this case, the lowest diversity index does not necessarily indicate the low diversity but may be associated with partial sampling of only lowland germplasm. The highest range of diversity across collection districts was observed for leaf rolling and the lowest range for awns (Table 3.4.3). Landraces from Ambasel and Gubalfto for leaf colour, Bugina for stay-green and awns, Habru for leaf rolling, and Sekota for awns exhibited the maximum diversity of unity suggesting that a wide range of variation existed within Districts. Ambsele and Gubalafto are relatively lowland wet areas with an average annual rainfall of 600-1000 mm and growing duration of 110-150 days, whereas Bugina, Habru and Sekota are lowland dry areas with an average annual rainfall of less than 600 mm and growing duration of 90-130 days (Ayana and Bekele, 2000). The maximum variation in stay-green, awns and leaf rolling observed in Bugina, Habru and Sekota, could be attributed by moisture stresses at these environments.

Table 3.4.3 Diversity index estimates of the 12 phenotypic characters in 262 sorghum accessions by nine districts of collection

	District ^b										
Character ^a	Amb	Bug	Dah	Gub	Hab	Kal	Kob	Sek	Ziq	Range	<u>+</u> SE
GRC	0.83	0.83	0.89	0.89	0.89	0.78	0.83	0.93	0.88	0.78-0.93	0.02
GLC	0.83	0.88	0.88	0.90	0.97	0.86	0.88	0.96	0.88	0.83-0.97	0.03
GLV	0.90	0.79	0.81	0.88	0.89	0.87	0.97	0.86	0.95	0.79-0.97	0.02
LO	0.79	0.86	0.99	0.88	0.92	0.79	0.75	0.94	0.76	0.75-0.99	0.06
LC	1.00	0.94	0.94	1.00	0.54	0.98	0.99	0.99	0.99	0.54-1.00	0.01
HS	0.90	0.78	0.74	0.92	0.97	0.88	0.85	0.96	0.88	0.74-0.97	0.01
PE	0.80	0.69	0.76	0.75	0.89	0.79	0.85	0.94	0.92	0.69-0.94	0.01
SG	0.82	1.00	0.72	0.74	0.88	0.78	0.99	0.76	0.99	0.72-1.00	0.02
AW	0.95	1.00	0.99	0.99	0.95	0.95	0.92	1.00	0.92	0.92-1.00	0.01
IC	0.69	0.81	0.72	0.69	0.95	0.70	0.99	0.85	0.95	0.69-0.99	0.06
LR	0.79	0.99	0.37	0.50	1.00	0.81	0.50	0.94	0.92	0.37-1.00	0.07
MDC	0.96	0.92	0.97	0.76	0.95	0.86	0.97	0.89	0.91	0.76-0.97	0.07
Mean	0.86	0.87	0.82	0.83	0.90	0.84	0.87	0.92	0.91		

^a GRC=Grain colour, GLC=Glume colour, GLV=Glume cover, LO=Leaf orientation, LC=Leaf colour, HS=Head shape, PE=Panicle exsertion, SG=Stay-green, AW=Awns, IC=Inflorescent compactness, LR=Leaf rolling, MDC=Midrib colour.

^b Amb=Ambasel, Bug=Bugna, Dah=Dahina, Gub=Gubalfto, Hab=Habru, Kal=Kalu, Kob=Kobo, Sek=Sekota, Ziq=Zequal

3.4.3 Principal component analysis

Principal component analysis was used to identify the characters most important in terms of the variation of the landraces within each of the districts of collection. The first four principal components (PCs) with eigenvalues greater than one, explained 82% of the total variation among the studied landraces for the twelve qualitative characters (Table 3.4.4). About 34% of the total variation accounted for by the first PC alone was due to the contrast between panicle exsertion and the average effects of stay-green, inflorescent compactness and grain colour. In the second PC, leaf rolling and midrib colour had significant contribution. Likewise, the third PC accounted for about 16% of the total variance of the landraces originated mainly from the contrast between glume cover and the average effects of awns and glume colour. Variation in head shape, leaf orientation and leaf colour constituted a large part of the total variation explained by the fourth PC. This result suggested that staygreen, leaf rolling, awns, head shape, panicle exsertion, inflorescence compactness and leaf orientation were important traits in differentiating accessions of different regions. This implies that these traits are vital for the variation in sorghum landraces collected from different regions. Since, this experiment was conducted under water stress conditions, the above mentioned traits are reported to be crucial for moisture stress tolerance (Xu et al., 2000; Tambussi et al., 2007; Khaliq et al., 2008; Samarah et al., 2009; Taketa et al., 2011). Rao et al. (1996) reported that these morphological characters are highly influenced by environmental factors. Ayana and Bekele (1999) also indicated the relevance of environmental factors in affecting the structural variation in sorghum accessions.

The districts were dispersed in all the four quadrants with the majority of the regions were found on the two positive quadrants of the first PC (data not shown). The first PC was much more important in separating the regions than the second PC. The first PC classified Zikuala, Sekota and Kobo in one group with negative principal scores and the rest of the region with positive scores. The extremes of the first axis were occupied by Kobo with low negative principal scores and Bugina and Zikuala with high positive principal scores. On the second PC, Habru had relatively high positive principal scores, while Sekota and Kalu had low negative principal scores. Gubalafto was the closest region to the center of the axis while Habru and Kobo were the farthest from the center, suggesting that accessions from the two regions were relatively divergent from those of the other regions. The clustering of the regions revealed that regions having similar climatic conditions and adjacent regions were grouped together.

Table 3.4.4 Principal component (PC) and discriminant function (DA) scores, eigenvalues, total variance and cumulative variance for 12 qualitative characters in sorghum landraces

Characters	nt	Discriminant function							
	PC1	PC2	PC3	PC4	1	2	3	4	5
Leaf rolling	0.08	0.87	0.20	-0.05	0.57	0.19	0.15	0.24	-0.39
Inflorescent compactness	0.89	-0.10	0.22	-0.12	0.55	-0.34	0.17	-0.31	0.31
Glume cover	-0.05	0.12	-0.90	-0.39	-0.24	0.45	0.31	0.23	-0.32
Glume colour	-0.06	-0.72	0.55	-0.07	0.16	0.41	-0.01	-0.25	-0.21
Leaf orientation	0.10	0.49	0.03	0.63	0.06	0.04	0.55	0.11	0.11
Midrib colour	-0.25	0.86	-0.11	0.01	-0.30	0.02	0.47	-0.02	0.11
Panicle exsertion	-0.72	-0.24	-0.19	-0.02	-0.12	0.22	-0.46	0.11	0.43
Head shape	-0.16	-0.03	0.03	0.94	-0.10	0.27	-0.19	0.62	0.06
Grain colour	0.55	-0.47	0.40	0.03	-0.12	-0.31	-0.14	0.50	0.32
Stay-green	0.95	-0.17	-0.12	0.18	0.42	0.32	-0.11	-0.28	0.54
Awns	0.31	0.11	0.92	-0.07	0.15	-0.27	0.12	-0.13	0.32
Leaf colour	-0.44	0.29	-0.29	-0.69	0.00	0.37	0.21	0.08	0.25
Eigenvalues	4.06	2.40	1.92	1.50	0.21	0.09	0.08	0.06	0.05
% total variance	33.82	20.01	15.98	12.50	38.70	18.00	14.90	10.70	9.70
% cumulative variance	33.82	53.83	69.81	82.31	38.70	56.70	71.60	82.30	92.00

Scatter plots of the 262 landraces using the first two PCs over districts of collection is presented in Figure 3.4.1. Plotting the landraces on the first two PC axes graphically demonstrated that landraces from different districts coverred the PC axes unequally. For example landraces from Ambasele, Bugina, Habru, Kobo, Sekota and Zekuala were unevenly distributed over the four quadrants of the two PCs axes. However, landraces from Kalu and Gubalafto were evenly distributed over the two PCs axes. The differences in the distribution of the landraces may be associated with natural and artificial selection over different climatic condition and use (Doggett 1988; Asante, 1995). Specific patterns were observed for each of the districts. The result showed that high phenotypic diversity in the Wello landraces collection as being differentially distributed among the districts with specific patterns drawn for each district of collection.

3.4.4 Discriminant analysis

Discriminant analysis (DA), using the districts of collection of the landraces as a grouping variable, revealed that 59% sorghum landraces were correctly classified to their respective regions of collection (Table 3.4.5). The regional classification was fully correct for all the nine landraces from Kobo. The percentage of landraces correctly classified was relatively higher for landraces from Dahina and Habru, while it was relatively low for landraces from Gubalafto. On the other hand, the grouping in to regions of collection showed correct

classification for 56%, 59%, 67% and 64% of the landraces from Ambasel, Kalu, Zikuala, Bugina and Sekota, respectively. The intermediate proportion of correct regional (59%) classification observed in this study can be attributed to the higher the diversity of the group (Pecetti and Damania, 1996). It was also hypothesis that the higher the diversity of the group, the higher is the probability of misclassification and vice versa (Holcomb *et al.*, 1977).

The DA also determines the contributions of one or more categorical predictor variables in the diversity assessment of accessions. It is a useful procedure when the variables are orthogonal to each other. In this study, five discriminant functions were identified that explained 92% of the variation among the landraces and districts of collection (Table 3.4.4). The first function that accounted for 39% of the total variation was found to be well correlated with leaf rolling and inflorescence compactness. In the second discriminant function, glume colour and cover were the most important variables that explained 18% of the total variations. The third and fourth discriminant functions were explained about 26% of the total variation and well correlated with leaf orientation and midrib colour; and head shape and grain colour, respectively. Likewise the fifth discriminant function was highly correlated with stay-green. From the analysis it is clear that leaf rolling, stay-green, inflorescence compactness, midrib colour, head shape, grain colour, glume cover and glume colour were orthogonal. This implies that these characters can be used for effective planning of future germplasm collection.

The dendrogram showing similarities of the nine districts of collection of 262 landraces of sorghum evaluated for 12 qualitative characters was presented in Figure 3.4.2. The dendrogram clearly showed that the close relationship between landraces from Gubalafto, Sekota, and Ambasel; Kalu and Zekuala; Bugina and Habru. However, landraces from Kobo remained distinct and ungrouped. The cluster analysis thus confirmed the distinctiveness of the accessions from Kobo from the accessions from the other regions as a whole, which was partially displayed by the bi-plot analysis of the principal components (data not shown).

Table 3.4.5 Summery of discriminant analysis for 262 landraces by region of collection

Region		No. entries	Predicted classification										
			1	2	3	4	5	6	7	8	9	10	% correct
1	Ambasel	63	35	2	1	12		9		2	1	1	56
2	Bugina	14	1	9	2	2							64
3	Dahina	14			13	1							93
4	Gubalafto	63	14	1	4	26	1	16		1			41
5	Habru	8	1				7						88
6	Kalu	68	9	3	4	8	1	40	1	2			59
7	Kobo	9							9				100
8	Sekota	14	2		1	1		1		9			64
9	Zikuala	9	2							1	6		67

3.5 Conclusion

The results suggest that very high diversity exists for all the 12 phenotypic characters studied. The proportion of total diversity obtained among the collection districts was less than that of within collection regions. It would be possible to make selection for any of the traits within a particular region, presuming that a significant portion of the phenotypic variation is genetic. The diversity available in the studied germplasm allows future exploitation by sorghum improvement programmes. It was also found that the landraces showed a wide range of variation for stay-green and leaf rolling characters useful in sorghum breeding towards drought stress tolerance. From a conservation point of view, examining the way in which genetic diversity within a species is distributed among its populations and regions of origin needs to be considered (Gray, 1996). In this study, four PCs that explained 82% of the total variation and five discriminant functions that explained 92% of the variation in the landraces/districts of collection were identified. Characters, such as stay-green, leaf rolling, head shape, inflorescence compactness and leaf orientation were important traits in differentiating landraces of different districts. Landraces from Kobo were found to be relatively divergent from the rest of the districts and they can be used as a good sources of genes.

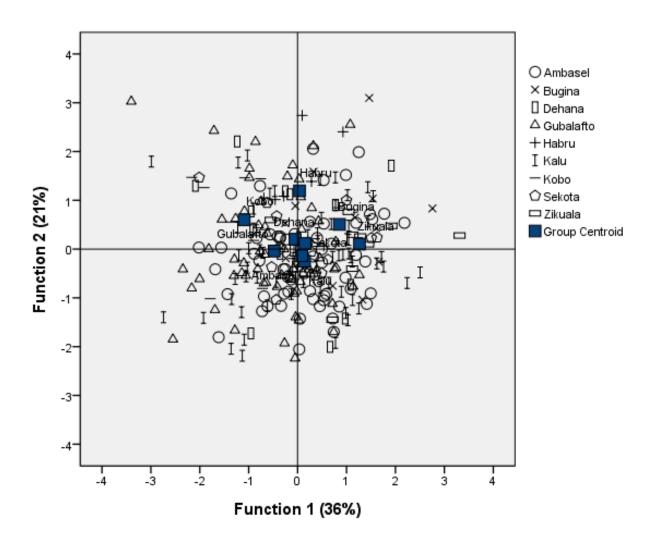


Figure 3.4.1 Scatter plots of landraces collected from nine districts using the first two PCs

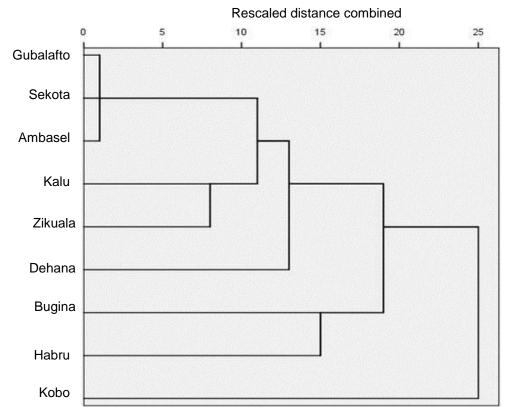


Figure 3.4.2 Dendrogram showing similarities of the nine districts of collection of 262 sorghum landraces evaluated for 12 qualitative characters

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CHAPTER 4

Genetic Diversity Assessment among Lowland Sorghum [Sorghum bicolor (L.) Moench] Landraces under Moisture Stress Environments using Quantitative Traits

4.1 Abstract

A total of 273 sorghum (Sorghum bicolor [L.] Moench) landraces collected from nine districts of north and north eastern Ethiopia were evaluated under moisture stress conditions to assess the extent of morphological variations inter- and intra-districts. Components of genetic variance, broad sense trait repeatability and phenotypic correlations were estimated for ten quantitative characters. Landraces revealed highly significant variation for all characters under study. The extent of variation was highly influenced by location and moisture stress condition. Landraces collected from drought prone districts showed low coefficient of variation (6.2 to 13.4%) for several characters, indicating comparatively high intra-district uniformity. High levels of trait repeatability values were recorded for days to 50% anthesis at 77%, panicle length (65%) and plant height (71%). Repeatability estimates of traits were influenced by the environment and genotype by environment interaction. Positive and highly significant correlation was exhibited between grain yield and head weight. Grain yield and head weight were negatively correlated with days to 50% flowering, days to 50% maturity and grain filling duration. The study identified marked morphological variants among collections of sorghum landraces from north and north eastern Ethiopia. Landraces including 239167-B, 244725-A, 242049-B, 242050-A, 244711-A, 202508, 244729-A, 242046, 75454, 73056-B and 72457 were identified as promising sources of drought resistance genes for sorghum improvement in the north and north eastern Ethiopia and other similar environments.

Keywords: Ethiopia, genetic variance, trait repeatability, Sorghum bicolor; Wello

4.2 Introduction

Sorghum (*Sorghum bicolor* L. Moench; 2n=2x=20) is the most important cereal crop worldwide after wheat, rice, maize, and barley (FAO, 1995, Poehlman, 1994). The crop has wide adaptation but its production has been limited due to water- and heat-stresses within subtropics and tropics. Sorghum is the main staple food crop in Ethiopia, currently ranks third after tef and maize in total production; ranks second after maize in productivity per hectare and in area harvested (CSA, 2012). It serves as an important source of income for small scale farmers because of its wide range of uses. Sorghum is mainly grown for its grain as food for humans and livestock feed and for production of local beverages, while the stalk is also used for animal feed and as a construction material (Teshome *et al.*, 1999; McGuire, 2000).

Ethiopia is the center of genetic diversity of many cultivated plants, including sorghum (Vavilov, 1951; Harlan, 1969). A large number of accessions (~14,000) have been collected by the joint efforts of the Ethiopian Sorghum Improvement project (ESPI) and the Institute of Biodiversity Conservation (IBC) (Gebrekidan 1973; Kebede, 1986, Teshome *et al.*, 2001). The country's wide environmental variability has an important implication of the diversity of plants. Sorghum is one of the most diverse crops distributed over a wide range of agroecologies in this country. Although most of the sorghum accessions are landraces, several wild, weedy, and intermediate forms of sorghum accessions are found (IBC, 1995).

Breeding for drought resistance requires sources of resistance to drought (Terán and Singh, 2002). Landraces are highly adapted to specific environmental conditions and are useful sources of genetic variation (Brown and Munday, 1982). The heterogeneity and heterozygosity nature of tropical landraces of sorghum enables to display high adaptation to drought stress (Rao *et al.* 2002; Habyarimana *et al.*, 2004; Dar *et al.*, 2006). Assessment of genetic variability in crops has strong impact on crop improvement programmes, and conservation of genetic resources (Assar *et al.*, 2005). Several previous studies on sorghum germplasm of Ethiopia have demonstrated the existence of wide variation in many phenotypic traits (Teshome *et al.*, 1997; Ayana and Bekele, 1998, 1999, 2000; Ayana *et al.*, 2000; Abdi *et al.*, 2002; Geleta and Labuschagne, 2005).

In Ethiopia, the prevalence of drought has increased from every five to six years to every three years in the past two decades. In the past, when the rainfall situation was relatively better, farmers used to grow different late maturing sorghum landraces that could be planted in April/May. While in times of delayed rainfall onset and a short rainy season, farmers switch to growing early maturing sorghum landraces. However, due to the prevalent shift in

the rainfall condition, farmers have completely changed from growing highly productive late maturing sorghum landraces to growing either early maturing sorghum varieties or other crops (Assefa et al. unpublished). Moreover, currently there is a very narrow range of short-duration varieties of sorghum which, in turn, limits farmers' choices and marks genetic erosion of local landraces. Early maturing landraces are generally of low productivity as compared to the late maturing sorghum landraces. The advantage of landraces, however, is that they perform well under moisture stress; they are best adapted to the local conditions; and they have farmers' preferred attributes, despite their low productivity (Rao *et al.*, 1996; Ayana and Bekele, 2000; Grenier *et al.*, 2004; Ali *et al.*, 2009).

Breeding for drought resistance requires agronomically suitable germplasm with adequate resistance (Terán and Singh, 2002). Wild species, traditional varieties, commercial cultivars, and breeding lines are used as sources of gene for drought resistance in most breeding programmes. Dar *et al.* (2006) indicated the importance of traditional germplasms as a source of resistance genes for drought in the semi-arid tropics where moisture stress is the greatest challenge for crop improvement. Similarly, Habyarimana *et al.* (2004) indicated that the effect of heterogeneity and heterozygosity of tropical landraces of sorghum enables to display high adaptation to drought stress. Although sorghum originated in Africa, it spread across wide geographic areas covering a wide range of latitude and longitude, altitude, day length, rainfall, and temperature regimes. Consequently, it adapts to a range of biotic and abiotic stress factors. This has resulted in the evolution of several landraces cultivated in their respective sub-districts (Rao *et al.*, 2002). Blum (2004) also indicated that sorghum is a warm-season and photoperiod sensitive grass that is characterized more by diversity than homogeneity.

Sorghum is an important crop in the lowland areas of Ethiopia where rainfall is unreliable and recurrent drought is common (Kebede and Menkir, 1987). Its adaptation to drought stress environments makes sorghum an important cereal crop in Ethiopia and worldwide (Doggett, 1988, House, 1995). The Ethiopian Sorghum Improvement Programme (ESIP) made a major collection of landraces during 1973 in the north eastern region where the largest diversity is found (Gebrekidan, 1975). In this region drought stress is a major crop production constraint. Although this region has the largest diversity of sorghum and the enormous potential of landraces recognized nationally and globally, assessment of the level of genetic diversity within and between water stressed environments is limited. The objectives of this study were to assess the extent of phenotypic and genetic diversity within and between the districts of collection, evaluate the performance of landraces under

moisture stress condition, estimate genetic variance, trait repeatability and phenotypic correlations using quantitative traits in sorghum.

4.3 Materials and Methods

4.3.1 Plant material

Out of the total collection of sorghum in Ethiopia, 273 landraces of lowland origin were used for this study. Landraces were collected by the Ethiopian Institute of Biodiversity Conservation (IBC) across altitudes ranging from 900-1540 masl. The landraces were collected from three Zones and nine Districts from northeastern Amhara region (Wello). Sixty seven landraces were included from Ambasel, Kalu (74), Kobo (9), Bugina (14), Gubalafto (64), Habru (8), Dahina (14) Zikuala (9), and Sekota (14). Five other entries were included as comparative controls. Of the five, three were improved cultivars acquired from the sorghum improvement programme of the Sirinka Agricultural Research Center (SARC), one local and one introduced variety developed for drought resistance by the International Crops Research Institute for the Semi-Arid Tropics (ICRISAT).

4.3.2 Field experiments and data collection

Field experiments were conducted at Sirinka Agricultural Research Center under rainfed and Kobo sub-center under both rainfed and irrigation. Sirinka and Kobo research sites are situated at 11°45' and 12°9' N latitude and 39°36' and 39°38' E longitude with an altitude of 1850 and 1400 masl, respectively. Data on rainfall pattern, pan evaporation, relative humidity, and maximum/minimum temperatures for the two sites is indicated in Table 4.4.1. Both locations have a bimodal rainfall pattern with a short rainy season (belg) from March to April and long rainy season (meher) from June to mid of September. The total rainfall received during the crop growing period at Sirinka was 886.1 mm while Kobo received 616.5 mm. One can question the drought proneness of the areas by simply viewing the rainfall figures. At both sites, the maximum rainfall is received during July and August, in those months the rain is very intensive and wasted through runoff. After September the rain becomes severely depilated. Additionally, there were 60 and 52 rainy days in Sirinka and Kobo, respectively. Growing period, for long cycle sorghum varieties, ranges from mid-July to mid-December and anthesis starts after mid-September. Growing period for short cycle varieties, on the other hand, stretches from mid-July to October. Consequently, sorghum accessions grown under rainfed conditions, at both sites, normally experience moisture stress at anthesis and post-anthesis growth stages. Additionally, to insure the genotypes experience moisture stress, the stress experiment was planted three weeks after the control exeperiment.

Table 4.4.1 Mean monthly metrological data at Sirinka and Kobo/Ethiopia during the last ten years (2000-2010)^a

RF 16.14	PEV	MNT	NA)/T				Kobo						
16.14			MXT	RH	RF	PEV	MNT	MXT	RH				
_	4.90	12.51	26.51	61.10	34.25	4.71	11.53	22.79	71.77				
7.12	6.53	13.20	28.97	51.55	35.43	5.78	12.43	25.25	65.27				
33.97	6.89	14.35	30.87	49.00	87.68	6.21	13.47	25.97	64.95				
60.31	7.29	14.80	31.80	47.36	166.05	6.81	14.83	25.88	60.44				
38.36	7.90	16.77	33.61	38.55	38.53	7.26	15.73	29.54	50.87				
16.09	9.15	19.09	34.84	29.80	29.59	8.79	16.72	30.80	45.08				
169.53	5.89	18.52	32.19	48.73	237.36	6.08	15.65	28.79	65.24				
223.66	6.05	17.57	30.96	58.64	297.80	5.90	14.86	27.20	66.59				
58.39	4.74	16.09	30.64	57.55	84.95	5.58	14.06	27.15	63.35				
36.99	5.65	13.68	29.93	48.64	69.16	5.45	12.29	25.95	60.65				
19.32	6.03	12.16	28.80	46.64	37.41	5.28	11.86	24.35	58.79				
16.85	5.64	11.90	27.37	52.20	30.82	5.66	10.77	23.05	67.86				
	33.97 60.31 38.36 16.09 169.53 223.66 58.39 36.99 19.32	33.97 6.89 60.31 7.29 38.36 7.90 16.09 9.15 169.53 5.89 223.66 6.05 58.39 4.74 36.99 5.65 19.32 6.03	33.97 6.89 14.35 60.31 7.29 14.80 38.36 7.90 16.77 16.09 9.15 19.09 169.53 5.89 18.52 223.66 6.05 17.57 58.39 4.74 16.09 36.99 5.65 13.68 19.32 6.03 12.16	33.97 6.89 14.35 30.87 60.31 7.29 14.80 31.80 38.36 7.90 16.77 33.61 16.09 9.15 19.09 34.84 169.53 5.89 18.52 32.19 223.66 6.05 17.57 30.96 58.39 4.74 16.09 30.64 36.99 5.65 13.68 29.93 19.32 6.03 12.16 28.80	33.97 6.89 14.35 30.87 49.00 60.31 7.29 14.80 31.80 47.36 38.36 7.90 16.77 33.61 38.55 16.09 9.15 19.09 34.84 29.80 169.53 5.89 18.52 32.19 48.73 223.66 6.05 17.57 30.96 58.64 58.39 4.74 16.09 30.64 57.55 36.99 5.65 13.68 29.93 48.64 19.32 6.03 12.16 28.80 46.64	33.97 6.89 14.35 30.87 49.00 87.68 60.31 7.29 14.80 31.80 47.36 166.05 38.36 7.90 16.77 33.61 38.55 38.53 16.09 9.15 19.09 34.84 29.80 29.59 169.53 5.89 18.52 32.19 48.73 237.36 223.66 6.05 17.57 30.96 58.64 297.80 58.39 4.74 16.09 30.64 57.55 84.95 36.99 5.65 13.68 29.93 48.64 69.16 19.32 6.03 12.16 28.80 46.64 37.41	33.97 6.89 14.35 30.87 49.00 87.68 6.21 60.31 7.29 14.80 31.80 47.36 166.05 6.81 38.36 7.90 16.77 33.61 38.55 38.53 7.26 16.09 9.15 19.09 34.84 29.80 29.59 8.79 169.53 5.89 18.52 32.19 48.73 237.36 6.08 223.66 6.05 17.57 30.96 58.64 297.80 5.90 58.39 4.74 16.09 30.64 57.55 84.95 5.58 36.99 5.65 13.68 29.93 48.64 69.16 5.45 19.32 6.03 12.16 28.80 46.64 37.41 5.28	33.97 6.89 14.35 30.87 49.00 87.68 6.21 13.47 60.31 7.29 14.80 31.80 47.36 166.05 6.81 14.83 38.36 7.90 16.77 33.61 38.55 38.53 7.26 15.73 16.09 9.15 19.09 34.84 29.80 29.59 8.79 16.72 169.53 5.89 18.52 32.19 48.73 237.36 6.08 15.65 223.66 6.05 17.57 30.96 58.64 297.80 5.90 14.86 58.39 4.74 16.09 30.64 57.55 84.95 5.58 14.06 36.99 5.65 13.68 29.93 48.64 69.16 5.45 12.29 19.32 6.03 12.16 28.80 46.64 37.41 5.28 11.86	33.97 6.89 14.35 30.87 49.00 87.68 6.21 13.47 25.97 60.31 7.29 14.80 31.80 47.36 166.05 6.81 14.83 25.88 38.36 7.90 16.77 33.61 38.55 38.53 7.26 15.73 29.54 16.09 9.15 19.09 34.84 29.80 29.59 8.79 16.72 30.80 169.53 5.89 18.52 32.19 48.73 237.36 6.08 15.65 28.79 223.66 6.05 17.57 30.96 58.64 297.80 5.90 14.86 27.20 58.39 4.74 16.09 30.64 57.55 84.95 5.58 14.06 27.15 36.99 5.65 13.68 29.93 48.64 69.16 5.45 12.29 25.95 19.32 6.03 12.16 28.80 46.64 37.41 5.28 11.86 24.35				

^a RF= rain fall (mm); PEV= pan evaporation (mm), MNT= minimum temperature (°C), MXT= maximum temperature (°C), RH= relative humidity (%)

At Kobo supplementary irrigation was applied every week from planting to grain filling stage. Non-stress plots received 10 supplementary irrigations (proximately 250 mm of water) 4 days before and 6 days after anthesis. This experiment was used as a control and the supplementary irrigation was sufficient for normal crop growth and development. The supplementary water requirement was calculated using CWR = KC x EVT, where CWR is crop water requirement, KC is crop coefficient determined at different growth and developmental stages and EVT is evapo-transpiration rate. The 278 accessions were grown in a single row of 3 m long at a seed rate of 10 kg ha⁻¹ and spacing of 0.75 m and 0.3 m between row and plants, respectively. The five control varieties were planted in each block. The experiment was carried out in an unbalanced incomplete block design of 10 blocks and 32 plots within each block in two replications. Diammonium phosphate (DAP) fertilizer was applied at the rate of 100 kg ha⁻¹ and urea at 50 kg ha⁻¹ as recommended for sorghum in the lowlands of Ethiopia. All DAP was applied at the time of planting while urea was applied in a split application. Other agronomic practices were applied uniformly to all the treatments.

For every accession in a plot, five individual plants were sampled and used for recording the data, except days to 50% anthesis and maturity, which was recorded on plot basis. The following parametric characters were studied: days to 50% anthesis (DTA), days to 50% maturity (DTM), plant height (PH), panicle length (PL), leaf length (LL), leaf width (LW), leaf number (LN), grain yield (GY), and head weight (HW). Additionally, Grain filling duration (GFD) was calculated by subtracting number of days to maturity from days to anthesis. Drought tolerance was quantified using stability performance test under drought stress conditions as described by Tuinstra *et al.* (1997). Yield stability (YS) was calculated as proportion of grain yield under stress to grain yield under full irrigation conditions.

4.3.3 Data analysis

Data on the 278 genotypes, grown at the two locations and two water treatments was subjected to statistical analysis using GenStat computer software, GenStat for Windows 11th Edition (Payne *et al.*, 2008). Analysis of variance was performed for the 11 quantitative characters. Homogeneity of the error among the two locations and the two water treatments was examined using Bartlett's test for each of the studied quantitative characters. The test showed that days to 50% flowering, days to 50% maturity, grain filling duration, grain yield, head weight, leaf length, leaf width and panicle length have homogenenous of varainces. For the other two characters i.e. number of leaves and plant height, failed to meet the assumptions of equality of variances for which logarithmic data transformation was performed. Phenotypic correlations between the characters were assessed based on means using the Pearson correlation procedure.

The two water treatments and locations were treated as environments. Genotypes were considered as fixed effect while replication and block were considered as random effects. The expected mean squares were estimated from the analysis of variance and the genetic parameters were estimated as indicated by Johnson *et al.* (1959). Trait repeatability estimates were calculated from the genetic parameters. Out of the 273 sorghum landraces characterized and evaluated five, ten and two landraces failed to grow at Sirinka rainfed, Kobo rainfed, and Kobo irrigation conditions, respectively. About 35% of the failures were from Ambasel, 29% from Kalu, 18% from Gubalafto, 12% from Dahina, and 6% from Sekota. Landrace 239179-A was unsuccessful in both locations and water treatments. Some landraces failed to reach maturity in both locations and water treatments and data were excluded from statistical analysis.

4.4 Results and discussion

4.4.1 Variation and variance components

Analyses of variance per test location and water regime revealed highly significant differences ($p \le 0.01$) among landraces for the all characters suggesting the existence of considerable variation. Thus a combined analysis of variance was conducted over two locations and two water regimes that showed significant genotype by location and genotype by water regime interactions for all traits (Tables 4.4.2 and 4.4.3). The effect of location was not significant on DTA and LW (Table 4.4.1). The analysis on water regimes showed a non-significant effect on LW (Table 2). Yield stability indices were significantly different among genotypes ($p \le 0.01$) (Tables 4.4.2 and 4.4.3). Ceccarelli *et al.* (1991) indicated that genetic differences in yield and yield stability under stress conditions are associated with differences in morphological and developmental traits. It is also evidenced that the overall yield performance of genotypes is determined by the interaction of traits rather than the expression of individual traits. Blum (1988) reported that yield by itself is not under direct genetic control since it is the integrated effect of the multitude of physiological and biochemical processes.

The magnitude of variance components are summarized in Table 4.4.4. Analysis across the two water regimes indicated that traits viz. DTA, DTM, LN, PH and PL had the highest genotypic variances estimated at 5.7, 16.5, 0.3, 143.4 and 2.59, respectively, while LL showed the least (Table 4.4.4). On the other hand, GFD, LW, LL, HW and GY revealed the highest genotype by water regime interaction variances (Table 4.4.4). DTA and LN were not affected by genotype and water regime interactions. Trait repeatability was the highest for DTA (77%) and LN (67%) followed by PL, PH and DTM (Table 4.4.4).

Across locations, characters such as DTA, DTM, LN, PH and PL exhibited the highest genotypic variance, whereas GFD, HW, LW and GY showed the lowest. Conversely, GFD, HW, GY, LL and LW were highly affected by genotype and location interactions. PH with 71%, DTA (66%) and PL (60%) expressed the highest repeatability followed by LN (48%) and DTM (45%). Generally, for most of the characters studied under rainfed condition, the repeatability estimates of the characters were relatively low (Table 4.4.3). Ten year data on mean monthly rainfall, pan evaporation, relative humidity, and maximum/minimum temperatures for the two locations is indicated in Table 4.4.1.

Table 4.4.2 Mean, coefficient of variation (CV), standard error (SE), range and mean squares on 10 quantitative traits of sorghum landraces evaluated at two locations^a.

Entry					Tra	its ^b					
-	DTA	DTM	GFD	LN	LW	LL	PH	PL	GY	HW	YS
ICSV 111	70.5	120.0	49.5	8.0	7.7	53.9	141.8	22.3	2564.0	80.3	2.0
Girana-1	72.3	121.2	48.9	8.9	8.4	67.1	178.0	20.8	2738.0	89.0	2.3
Misikir	70.3	124.3	53.9	8.9	7.9	63.5	154.2	21.6	2802.0	86.4	2.3
72458	76.9	126.2	49.3	8.6	5.7	54.3	181.7	20.4	1469.0	52.1	0.4
244725-a	76.8	127.1	50.4	10.2	8.1	72.8	199.7	17.8	1637.0	70.6	1.1
72447	72.7	127.1	54.5	9.3	7.7	57.8	135.5	22.7	1774.0	58.8	1.6
72472	78.6	127.2	48.5	9.8	7.2	68.6	186.4	23.6	1200.0	57.1	2.3
244711-a	84.0	127.6	43.6	10.8	7.3	63.5	168.7	22.6	1307.0	59.0	0.8
242049-a	79.9	127.8	48.0	9.9	8.3	69.5	223.1	24.1	2072.0	65.2	1.0
202508	77.8	127.9	50.1	10.2	8.1	60.5	210.7	24.4	1675.0	65.9	2.7
239167-b	80.2	128.1	48.0	11.9	8.8	75.2	241.8	28.7	1660.0	88.4	1.2
Raya	77.7	128.1	50.4	9.1	8.6	69.1	167.3	20.1	2913.0	95.8	1.8
239167-a	76.4	128.7	52.3	9.5	7.9	66.7	178.9	28.0	1998.0	85.6	0.0
239208	82.1	128.9	46.8	12.4	8.5	72.5	226.0	21.4	1676.0	80.3	0.3
244715	76.3	129.4	53.1	11.4	7.4	71.0	250.7	22.9	2352.0	82.2	3.7
214855	78.9	129.6	50.7	10.5	9.0	67.7	195.4	15.0	2455.0	95.7	1.9
242046	78.9	129.6	50.7	11.1	7.8	70.4	209.8	23.7	1552.0	64.2	1.6
239188	86.3	129.9	43.6	10.8	8.0	77.9	227.3	31.3	1781.0	86.6	0.9
244722	86.7	129.9	43.3	12.9	7.4	74.5	191.2	24.9	1729.0	72.1	1.0
72491-a	77.9	129.9	52.1	11.3	7.7	69.8	248.6	23.8	2246.0	83.6	1.0
242050-a	78.2	130.2	52.0	11.1	8.2	68.7	250.9	25.4	2037.0	84.5	2.0
239170	80.0	130.4	50.4	8.9	8.2	69.2	210.8	21.1	1581.0	85.1	2.0
242050-b	77.9	130.4	52.5	10.8	9.2	66.3	221.2	20.7	1028.0	78.9	2.1
244725-b	79.9	130.4	50.5	12.4	7.5	65.1	204.9	13.8	2598.0	73.1	3.5
244711-b	79.5	130.9	51.4	10.3	7.4	68.1	216.8	19.7	1524.0	58.3	3.2
244729-a	80.2	131.2	51.0	9.0	8.0	65.5	209.2	22.1	1568.0	62.0	0.6
244727	81.2	131.3	50.2	10.8	7.8	64.9	214.3	22.3	2345.0	90.2	1.2
242049-b	78.3	131.5	53.2	11.7	7.9	70.0	250.6	24.2	2490.0	88.3	3.7
211239-a	76.7	132.0	55.3	10.3	8.9	65.4	223.5	29.1	2093.0	82.9	0.9
244720	78.2	132.2	54.0	9.9	9.1	65.3	135.1	25.0	2190.0	92.0	1.4
No. entry	271	271	271	271	271	271	271	271	264	264	264
CV (%)	5.03	3.40	8.10	8.40	8.40	8.50	9.10	6.90	7.40	7.80	14.4
Mean + SE	90 <u>+</u> 4.5	143.6 <u>+</u> 4.9	53.6 <u>+</u> 4.3	12.8 <u>+</u> 1	8.4 <u>+</u> 0.7	71.0 <u>+</u> 6	208.2 <u>+</u> 19	20.8 <u>+</u> 1.4	1709 <u>+</u> 125.4	83.4 <u>+</u> 6.4	1.5 <u>+</u> 0.21
Range	60 -117	105 -185	20 - 84	6 - 28	5 - 14	12 - 96	90 - 375	3 - 39	209 - 4140	31 - 215	0 - 7
Genotype MS	504.86**	765.54**	245.63**	16.6**	1.9**	166.3**	6438.8**	125.3**	2211746**	1668.7**	3.5**
Location MS	0.01	3.75**	3.32**	0.09**	0.03	0.15**	3.5**	0.02**	18021.2**	13.9**	-
GxL MS	4.65**	11.13**	13.75**	0.2**	0.05**	2.6**	46.4**	1.5**	99137.2**	87.7**	

a data showing 10% of the tested landraces, ^b DTA= days to 50% anthesis, DTM= days to 50% maturity, GFD= grain filling duration, HW= head weight, GY= grain yield, LN= number of leaves, LW= leaf width, LL= leaf length, PH= plant height, PL=panicle length, YS= yield stability * and ** denote significant differences at 0.05 and 0.01 probability levels, respectively.

Table 4.4.3 Mean, coefficient of variation (CV), standard error (SE), range and mean squares on 10 quantitative traits of sorghum landraces evaluated at two water regimes^a.

						traits ^b					
Entry	DTA	DTM	GFD	LN	LW	LL	PH	PL	GY	HW	YS
ICSV 111	71.57	111.4	39.86	8.18	7.884	67.8	147.3	23.21	3491	99.91	1.009
Girana-1	73.02	113.3	40.27	9.45	8.917	73.43	186.3	20.07	3079	104.63	1.626
Misikir	71.18	116.8	45.62	9.31	8.472	70.11	154.9	21.9	3218	102.98	1.565
239167-a	80.24	119.6	39.35	10.43	8.317	71.77	187.4	30.03	1803	106.64	1.491
75460	79.07	120.2	41.15	11.4	8.406	70.02	248.5	16.72	2027	74.32	2.048
244729-a	76.73	120.3	43.61	9.18	6.893	66.95	228.3	19.19	1430	64.05	0.721
244711-a	86.01	121.5	35.47	9.92	7.201	65.49	180	21.86	1015	76.31	1.325
244725-a	79.16	121.6	42.46	10.91	8.046	75.43	222.7	19.56	2446	95.78	0.569
242049-a	82.26	122.1	39.86	11.22	8.964	76.17	241.3	23.87	2079	96.56	0.997
73056-b	79.94	122.2	42.3	11.48	9.077	73.7	227.2	21.71	1760	93.87	1.91
242049-b	80.55	122.8	42.21	11.46	8.383	73.94	264.2	25.43	3169	122.34	1.705
202508	79.39	123.5	44.13	12.19	8.407	62.88	225.3	24.45	2566	87.69	0.967
242046	81.8	123.5	41.67	11.28	8.238	74.24	223.1	23.39	1876	89.78	1.04
244733	80.3	123.5	43.18	11.8	8.889	75.75	255.1	18.77	3510	120.3	0.805
244732-a	81.87	123.8	41.9	11.54	7.848	64.9	200.1	13.35	3088	112.22	0.688
244725-b	77.22	124	46.81	11.01	7.638	62.6	221.8	15.1	2468	109.61	4.227
72491-a	81.85	124	42.2	10.74	8.602	76.46	263.1	24.1	2093	92.46	2.433
72458	77.65	124.2	46.51	9.15	6.492	63.83	193.4	21.28	1268	65.9	3.692
237260	81.74	124.9	43.2	12.04	8.737	72.61	255.4	23.52	3081	103.67	1.225
242039-b	80.74	125.2	44.5	13.08	7.91	66.26	221.2	13.72	1985	117.16	2.69
73092	85.05	125.5	40.4	12.52	8.136	73.85	252.8	19.76	2846	129.14	2.556
239192-b	83.77	125.6	41.84	12.85	8.825	73.44	256.3	14.5	2371	98.26	2.469
73088-b	79.81	125.8	45.95	11.21	8.758	74.88	236.3	16.57	2002	127.77	1.988
244711-b	81.4	125.9	44.46	11.24	7.539	68.79	220.3	21.07	1739	82.95	2.089
Jigurty	80.68	126.4	45.72	11.65	8.394	69.67	273.2	24.64	2481	118.89	1.669
239208	84.94	126.6	41.68	13.42	8.273	72.21	250.3	21.49	2690	84.85	0.574
244712	80.47	126.7	46.25	12.08	7.956	70.18	218.4	12.89	2789	111.08	1.254
244727	82.82	126.7	43.84	11.01	8.208	69.59	246.7	22.06	2083	89.47	1.522
239170	81.24	126.8	45.6	10.79	8.653	74.68	230.3	20.67	2456	84.22	0.89
73045-c	78.95	127.4	48.45	10.44	6.892	64.29	222.9	25.09	1892	71.81	1.049
No. entry	272	272	272	272	272	272	272	272	266	266	266
CV (%)	4.3	3.1	8.0	10.9	8.2	8.8	5.4	6.2	6.1	6.9	10
Mean + SE	87.4 <u>+</u> 3.8	138.3 <u>+</u> 4.3	50.9 <u>+</u> 4.1	12.8 <u>+</u> 1.4	8.4 <u>+</u> 0.7	73.9 <u>+</u> 4.0	223.9 <u>+</u> 19.8	21 <u>+</u> 0.3	1907 <u>+</u> 132.5	89.3 <u>+</u> 5.4	1.16 <u>+</u> 0.1
Range	68 -112	109 -180	20 - 85	27 - 215	6 - 28	42 - 94	90 - 375	3 - 37	27 - 215	209 - 4639	0 - 9
Genotype MS	270.0**	881.4**	312.2**	17.5**	1.9**	277.1**	4265.7**	134.9**	2192.4**	3425351**	2.5**
Water regime	6149.7**	1558.7**	1516.3**	395.9**	1.2	5323.8**	166517.3**	110.9**	4309.6**	14562257**	=
GxE	26.9**	204.41**	169.4**	3.5**	1.3**	58.7**	1372**	29.5**	1097.5**	1592932**	

^a data showing 10% of the tested landraces, ^b DTA= days to 50% flowering, DTM= days to 50% maturity, GFD= grain filling duration, HW= head weight, GY= grain yield, LN= number of leaves, LW= leaf width, LL= leaf length, PH= plant height, PL=panicle length, YS= yield stability * and ** denote significant differences at 0.05 and 0.01 probability levels, respectively.

Table 4.4.4 Variance components and trait repeatability values for 10 quantitative characters in 278 sorghum landraces after evaluation in two water regimes and two locations

		Combined	over water i		Combined over locations ^b						
Characters ^a	σ_{p}^{2}	σ_{g}^{2}	σ² _e	σ^2_{ge}	R (%)	σ^2_p	σ_{g}^{2}	σ_{e}^{2}	$\sigma^2_{ m ge}$	R (%)	
DTA	7.48	5.72	1.12	0.63	76.51	13.95	9.27	0.01	4.67	66.42	
DTM	26.03	16.47	0.25	9.31	63.28	27.26	12.38	3.75	11.13	45.39	
GFD	11.03	3.15	0.24	7.63	28.58	18.74	1.67	3.32	13.75	8.93	
HW	80.62	26.63	0.60	53.40	33.03	105.73	4.18	13.88	87.67	3.96	
GY	126575.67	45371.83	2434.54	78769.30	35.85	122102.68	4944.33	18021.21	99137.15	4.05	
LN	0.45	0.30	0.07	0.08	67.05	0.56	0.27	0.09	0.20	48.03	
LW	0.04	0.00	0.00	0.04	8.13	0.05	0.00	0.00	0.05	0.40	
LL	4.07	0.97	0.96	2.14	23.78	3.79	1.06	0.15	2.58	27.93	
PH	222.76	143.39	30.29	49.09	64.37	169.64	119.73	3.48	46.44	70.57	
PL	4.00	2.59	0.01	1.39	64.84	3.79	2.27	0.00	1.53	59.79	

^a DTA= days to 50% anthesis, DTM= days to 50% maturity, GFD= grain filling duration, HW= head weight, GY= grain yield, LN= number of leaves, LW= leaf width, LL= leaf length, PH= plant height, PL=panicle length,

R = trait repeatability

Both locations have a bimodal rainfall pattern with a short rainy season (Belg) from March to April and long rainy season (Meher) from June to mid of September. The Kobo site experiences high temperature (15 - 31°C) than Sirinka (14 - 26 °C) (Table 4.4.1). Plant growth is a function of the genotype, the environment, and the interaction of the two. The genotype and the environment components are recognized as the primary sources of variability in agronomic and genetic studies (Ceccarelli *et al.*, 1991). The environmental factors consist of location, year, and growing-season. Temperature and rainfall are the most crucial weather factors that determine actual sorghum yield and the two factors accounted for more than half of the environmental and G x E variances (Saeed *et al.*, 1984). Stress environments cause a reduction in the repeatability of yield and other yield components. Such environments are usually characterized by larger environmental variance than non-stressed environments which would increase the challenges in relating phenotype with genotype (Ceccarelli *et al.*, 1991).

^b σ_p^2 = phenotypic variance, σ_g^2 = genotypic variance, σ_e^2 = environmental variance, σ_{ge}^2 = variance for genotype x environment,

4.4.2 Variation within and among collection sites

Landraces demonstrated a wide range of variation between and within the districts of collection (Table 4.4.5). Landraces from Ambasel showed a wider range of variation for DTA (69 - 111 days), DTM (111- 182 days), GFD (24 - 82 days), LN (7 - 28) and HW (27 - 15 gm) (Table 4.4.5). Collections from Dahina and Gubalafto had variations in PH (75 -112 cm). Dahina had the widest range of variation for GY (311-4639 kg/ha). Significant range of variations for DTM (113-182 days), GFD (20 -82 days), PH (90 - 336 cm), and LL (42 - 94 cm) was observed from landraces collected from Kalu. In this district PL ranged from 8 to 38.2 cm with a mean of 21.17 cm, and LW and LL displayed a wide range of variations i.e. 5.4 to 14.4 cm and 42 to 94.4 cm, respectively. Panicle length is an important character which contributes to yield and represents features of a specific race (Harlan, 1992; Doggett, 1988). DTA, DTM and GFD varied between and within districts with 65, 57, and 71 days, respectively. The maximum value record was 22 times higher than the minimum value for GY, 8 times for HW and 4 times for PH (Table 4.4.5).

4.4.3 Duncan's multiple range test

Means of the top 30 selected entries across districts of collections at two water regimes and two localities are summarized in Tables 4.4.6 and 4.4.7. Landraces with large compact and round panicles were found in Dahina, Gubalfto and Sekota in relatively high proportion whereas landraces with small panicles were common in the Bugina and Kalu collections (Tables 4.4.6 and 4.4.7). The highest mean LN per plant (14) was observed at Ambasel and Kalu landraces whereas the control varieties showed the lowest number of leaves (10) per plant (Table 4.4.6). The mean LW of the landraces collected from Ambasel was significantly higher than the other districts whereas landraces from Dahina, Kobo and Ziquala exhibited lower LW. The mean PH for the landraces from Kobo (229 cm) and Sekota (239 cm) were significantly higher while the control varieties had shorter PH (213 cm) than collections from the other districts (Table 4.4.6).

Table 4.4.5 Minimum and maximum values on 10 quantitative characters among 273 sorghum landraces collected from nine districts and 5 controls.

	Traits ^a												
District	DTA	DTM	GFD	HW	GY	LN	LW	LL	PH	PL			
Ambasel	69.0 - 111.0	111.0 - 182.0	24.0 - 82.0	26.6 - 214.8	226.0 - 4076.0	7.0 - 27.6	4.9 - 14.4	41.6 - 92.4	97.0 - 336.0	8.0 - 36.0			
Bugina	76.0 - 110.0	119.0 -180.0	30.0 - 80.0	41.1 - 132.3	506.0 - 3689.0	9.4 - 17.8	6.0 - 11.4	49.2 - 90.2	139.0 - 311.7	9.0 - 28.3			
Dahina	75.0 - 112.0	120.0 - 180.0	31.0 - 80.0	35.5 - 133.9	311.0 - 4639.0	8.6 - 16.6	6.4 - 10.2	58.8 - 92.4	120.0 - 340.0	9.0 - 37.2			
Gubalafto	75.0 - 111.0	118.0 - 183.0	30.0 -82.0	31.1 - 166.2	231.0 - 4256.0	7.4 - 20.8	5.8 - 12.4	54.0 - 91.0	92.5 - 354.0	8.0 - 38.2			
Habru	77.0 - 117.0	119.0 - 180.0	39.0 - 84.0	62.6 - 182.9	267.0 - 4084.0	9.4 - 16.4	5.8 - 10.3	58.6 - 86.4	136.0 - 291.0	8.0 - 31.2			
Kalu	78.0 - 111.0	113.0 - 182.0	20.0- 82.0	31.1 - 170.3	209.0 - 4124.0	7.0 - 19.8	5.4 - 11.6	42.0 - 94.4	90.0 - 336.2	9.0 - 36.2			
Kobo	81.0 - 111.0	126.0 - 185.0	37.0 - 81.0	40.1 - 128.0	352.0 - 3080.0	9.0 - 17.0	6.6 - 11.2	54.6 - 89.6	148.0 - 302.5	13.0 - 34.6			
Sekota	70.0 - 96.0	120.0 - 165.0	29.0 - 85.0	47.1 - 142.1	399.0 - 4082.0	7.8 - 16.4	5.8 - 12.0	53.8 - 96.2	167.5 - 375.0	9.2 - 30.8			
Ziquala	77.0 - 96.0	120.0 - 160.0	24.0 - 74.0	49.4 - 132.7	709.0 - 3840.0	6.3 - 16.4	6.4 - 10.0	55.2 - 96.0	125.0 - 326.2	10.8 - 34.6			
Controls	60.0 - 86.0	105.0 - 150.0	24.0 - 57.0	80.0 - 152.4	932.0 - 4140.0	5.6 - 15.0	5.4 - 10.6	51.2 - 84.2	118.0 - 330.0	17.2 - 30.8			
Entire	60.0 - 117.0	105.0 -180.0	20.0 - 85.0	26.6 - 214.8	209.0 - 4639.0	5.6 - 27.6	5.4 - 14.4	42.0 - 94.4	90.0 - 375.0	4.0 - 38.2			
data													

^a DTA= days to 50% anthesis, DTM= days to 50% maturity, GFD= grain filling duration, HW= head weight, GY= grain yield, LN= number of leaves, LW= leaf width, LL= leaf length, PH= plant height, PL=panicle length,

The average number of DTA for landraces from Kalu and Kobo was 91 days and Dahina 90 days that were significantly higher than other districts. Landraces from Sekota and Ziquala were significantly earlier to flower at 85 and 86 days, respectively and relatively closer to the control varieties. Landraces from Habru, Kalu, Kobo and Sekota were late maturing as compared to landraces from Ziquala and the controls. Significant differences were not observed between the control varieties and landraces from Bugina, Sekota and Ziquala in GFD. Accessions from Gubalafto and Kobo exhibited significantly stable yield performance followed by accessions from Habru, Bugina, Ambasel and the control varieties (Table 4.4.6).

Mean GFD for the landraces from Kalu was significantly longer than other districts whereas landraces from Ziquala exhibited the shortest. Landraces from Dahina had significantly higher GY (2651 kg/ha) and HW (90 g/plant) followed by landraces from Ziquala and the controls (Table 4.4.7). However, accessions from Gubalafto, Kalu, Habru and Kobo displayed the lowest mean GY. The longest PL (24 cm) was observed from Gubalfto landraces while landraces from Bugina showed short panicles (19 cm) (Table 4.4.7). The controls and landraces from Dahina, Kalu and Ziquala, performed well under Kobo conditions (Table 4.4.6). On the other hand, landraces from Dahina, Ziquala, and the controls performed well under stress condition. This suggests that landraces collected from the drought prone area were well adapted to stress environment.

Under rainfed condition, landraces collected from Dahina flowered significantly earlier (74 days) than the rest of the districts, whereas flowering at Kobo and Sekota was late at 94 and 97 days, respectively (Table 4.4.7). This suggested that the stress environment induced landraces from Dahina to flower and mature earlier. Environmental factors, such as photoperiod, temperature, and water availability, show regular seasonal changes that control the transition to flowering (Bernier et al., 1993). Moreover, under irrigation condition, landraces from Dahina produced significantly longer leaf (76 cm), whereas under rainfed condition they had significantly shorter leaf length (64 cm) (Tables 4.4.6 and 4.4.7). Generally, more regional mean differentiation was observed for GFD, GY, PH and YS in the rainfed experiment, whereas more regional mean differentiation was observed for HW, LW, LL and PL in the irrigation trials (Tables 4.4.6 and 4.4.7). Ceccarelli (1994) indicated that the expression of morphological and physiological plant characteristics associated with yield in optimal and stress conditions are different.

Reduced leaf area index, small and narrow leaf structure, reduced plant stature, and low tillering ability are reported as drought adaptive mechanisms in plants (Richards *et al.*, 2002). Larger plants have larger leaf area and transpire more water than smaller plants and transpiration is first restricted by reduction of leaf expansion. Short cultivars are efficient in

balancing assimilate translocation between the developing grain and other vegetative organs as compared to tall genotypes (Kouressy *et al.*, 2008). Hence genetic dwarfing of tall genotypes improves the grain yield potential of sorghum in arid and semi-arid environments through increased harvest index.

Short growing duration is considered as an important trait of drought escape. The advantage of early maturing genotypes as in landraces from Dahina, Ziquala, Sekota, and Bugina in drought affected areas has long been realized by farmers and breeders. On the other hand, most studies showed that high yield potential and late maturity are positively correlated under favorable conditions (van Oosterom *et al.*, 2006). Hence, drought escape by shortening the growing period is made at the expense of the crop's potential yield (Blum, 1988). The traditional tall varieties denominated as 72995, 239147, 72439-B, 71160-B, 70023, 206114-A, and 73079, in this study are characterized by extended growth duration, with moderately high biomass yield, and low grain yield under drought. The short landraces (244745, 244732-A, 72439-C, 69251, and 72493), on the other hand, have reduced biomass yield and relatively high grain yield.

Table 4.4.6 Means of sorghum landraces collected from nine districts with controls on 11 quantitative traits (averaged from two water regimes)

	Traits ^{a,b}											
District	DTA	DTM	GFD	HW	GY	LN	LW	LL	PH	PL	YS	
Ambasel	89.60 ^{bc}	141.10 ^{ab}	51.53 ^{ab}	91.70 ^{bcd}	1768 ^{bcd}	13.55 ^a	8.59 ^a	75.70 ^a	234.60 ^{ab}	20.91 ^{cde}	1.28 ^{cd}	
Bugina	88.95 ^c	137.40 ^{abc}	48.42 ^{abc}	81.95 ^{def}	1667 ^{bcd}	13.41 ^a	8.33 ^{abc}	74.09 ^{bc}	228.60 ^b	18.49 ^e	1.25 ^{cd}	
Dahina	89.93 ^{abc}	117.70 ^d	51.70 ^{ab}	107.82 ^a	3181 ^a	13.02 ^a	8.13 ^c	75.55 ^a	188.30 ^d	22.10 ^{bc}	1.42 ^{bc}	
Sublafto	89.61 ^{bc}	141.60 ^{ab}	54.19 ^a	73.59 ^f	1430 ^d	13.22 ^a	8.30 ^{abc}	74.17 ^b	211.70 ^c	25.02 ^a	0.80 ^e	
labru	89.01 ^c	143.80 ^a	55.85 ^a	84.81 ^{cde}	1598 ^{bcd}	13.20 ^a	8.33 ^{abc}	74.94 ^{ab}	231.00 ^{ab}	21.37 ^{cd}	1.11 ^d	
alu	91.02 ^a	144.90 ^a	53.54 ^{ab}	101.04 ^{ab}	2099 ^b	13.52 ^a	8.39 ^{abc}	74.30 ^b	230.60 ^b	19.22 ^{de}	0.64 ^e	
obo	90.60 ^{ab}	144.60 ^a	53.76 ^a	80.51 ^{ef}	1522 ^{cd}	13.31 ^a	8.09 ^c	72.76 ^{cd}	228.90 ^b	20.35 ^{cde}	0.84 ^{de}	
Sekota	84.69 ^d	144.40 ^a	45.70 ^{bc}	82.53 ^{def}	1490 ^{cd}	12.28 ^a	8.25 ^{bc}	71.88 ^{de}	238.80 ^a	24.00 ^{ab}	1.71 ^a	
Ziquala	85.78 ^d	130.40 ^c	48.12 ^{abc}	94.57 ^{bc}	2023 ^{bc}	12.26 ^a	8.05 ^c	72.57 ^d	237.80 ^a	20.70 ^{cde}	1.59 ^{ab}	
Controls	75.45 ^e	133.90 ^{bc}	42.80 ^c	91.23 ^{bcd}	1997 ^{bc}	9.70 ^b	8.49 ^{ab}	70.87 ^e	212.90 ^c	20.47 ^{cde}	1.43 ^{bc}	

^a DTA= days to 50% anthesis, DTM= days to 50% maturity, GFD= grain filling duration, HW= head weight, GY= grain yield,

LN= number of leaves, LW= leaf width, LL= leaf length, PL=panicle length, PH= plant height, YS= yield stability

^b Means followed by the same letter in a column are not significantly different after the Duncan's Multiple Range Test procedure at P=0.01

Table 4.4.7 Means of sorghum landraces collected from nine districts with controls on 11 quantitative traits (averaged from two locations)

Traits ^{a,b}											
District	DTA	DTM	GFD	HW	GY	LN	LW	LL	PH	PL	YS
Ambasel	92.57 ^{ab}	144.80 ^{ab}	52.26 ^{def}	87.47 ^{ab}	1653 ^{cd}	9.17 ^b	8.06 ^b	72.91 ^a	217.70 ^{ab}	20.41 ^{bcd}	1.44 ^d
Bugina	93.50 ^{ab}	145.50 ^{ab}	52.05 ^{def}	81.27 ^{cd}	1564 ^{cde}	11.76 ^a	8.57 ^a	70.79 ^c	216.30 ^b	18.50 ^d	1.93 ^{bc}
Dahina	73.69 ^d	124.9 ^c	51.21 ^{ef}	90.33 ^a	2651 ^a	11.89 ^a	8.37 ^{ab}	63.95 ^d	179.90 ^e	21.99 ^{ab}	2.305 ^a
Gublafto	93.03 ^{ab}	145.80 ^{ab}	52.81 ^{de}	79.51 ^{cd}	1354 ^{ef}	11.90 ^a	8.128 ^b	72.00 ^{abc}	192.40 ^d	24.42 ^a	0.65 ^f
Habru	93.22 ^{ab}	148.70 ^a	55.48 ^{bc}	78.60 ^d	1432 ^{det}	12.61 ^a	8.53 ^a	71.79 ^{abc}	211.50 ^{bc}	21.34 ^{bc}	1.46 ^d
Kalu	93.88 ^{ab}	152.60 ^a	58.75 ^a	87.32 ^{ab}	1682 ^{bc}	12.82 ^a	8.40 ^{ab}	71.74 ^{abc}	215.60 ^b	19.07 ^{cd}	0.77 ^{ef}
Kobo	94.11 ^a	149.10 ^a	54.95 ^c	80.79 ^{cd}	1427 ^{ef}	12.89 ^a	8.30 ^{ab}	72.54 ^{ab}	212.40 ^b	20.31 ^{bcd}	0.96 ^e
Sekota	96.55 ^a	152.40 ^a	55.83 ^b	71.82 ^e	1195 ^f	12.94 ^a	8.61 ^a	72.50 ^{ab}	215.60 ^b	22.86 ^{ab}	2.05 ^b
Ziquala	87.79 ^{bc}	138.50 ^b	50.74 ^f	83.82 ^{bc}	1830 ^b	12.95 ^a	8.30 ^{ab}	71.33 ^{bc}	223.70 ^a	20.87 ^{bcd}	1.97 ^{bc}
Controls	84.90 ^{bc}	138.60 ^b	53.67 ^{cd}	82.32 ^{bcd}	1853 ^b	13.03 ^a	8.44 ^{ab}	71.48 ^{abc}	204.90 ^c	21.00 ^{bcd}	1.69 ^{cd}

^a DTA= days to 50% anthesis, DTM= days to 50% maturity, GFD= grain filling duration, HW= head weight, GY= grain yield,

LN= number of leaves, LW= leaf width, LL= leaf length, PH= plant height, PL=panicle length, YS= yield stability

^b Means followed by the same letter in a column are not significantly different after the Duncan's Multiple Range Test procedure at P=0.01

The present study showed the existence of landraces with short duration and reduced plant height from collections of Wello including Ziquala, Sekota, Dahina and Kobo. In Ethiopia, more than 40 improved sorghum varieties have been released since 1973, out of which, 16 varieties were released for the lowlands and the other for the highland agro-ecologies (McGuire, 2008). Developing improved varieties for specific stress conditions requires exploring locally adapted germplasm and performing the selection under specific stressful conditions (Ceccarelli *et al.*, 2001). Availability of genetic diversity will enable plant breeders to develop cultivars that combine farmers' desirable attributes.

4.4.4 Correlations of yield and its components

Grain yield was positively and significantly correlated with head weight but negatively correlated with days to 50% flowering, days to 50% maturity and grain filling duration (Table 4.4.8). Similarly, grain yield was found to be positively and significantly correlated with plant height. Days to 50% flowering was positively and significantly correlated with days to 50% maturity (r = 0.67, p < 0.001) number of leaves (r = 0.62, p < 0.001), leaf length (r = 0.33, p = 0.01), and leaf width (r = 0.18, p = 0.05) but negatively correlated with grain yield, head weight and panicle length. These results are in conformity with those reported by Ayana and Bekele (2000). Previous studies have shown that reduced leaf area index, small and narrow leaf structure, reduced plant stature, and low tillering ability are drought adaptive mechanisms in plants (Richards, 2000; Richards et al., 2002). Mortlock and Hammer (1999) observed that larger plants have larger leaf area and transpire more water than smaller plants. In water limited condition, transpiration is first restricted by reduction of leaf expansion (Sinclair et al., 2005). In addition, genetic dwarfing of tall genotypes improves the grain yield potential of sorghum in arid and semi-arid environments. Dwarf cultivars are efficient in balancing assimilate translocation between the developing grain and other vegetative organs as compared to tall genotypes (Kouressy et al., 2008).

Table 4.4.8 Pair-wise correlation coefficients indicating the association of grain yield and its components in sorghum.

	DTA	DTM	GFD	HW	LL	LN	LW	PH	PL	GY
DTA	1.00									
DTM	0.67**	1.00								
GFD	-0.09	0.67**	1.00							
HW	-0.27*	-0.40**	-0.27*	1.00						
LL	0.33**	0.36**	0.15*	0.08	1.00					
LN	0.62**	0.56**	0.13*	-0.08	0.50**	1.00				
LW	0.18*	0.20*	0.09	0.07	0.31**	0.22*	1.00			
PH	0.07	0.05	0.00	0.22*	0.40**	0.42**	0.10	1.00		
PL	-0.26*	-0.15*	0.05	-0.01	-0.06	-0.21*	-0.12	0.05	1.00	
GY	-0.27*	-0.40**	-0.27*	1.00**	0.08	-0.08	0.07	0.22*	-0.01	1.00

^{*} and ** denote significant differences at 0.05 and 0.01 probability levels, respectively.

DTA= days to 50% anthesis, DTM= days to 50% maturity, GFD= grain filling duration, HW= head weight, LN= number of leaves, LW= leaf width, LL= leaf length, PH= plant height, PL=panicle length, GY= grain yield

Conclusion

In conclusion, the study identified considerable magnitude of genetic variation among landraces between and within districts of collection for most quantitative traits. Landraces designated as 214838-A, 72476, 73090, 244733, 239150, 200113, 214855, 239255-B, and 69251 were identified as suitable genotypes with high grain yield and head weight, increased panicle length, medium maturity and long stalk. On the other hand, landraces 244745, 244732-A, 72439-C, 69251, and 72493 were selected for short stalk, reduced biomass yield and relatively high grain yield. Similarly landraces 239167-B, 244725-A, 242049-B, 242050-A, 244711-A, 202508, 244729-A, 242046, 75454, 73056-B and 72457 were recognized as early maturing with relatively medium grain yield and intermediate stalk. These germplasm will serve as sources of drought tolerance genes in sorghum improvement programmes for the north and north eastern Ethiopia and other similar environments. Further breeding will be initiated using the selected genetic resources to enhance high and stable yield, drought tolerance and early maturity.

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CHAPTER 5

Molecular Genetic Variation in Lowland Sorghum (*Sorghum bicolor* (L.) Moench) Landraces of Ethiopia Assessed by Simple Sequence Repeats (SSRs)

5.1 Abstract

The North Eastern regions of Ethiopia are known for thier high level of sorghum genetic diversity, and for being drought prone. Sorghum has been developed and maintained by farmers under these marginal and heterogeneous environments for centuries. The objective of this study was to examine the genetic diversity and population structure of 200 landraces collected from seven districts using 30 microsatellite markers sampled from all linkage groups of the sorghum genome. Both distance-based and model-based analytical approaches were used to analyze the data. Extensive variation was observed for all the markers used in this study. A total of 316 putative alleles were recorded with an average of 10.5 alleles per locus. The PIC values ranged from 0.26 to 0.88 with a mean value of 0.61, reflecting the high discriminating ability of the markers used. The total gene diversity was 0.69, which partitioned 86% among and 14% within landraces. The first was largely due to variation among genotypes within geographic origins. No genetic variation was observed between geographic origins. Landraces from different collection sites appeared to cluster together indicating the existence of high level of gene flow among regions. The low level of genetic differentiation and high level of gene flow between the different collection sites indicates that a relatively small random collection of landraces would capture the bulk of genetic diversity in the target area. However, the modelbased analysis clustered the landraces into two distinct sub-groups with relatively high genetic differentiation. This would enable breeders to select distinctive alleles and exploit the potential of transgressive segregation between the two sub-groups. Given that the target area is marginal, drought prone and heterogeneous, the sorghum landraces' diversity studies could serve as an important source of valuable alleles for drought tolerance in future sorghum improvement programmes.

Keywords: Genetic diversity, drought tolerance, lowland sorghum, microsatellite markers, Sorghum bicolor

5.2 Introduction

Genetic analysis involves investigation of variations among individuals, groups or populations, which can be conducted by a specific method or combinations of methods. Genetic diversity is a necessary condition to achieve yield stability. It is needed for plant breeders to develop improved crop varieties. Genetic diversity studies can be performed at a phenotypic, biochemical or molecular level. Biochemical markers such as isozymes and storage proteins have been used extensively to characterize plant genetic resources (Ayana *et al.*, 2001; Gregova *et al.*, 2004; Zong *et al.*, 2005). The applications of protein markers have been limited by the relatively low levels of polymorphisms detected.

DNA based molecular markers, on the other hand, are more reliable and robust methods for the characterization of genetic diversity. Currently, there exists a wide range of molecular markers that can be used to characterize genetic diversity. A range of molecular markers have been used on sorghum such as restriction fragment Length polymorphism (RFLP) (Smith *et al.* 1997; Menz *et al.*, 2004; Perumal *et al.*, 2007), random amplified polymorphic *DNA* (*RAPD*) (Ayana *et al.*, 2000; Dahlberg *et al.*, 2002; Agrama and Tuinstra, 2003), Amplified Fragment Length Polymorphism (AFLP) (Menz *et al.*, 2004; Geleta *et al.*, 2006; Perumal *et al.*, 2007) and simple sequence repeats (SSR) (Smith *et al.*, 2000; Dje *et al.*, 2000; Uptmoor *et al.*, 2005; Dillon *et al.*, 2005; Manzelli *et al.*, 2007; Ganapathy *et al.*, 2012). DNA marker applications have facilitated the identification of agronomic traits in wild, traditional and improved germplasm through the dissection of quantitative traits using linkage map based approaches (Tanksely and McCouch, 1997).

Simple sequence repeats (SSR) markers are among the most widely used DNA based molecular markers. SSR markers consist of tandemly repeated, short DNA sequence motifs that tend to occur in non-coding regions of the genome. They are ubiquitous components of all eukaryotic genomes and are also found in prokaryotes (Tóth, *et al.*, 2000). In sorghum, SSR markers have been widely applied for assessment of genetic diversity and characterization of germplasm (Smith *et al.*, 2000; Anas and Yoshida, 2004; Assar *et al.*, 2005; Geleta *et al.*, 2006; Perumal *et al.*, 2007; Deu *et al.*, 2008), identification and fingerprinting of genotypes (Ellis and Burke, 2007; Menz *et al.*, 2002), estimation of genetic distances between and within populations (Smith *et al.*, 2000; Anas and Yoshida, 2004; Geleta *et al.*, 2006), detection and mapping of QTLs (Haussmann *et al.*, 2002; Nagaraj *et al.*, 2005; Brown *et al.*, 2008; Knoll and Ejeta, 2008), and gene tagging and mapping (Bhattramakki *et al.*, 2000; Klein *et al.*, 2001; Mukta and Boora,

2005). It has been frequently reported that SSRs are highly polymorphic and provide a wider genome coverage than other molecular markers (Anas and Yoshida, 2004). DNA fingerprinting is an increasingly popular method for crop diversity studies through estimation of genetic distances. This method is relatively simple and cost effective because of the availability of large numbers of public SSR markers.

Developing species specific SSR markers requires significant resources since SSR markers entail prior DNA sequence data. However, once the SSRs are developed, they can be used to provide DNA profiles that are highly discriminative among genotypes of many species (Senior and Heun, 1993; Roder et al., 1995; Rongwen et al., 1995; Charters et al., 1996; Smith et al., 1997; Smith et al., 2000). Although no information is available on SSR motifs and primers, the sorghum genome sequence project identified 71, 000 SSRs in the genome (Paterson et al., 2009). The development of high throughput PCR-based marker technology and the availability of this huge number of SSR markers provide a more cost effective and rapid method for DNA profiling (Smith et al., 2000). Microsatellite markers combine the property of high polymorphism, higher relative abundance, high reproducibility, co-dominance, being multi-allelic and wide genome coverage (Jones et al., 1997). A single SSR marker can be mapped as a discrete locus and the location of this is stably inherited within a species unlike RFLP and RAPDs markers (Senior et al., 1996). Additionally, a relatively small set of SSR markers can be used as a tool for measuring genetic diversity and provide information valuable for genetic conservation (Smith et al., 2000).

In Ethiopia, the most important staple cereal crops include tef [*Eragrostis tef* (Zucc.) Trotter.], maize (*Zea mays* L.), sorghum [*Sorghum bicolour* (L.) Moench] and wheat (*Triticum aestivum* L.) accounting for 28.5, 20.3, 19.6, and 16% of the total cereal crop cultivated area, respectively (CSA, 2011). Sorghum is grown in almost all regions by subsistence farmers for multiple uses such as food, animal feed and as the basis of some local beverages. However, its productivity has remained relatively low with an estimated national mean yield of < 2 t ha⁻¹ (CSA, 2012) due to several biotic, abiotic and socio-economic constraints. Sorghum productivity in regions of Ethiopia could be enhanced through effective breeding using locally adapted and well-characterized germplasm.

An understanding of genetic diversity among inbred lines can be particularly useful in planning crosses, in assigning lines to specific heterotic groups, and for precise identification of plant material for strategic conservation (Geleta *et al.*, 2006; Perumal *et al.*, 2007; Deu *et al.*, 2008).

Analysis of genetic diversity in germplasm collection can facilitate reliable classification of genotypes and identification of groups of core genotypes with utility for specific breeding purposes (Grenier et al., 2001). Although molecular markers have proven to be a valuable tool in the study of plant genetic diversity, they have not been applied widely in Ethiopia for genetic diversity analysis or sorghum germplasm development. Genetic analysis of Ethiopian lowland sorghum germplasm using SSR markers is scanty. Typically, sorghum breeding programmes in the country establish the genetic relationships through phenotypic evaluations, often with no application of molecular methods. To date, few diversity assessments have been made by breeders in the country. Menkir et al. (1997) applied RAPD markers and studied the genetic diversity of sorghum sampled from different parts of Ethiopia. Ayana et al. (2000) used the same RAPD markers and determined sorghum diversity collected from western Ethiopia. Geleta et al. (2006) used SSR and AFLP markers for genetic diversity analysis of sorghum collections from eastern Ethiopia. However, the previous reports did not cover landraces adapted to the lowland areas where drought stress and stalk borer insect damage largely limits sorghum productivity. Thus, further studies that include sorghum landraces evolved and maintained in the droughtprone environments is vital for effective breeding. This study, therefore, aimed to systematically assess the genetic diversity and interrelationships among and within genotypes collected from drought-prone lowland areas of North Eastern Ethiopia, using polymorphic SSR markers sampled from the 10 linkage groups.

5.3 Materials and methods

5.3.1 Plant material

Out of 273 sorghum genotypes collected across a wide altitudinal range (900-1540 masl), 200 were selected on the basis of their morphological distinctiveness and drought tolerance, in terms of earliness and yield stability, when evaluated across different environments. The selected sorghum germplasm included both sweet stem and grain sorghum genotypes. The 200 genotypes were sampled from three administrative zones representing seven districts of which 46 landraces were from Ambasel, 54 from Kalu, 10 from Kobo, 12 from Bugina, 44 from Gubalafto, 14 from Zikuala, and 16 from Sekota. Four known genotypes, two sweet stem (AS-27 and AS-103) and two grain types (*Jigurte* and *Raya*) sorghum genotypes were included as comparative controls in the study. For each genotype, a bulk sample of leaf tips from five seedlings was harvested 4 weeks after germination. The leaf samples were dried using a silica gel protocol (Chase and Hills, 1991; Rogstad, 2003). The samples were arranged in 3 x 96-well

plates and sent to DNA LandMarks Inc., Canada for SSR analysis. Genomic DNA was extracted from the dried leaf samples (Sharma *at al.*, 2002) and the quality of the extracted DNA was evaluated on 1% agarose gel.

5.3.2 Microsatellite marker evaluation

For molecular level diversity assessement, 30 microstatelite markers were used. The markers used in this study were selected from SSR diversity kit (http://sat.cirad.fr/sat/sorghum_SSR_kit) from all the linkage groups of sorghum (Table 1). Genotyping was conducted at the genotyping service labratory of DNA LandMarks Inc., Canada. For electrophoresis, M13 forward primer method (5'CACGACGTTGTAAAACGAC3') was used at the 5' end of each primer. M13-forward primer was labelled with either one of the four fluorescence dyes (6FAM, PET, NED or VIC) (Applied Biosystems) for PCR products detection using the ABI 3730xl Genetic Analyzer (Applied Biosystems, USA). Multiplex PCR reactions were performed in a total of 20µl reaction volume using GeneAmp PCR system 9700 thermal cycler (Applied Biosystems, USA).

The SSR reaction contained 2µl (25ng) genomic DNA, 0.75µl M13-tailed forward primer (2pM/µl), 0.75µl reveres primer (2pM/µl), 0.8µl MqCl₂ (25mM), 2µl 10x reaction buffer, 0.4 dNTPs (100mM), 0.1μl Taq DNA polymerase (10U/μl) and 13.2 ddH₂O. The PCR reaction conditions consisted of 5 min at 94°C for initial denaturation, followed by 35 cycles of polymerization reaction, each consisting of denaturation step of 1 min at 94°C, an annealing at 61°C for 1 min (temperature reduced by 1°C for each cycle), and a polymerization step of 1 min at 72°C. This step was repeated for 35 cycles of denaturation of 1 min at 94°C, an annealing step of 1 min at 54°C, and a polymerization step of 1 min at 72°C, followed by a final polymerization step of 10 min at 72°C based on their expected amplicon size and dye. PCR products were pooled together along with internal size standards (GeneScan[™], 500 LIZ® from Applied Biosystems, USA). The data was generated through electrophoresis and then analyzed using GeneMapper software (Applied Biosystems, USA) and product size was scored in base pairs based on the relative migration of the internal size standard. Information generated from GeneMapper software was then used to determine diversity parameters including major allele frequency (A_f), number of alleles (N_a), number of effective alleles (N_e), observed heterozygosity (H₀), total gene diversity (H_t), average gene diversity with genotypes (H_e), gene flow per locus and geographic origin (N_m) , polymorphic information content (PIC), fixation index (F) and outcrossing rate (t).

5.3.3 Genetic diversity analysis

The Chi-square test was performed to determine the differences in allele frequencies among SSR markers. Genetic diversity parameters were determined, such as the number of different alleles ($N_{\rm e}$), the number of effective alleles ($N_{\rm e}$), allele richness ($N_{\rm e}$), observed heterozygosity ($N_{\rm e}$), total gene diversity ($N_{\rm e}$), average gene diversity ($N_{\rm e}$) using the protocol of Nei (1978). Furthermore, fixation index ($N_{\rm e}$), out-crossing rate (t) calculated as (1 - F)/(1 + F), and percent polymorphic loci ($N_{\rm e}$) were estimated with FSTAT version 2.9.3 (Goudet 2001) and GenAlex (Peakall and Smouse, 2006) for each pre-determined group, based on districts of collection of the genotypes.

Global and pairwise F_{ST} were computed using Weir and Cockerham's Q methods using FSTAT (Weir and Cockerham, 1984). Other parameters such as gene flow, Nei's unbiased genetic distance, and identity were estimated to examine the degree of population differentiation using GenAlex. Partitioning of the total genetic variation into Within and Among districts of collection was performed with a molecular analysis of variance (AMOVA) procedure using GenAlex (Peakall and Smouse, 2007).

5.3.4 Cluster analysis

The pattern of genetic diversity within and among genotypes and across the districts of collection was assessed using relevant software programmes. Genetic relationships within and among genotypes from the seven districts were assayed with neighbor-joining method in DARwin 5.0 software (Perrier *et al.*, 2003; Perrier and Jacquemoud-Collet, 2006). Bootstrap analysis was performed for node construction using 1000 bootstrap values.

The Bayesian genotypic clustering approach of STRUCTURE (Pritchard *et al.*, 2000) was used to validate population structures among the genotypes. Admixture model with independent allele frequency was used to simulate the population. For joint inference of population sub-structure, the model was run with 200000 MCMC iteration steps and 100000 burn-ins, assuming different starting points. Graphical representation of population assignments from STRUCTURE were produced from the programme DISTRUCT (Rosenberg, 2002).

5.4 Results and discussion

5.4.1. Genetic diversity within and among accessions

The 200 sorghum genotypes evaluated in this study were differentiated uniquely, using the 30 SSR markers. The 30 SSR loci were able to generate clear bands within the expected size range (Table 5.4.1). A total of 316 putative alleles (different fragment sizes) were detected among the 200 sorghum genotypes. The higher level of allelic diversity of SSR loci examined in this study was probably associated with the wide range of genetic diversity represented in the germplasm of Ethiopian lowland sorghum tested. The statistics of genetic diversity parameters within and among genotypes are given in Table 5.4.1. The total number of polymorphic alleles amplified per locus over all genotypes ranged from 2 (Xcup61) to 24 (mSbCIR240), while the mean number of alleles per locus for the whole set of genotypes was 10.5. The values in this study are higher than earlier results of Smith et al. (2000), Anas and Yoshida (2004), Menz et al. (2004), Assar et al. (2005) and Ganapathy et al. (2012). On the other hand, the effective numbers of alleles (N_e) for 17% of the loci were less than 2, and 27% of the loci had N_e values of more than 5, with a mean of 3.7 alleles per locus. The differences observed between total (N_a) and effective number of alleles (N_e) was due to the variation in the frequency of major alleles among genotypes. The distribution of allele frequency at a single locus varies among genotypes.

All the microsatellite markers used in this study were highly polymorphic. The PIC values of loci provide an estimate of the discriminatory power of loci considering the number of alleles and their relative frequencies (Smith *et al.*, 2000). The PIC values for SSR loci ranged from 0.26 for mSbCIR223 to 0.88 for Xgap206 and Xtxp145, with the mean value of 0.61. The present findings suggested that the level of diversity shown by the 30 microsatellites over all genotypes was slightly higher than the level described by Brown *et al.* (1996) with a mean diversity index of 0.56. On the other hand, the level of polymorphism in this study is lower than Taramino *et al.* (1997) and Kong *et al.* (2000), who reported a mean diversity index of 0.80 and 0.89, respectively. In general, the current results concur with the results of Smith *et al.* (2000), Agrama and Tuinstra (2003) and Geleta *et al.* (2006). It was also indicated that markers with moderate PIC values can uniquely classify most of the inbred lines and detect the polymorphism rate at a specific locus (DeWoody *et al.*, 1995; Smith *et al.*, 2000). Forty percent of the loci used in this study had PIC values of more than 0.75, indicating that these loci have high discriminatory powers that distinctively classify most of the genotypes.

The SSR loci scored in this study produced a wide range of amplified fragment sizes (the difference between the longest and shortest fragments) from 4 to 134 bp, which is much wider than initially reported with ranges of 4 to 60 (Table 5.4.1). Similarly, the total number of alleles is much higher than those reported in earlier publications (Brown *et al.* 1996; Taramino *et al.* 1997; Bhattramakki *et al.* 2000; Kong *et al.* 2000; Schloss *at al.* 2002; Geleta *et al.* 2006). This is probably observed due to the large sample size and the differences in the geographic origin of genotypes used in this study. The highest allele size variation was observed in Locus Xgap265 and could have been caused by slip strand mispairing during DNA replication (Levinson and Gutman, 1987), or caused by lower primer binding stringency due to potential mutation on a primer binding site (Dillon *et al.*, 2005).

Chi-square test results showed significant differences in allele frequencies at all loci for all the genotype sets. Total genetic diversity (H₁) ranged from 0.28 (mSbCIR223) to 0.89 (Xtxp145 and Xgap206). Similarly the mean gene diversity (H_e) was observed to be 0.56, with maximum and minimum values recorded by microsatellite markers mSbCIR223 (0.21) and Xgap206 (0.81). The mean total gene diversity was 0.69. The observed heterozygosity at each locus ranged from 0.02 (mSbCIR223 and Xtxp040) to 0.79 (Xgap265) with a mean value for all the loci of 0.11. Similarly, except for Xtxp265, the fixation indices (F) for all the markers were more than 0.75. The observed low heterozygosity and high fixation index for all the germplasm included in this study indicated that the landraces were distinct and predominantly homozygous, which is maintained by continued self-fertilization. An indirect estimate of the level of gene flow between genotypes was calculated using Nm = $0.25 (1-F_{st})/F_{st}$ following Nei (1987). The level of gene flow was high for Loci Xtxp010, Xgap72 and Xcup14 and low for loci Xtxp021 and Xtxp321, with the mean gene flow of 3.7. The observed low level of genetic differentiation $F_{ST} = 0.06$, may be explained by the high level of gene flow. This may be attributed to the fact that farmers in Ethiopia maintained large numbers of varieties on a single plot, resulting in a continuous exchange of genes through pollen flow (Manzelli et al., 2007; Barnaud et al., 2008).

In this study, when reviewing SSR classes based on the number of repeats and motifs, the compound microsatellite markers (gpsb123, mSbCIR283, Xgap206, and Xtxp321) showed higher allele numbers (average 16 per locus) and PIC values (average 0.78 per marker) followed by dinucleotide (mean alleles 11 per locus; PIC value mean 0.62 per marker) and trinucleotide SSRs (mean alleles 9 per locus; PIC value mean 0.52 per marker). However, Smith *et al.* (1997) and Mbeyagala *et al.* (2012) reported that di- and tri-nucleotide repeats

presented high PIC values compared to compound repeats. Among dinucleotide SSRs, TG/TC repeat motifs exhibited more informativeness (mean alleles of 12 per locus and PIC value with a mean of 0.64 per marker) as compared to AG/AC repeat motifs (mean alleles of 10 per locus and a PIC mean 0.61 per marker (Table 5.4.1). These results correspond with those of Dillon *et al.* (2005). Although, Taramino *et al.* (1997) and Kong *et al.* (2000) reported that AG repeats were more prolific and more widely scattered throughout the sorghum genome than other repeat types, in the current result AG repeats were less informative than TG/TC repeats. Smith *et al.* (1997) pointed out that generally di-nucleotide repeats are associated with shutter bands that complicate the accuracy of fingerprinting. Although this needs further investigation, it was found that there was a relationship between repeat length and type, and degree of polymorphism, using the 30 SSR markers available for *S. bicolor*. This also may have implication on selection of molecular markers for genetic diversity analysis and their application in marker-assisted selection.

The mean number of observed and effective alleles per locus for each collection site was highly correlated with the sample size, showing more than 6 alleles per locus per collection district (Table 5.4.2). At the zonal level, the highest number of alleles per locus per zone was observed from South Welo (9.1) and the smallest from Wag-Hemra (6.7). The mean number of effective alleles per locus was slightly higher in South Welo collections (3.7) followed by North Welo (3.6) and Wag-hemra (3.5). However, the number of effective alleles in the control genotypes at 2.4 was significantly lower than the landraces collected from the three zones. No significant variation was observed in allelic richness at the zonal level. However, allelic richness at district level showed that the mean number of alleles per locus per collection district ranged from 2.4 in the control genotypes to 3.7 in genotypes collected from Gubalafto and Kalu. The control genotypes were commercial varieties selected for improved yield, drought tolerance and high stalk sugar content. The continued exclusive breeding of these genotypes would result in narrow genetic diversity and low allelic richness when compared to the tested landraces.

Wright (1965) clarified three F-statistics to quantify the extent of genetic differentiation among and within population. F_{IT} measures correlations of alleles within individuals in the overall population, F_{ST} measures correlation of alleles of different individuals in the same population, and F_{IS} quantifies correlation of alleles within individuals within populations. In other words, F-statistics quantify the probability that two homologous alleles drawn at random are identical by descent (Falconer, 1981). Analysis of molecular variation (AMOVA) results of the total genetic variation among the sorghum landraces, among zones and districts showed that genetic

differentiation at zonal and district level appeared to be very low. However, out of the total genetic variation, 84% was attributed to variation among genotypes while only 16% was due to variation within genotypes (data not shown). Similar estimates of genetic variation among genotypes have been reported by Menkir *et al.* (1997) and Nguni *et al.* (2011) using RAPD and SSR markers, respectively.

Table 5.4.1 Genetic diversity within and among 200 sorghum genotypes based on 30 microsatellite markers

Marker	Motif				Ge	netic p	arame	eter			
		LG	N _a	N _e	H _t	H _o	H _e	N _m	F	Т	PIC
mSbCIR223	(AC) ₆	B(2)	3	1.40	0.23	0.02	0.21	3.43	0.92	0.08	0.26
mSbCIR276	(AC) ₉	C(3)	3	2.22	0.63	0.10	0.53	6.57	0.82	0.15	0.49
mSbCIR286	(AC) ₉	A(1)	11	2.15	0.50	0.03	0.47	2.35	0.95	0.04	0.52
SbAGB02	(AG) ₃₅	E(7)	12	5.19	0.85	0.10	0.75	2.70	0.89	0.10	0.78
Xcup14	(AG) ₁₀	C(3)	3	1.57	0.37	0.03	0.35	7.40	0.95	0.05	0.33
Xgap72	(AG) ₁₆	I(6)	11	3.41	0.74	0.08	0.67	7.02	0.85	0.13	0.66
Xgap84	(AG) ₁₄	B(2)	16	6.71	0.95	0.15	0.80	5.43	0.77	0.19	0.83
Xtxp021	(AG) ₁₈	D(4)	12	2.26	0.55	0.06	0.49	1.87	0.85	0.13	0.54
Xtxp141	(GA) ₂₃	G(10)	15	5.76	0.86	0.09	0.77	4.34	0.86	0.12	0.81
Xtxp145	(AG) ₂₂	I(6)	17	9.23	0.94	0.10	0.84	4.33	0.90	0.09	0.88
Di-nucleotides	(AC/AG)n repeats		10.3	3.99	0.66	0.08	0.59	4.54	0.88	0.11	0.61
gpsb067	(GT)10	H(8)	8	2.74	0.63	0.09	0.54	3.37	0.86	0.12	0.60
mSbCIR240	(TG) ₉	H(8)	24	4.95	0.88	0.10	0.78	4.85	0.82	0.15	0.79
mSbCIR306	(GT) ₇	A(1)	3	2.35	0.59	0.07	0.52	3.51	0.88	0.11	0.48
Xtxp057	(GT) ₂₁	I(6)	13	4.76	0.84	0.11	0.74	2.69	0.88	0.11	0.76
Xtxp010	(CT) ₁₄	F(9)	9	1.74	0.46	0.07	0.39	7.75	0.85	0.13	0.41
Xtxp012	(CT) ₂₂	D(4)	14	5.18	0.87	0.11	0.76	5.11	0.82	0.15	0.78
Xtxp015	(TC) ₁₆	J(5)	10	2.77	0.79	0.16	0.63	3.77	0.70	0.23	0.61
Di-nucleotides	(TG/TG)n repeats		11.6	3.5	0.72	0.10	0.62	4.44	0.83	0.14	0.63
Xcup02	(GCA) ₆	F(9)	4	2.20	0.58	0.07	0.52	6.25	0.90	0.09	0.44
Xcup53	(TTTA) ₅	A(1)	4	2.01	0.50	0.07	0.43	2.89	0.84	0.14	0.44
Xcup61	(CAG) ₇	C(3)	2	1.97	0.50	0.05	0.45	2.47	0.90	0.09	0.37
Xtxp040	(GGA) ₇	E(7)	4	1.53	0.34	0.02	0.32	2.19	0.95	0.05	0.30
Xtxp114	(AGG) ₈	C(3)	4	1.51	0.34	0.04	0.30	3.96	0.94	0.06	0.29
Xtxp265	(GAA) ₁₉	I(6)	21	5.77	1.51	0.73	0.78	6.08	0.11	0.47	0.81
Xtxp273	(TTG) ₂₀	H(8)	16	6.91	0.92	0.13	0.79	3.54	0.85	0.13	0.84
Xtxp278	(TTG) ₁₂	E(7)	9	1.91	0.53	0.10	0.43	3.04	0.81	0.16	0.44
Xtxp320	(AAG) ₂₀	A(1)	14	3.56	0.76	0.08	0.68	4.23	0.86	0.12	0.70
Tri-nucleotides		B(2)	8.7	3.0	0.66	0.14	0.52	3.85	0.80	0.15	0.51
Xgap206	(AC) ₁₃ (AG) ₂₀	F(9)	19	8.87	0.92	0.09	0.83	4.80	0.88	0.11	0.88
mSbCIR283	(CT) ₈ (GT) ₉	G(10)	14	5.08	0.79	0.12	0.67	2.52	0.83	0.15	0.79
Xtxp321	$(GT)_4(AT)_6(CT)_{21}$		15	3.31	0.74	0.11	0.63	1.82	0.84	0.14	0.68
gpsb123 (CA)7(GA)5		H(8)	6	2.69	0.64	0.06	0.58	5.51	0.92	0.08	0.56
Compound nucleo	Compound nucleotides		13.5	5.0	0.77	0.10	0.68	3.66	0.87	0.12	0.73
Overall mean			10.5	3.72	0.69	0.10	0.59	3.66	0.83	0.15	0.60
SD			6.03	2.20	0.06	0.12	0.18	1.40	0.04	0.03	0.20

LG Linkage group, N_a Total number of alleles per locus, N_e Numbers of effective alleles per locus, N_m Gene flow per locus, H_t Total gene diversity, H_e Average gene diversity within genotypes, PIC Polymorphic information content, F Fixation index, SD Standard deviation

Table 5.4.2 Genetic diversity within and among sorghum genotypes classified by collection zones for the overall sample of 200 genotypes

Zone/district					Genetic p	arameter								
•	N _a	N _e	A _r	H _t	H _o	H _e	F	t	I	Р				
				Administ	rative Zor	ne								
South Welo	9.10	3.72	3.628	0.738	0.097	0.642	0.855	0.078	1.442	100.0				
North Welo	8.53	3.62	3.598	0.756	0.119	0.637	0.828	0.094	1.416	100.0				
Wag-Hemra	6.73	3.53	3.522	0.716	0.091	0.625	0.865	0.072	1.329	100.0				
		District of collection												
Ambasel	7.57	3.55	3.55	0.735	0.099	0.637	0.851	0.080	1.387	100.0				
Kalu	7.97	3.66	3.68	0.762	0.115	0.648	0.834	0.090	1.417	100.0				
Bugina	4.33	2.93	3.17	0.703	0.103	0.600	0.834	0.090	1.111	96.7				
Gubalfto	7.40	3.72	3.69	0.745	0.101	0.644	0.849	0.081	1.400	100.0				
Kobo	4.23	2.84	2.83	0.691	0.110	0.581	0.808	0.106	1.068	100.0				
Sekota	4.83	2.99	3.34	0.669	0.088	0.581	0.858	0.076	1.133	96.7				
Ziquala	4.97	3.34	3.23	0.724	0.095	0.629	0.851	0.081	1.226	100.0				
Control	2.73	2.41	2.40	0.669	0.100	0.569	0.782	0.122	0.844	90.0				
Overall	5.50	3.18	3.72	0.712	0.101	0.611	0.834	0.090	1.198	97.9				
SD	0.23	0.12	0.45	0.033	0.008	0.030	0.024	0.015	0.187	3.30				

 N_A number of different alleles, N_e number of effective alleles, A_r allele richness, H_t Total gene diversity, H_o gene diversity within genotypes, He gene diversity among genotypes, F fixation index, t out-crossing rate t = (1 - F)/(1 + F), I Shannon's information index, P percent polymorphic loci

5.4.2 Distance-based population differentiation

The F-statistics over the 200 genotypes indicated that the mean inbreeding coefficient, F was 0.83 ± 0.024 , implying that the sorghum landraces collected from different districts were highly homozygous. There were high levels of inbreeding within landraces, with a mean value $F_{IS} = 0.81 \pm 0.015$. Genetic differentiation among genotypes was insignificant with a mean value of $F_{ST} = 0.06 \pm 0.03$. The possibility of two randomly sampled alleles in a given genotype

collections to be different was higher than 61% (mean gene diversity = 0.61). The highest overall gene diversity was recorded for genotypes collected from Kalu and Gubalafto (0.76) and the lowest value of H_t was detected for control genotypes. However, considering the administrative zones as a whole, no significant variation was observed in gene diversity.

The high genetic variability observed among genotypes signified that the genotypes were not under artificial selection and that there was a continuous exchange of alleles among genotypes (Manzelli, et al., 2007). Farmers in Ethiopia practice mass selection, i.e., healthy and big panicles are selected in a field every year and this seed is used in the following planting season. According to Harlan (1975), such practices exert a particular balance of selection pressure and allow for genetic variability within populations. Farmers select panicles based on appearance, not considering the genetic purity (Teshome et al., 2007). High genetic polymorphism was observed within genotypes, showing that on average, 98% of the loci were polymorphic. The proportions of polymorphic loci were 90% in the control genotypes and 100% in the gremplasm from Ambasel, Kalu, Gubalafto and Ziquala. Similarly Shannon's Information Index was slightly higher for Ambasel, Kalu and Gubalfto whereas the control genotypes had minimal values.

Genetic distance is the measure of the extent of genetic differences that exist between individuals, populations, or species, which is measured by some numerical quantity and can be described by allelic variation (Nei, 1987). The genetic distance calculated using the 30 SSR markers based on Nei (1987) indicated that the average unbiased Nei's genetic distance calculated among and within collection districts was higher (0.107 – 0.146) between the control genotypes and genotypes from Bugina, Kobo, Sekota and Ziquala but low between genotypes from Gubalafto, Ambasel and Kalu (0.01) (Table 5.4.3).

Table 5.4.3 Unbiased Nei genetic distances and identity among and between 200 sorghum genotypes collected from seven districts along with the control lines

		Genetic identity (GI)											
District	AM	KA	BU	GBL	KO	SE	ZQ	CO					
Ambasel		0.984	0.964	0.986	0.956	0.970	0.975	0.916					
Kalu	0.016		0.948	0.986	0.959	0.963	0.960	0.929					
Bugina	0.036	0.053		0.949	0.923	0.926	0.937	0.864					
Gubalfto	0.014	0.014	0.052		0.964	0.964	0.963	0.918					
Kobo	0.045	0.042	0.081	0.037		0.938	0.942	0.898					
Sekota	0.030	0.038	0.077	0.037	0.064		0.944	0.891					
Ziquala	0.026	0.041	0.065	0.037	0.060	0.058		0.899					
Control	0.088	0.073	0.146	0.086	0.107	0.115	0.107						
			G	enetic dis	tance (GD))							

The lowest genetic distance was observed between South and North Welo (0.008) and the highest between North Welo and the control genotypes (0.06). Similarly, genetic identity was higher among zones (0.94 - 0.99); however it was low between the control genotypes and most of the districts (Table 5.4.3). The narrow genetic distance among collection districts could be attributed to the racial specificity of most of the local genotypes belonging to durra race, combined with the high level of gene flow among districts

Genetic differentiation in the Ethiopian sorghum landraces collected from different zones analyzed with FSTAT revealed that F_{ST} value among pairs of populations ranged from 0.01 to 0.09 with an overall mean of 0.06 (Table 5.4.4). Population differentiation was relatively high between the control genotypes (F_{ST} ranged from 0.06 to 0.09) and the different districts of collection. However, population differentiation among the districts of collection was very similar to one another but was low (F_{ST} ranges from 0.01 to 0.05). Dje *et al.* (2000) reported an F_{ST} 0.68 among genotypes of cultivated sorghum. Whereas Ganapathy *et al.* (2012) reported an F_{ST} value of 0.51 between rainy and post rainy genotypes. Based on allozyme data, Ayana *et al.* (2001) reported an F_{ST} = 0.44 for landraces collected from three altitudinal zones of Ethiopia, while Zongo *et al.* (2005) measured an F_{ST} value of 0.81 based on landraces collected from Burkina Faso. However, the present estimate of F_{ST} is much lower than values reported by the previous authors.

Table 5.4.4 Pairwise genetic differentiation and gene flow among and between sorghum genotypes collected from seven districts along with the controls

				Gene flo	w (Nm)*			
Districts	AM	KA	BU	GBL	KO	SE	ZQ	CO
Ambasel		23.28	9.90	26.22	8.710	10.84	12.80	3.810
Kalu	0.011		7.554	28.105	8.697	10.494	10.242	4.163
Bugina	0.025	0.032		8.057	4.894	5.167	5.864	2.493
Gubalfto	0.009	0.009	0.030		9.472	10.573	11.139	3.959
Kobo	0.028	0.028	0.049	0.026		5.872	6.252	2.890
Sekota	0.023	0.023	0.046	0.023	0.041		6.369	3.058
Ziquala	0.019	0.024	0.041	0.022	0.038	0.038		3.108
Control	0.062	0.057	0.091	0.059	0.080	0.076	0.074	
-			Ger	netic differe	entiation (F _{st})		

^{*} Nm = $0.25 (1-F_{ST})/F_{ST}$

The low genetic differentiation among the population can be caused either by frequent occurrence of gene flow, or low genetic drift due to a high effective population size (Dje *et al.*, 1999). Correspondingly, high level of gene flow (23 - 28) exists between neighboring districts such as Ambasel, Kalu and Gubalfto. Some level of gene flow was also observed between Gubalafto, Sekota and Ziquala. The low F_{ST} in this study can be explained by the observed high level of gene flow, which leads to genetic homogeneity among the different districts. Teshome *et al.* (1997) reported the existence of gene flow among bordering districts. This could be further explained by the movement of farmers for trading and seasonal labour, and informal seed exchange among them.

Cluster analysis based on genetic dissimilarity using the neighbor-joining method in DARwin 5.0 classified the 200 sorghum genotypes into two distinct major clusters (Figure 5.4.1). However, the cluster analysis generated using SSR markers did not correspond with the predefined population structure based on districts of collection. This is expected because most of the genotypes were collected from lowland agro-ecologies of the different districts in the North East Ethiopia. Cluster I consisted of 52% of the total genotypes and composed of three sub-clusters (2, 3 & 4) (Figure 5.4.1). The second cluster contains 49% of the total genotypes and it also comprised of three sub-clusters (1, 5 & 6). In Cluster I, landraces from Bugina (67%), Gubalafto

(55%) and Ambasel (54%) were more frequent. On the other hand, landraces from Kalu (59%) and Sekota (56%) were more common in Cluster II. The three controls (AS-27, AS-103 and *Jigurte*) were also grouped in this set of genotypes.

Sixteen sweet sorghum genotypes were included in this study and they were widely scattered in different sub-clusters. In the present study, the sweet and grain sorghum varieties were not separated from each other and no specific pattern was observed. Ali *et al.* (2008) and Kimberley *et al.* (2007) in their studies using RLFP and SSR markers, also reported weak differentiation between the two types of sorghum (sweet stem and grain) and they could not be significantly separated. The sweet stem sorghum (SS) line AS-27 from South Africa was grouped with 239192A and 239234 SS landraces from Ethiopia while AS-103 sweet sorghum line from the U.S.A. was grouped with 72583A in Sub-clusters 1 & 4, respectively. Five of the sweet sorghum genotypes (69286-A, 73031, 73041, 73059, & 200538) from Ethiopia were grouped in Sub-cluster 2.

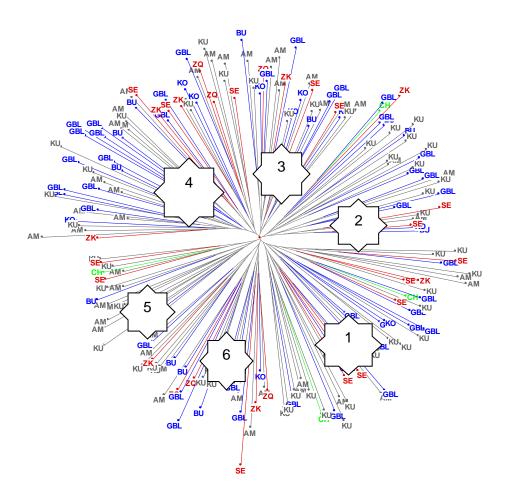


Figure 5.4.1 Neighbor-joining dendograms depicting genetic relationship between sorghum genotypes collected from three zones and seven districts of lowland environments in Ethiopia with respect to four controls (two sweet and two grain sorghum lines).

Different line shading represents different collection zones.

5.4.3 Model-based population differentiation and structure

The model-based structure analysis using STRUCTUR confirmed the existence of two distinct sets of genotypes, which was similar with NJ cluster analysis (Figure 5.4.2A). However, both the number and list of germplasm in the two analyses were not identical. In the first analysis, the model assumes that individuals may have mixed ancestry and inherited a fraction of their genome from ancestors in K populations. The model assigned individuals to a given population based on their genotypes and allele frequencies (Pritchard *et al.*, 2000). At K = 2, the 200 genotypes were clustered into two discrete groups with moderate level of admixtures. Further increasing the population to K = 3 did not increase separation except escalating the level of

admixtures to the two sub-groups. Population structure analysis on the basis of geographic origin did not reveal any meaningful population structure (Figure 5.4.2B). This result is congruent with AMOVA analysis that showed low differentiation among districts. It is apparent that no further sub-grouping in the study materials could be modeled and that the optimum population structure predicted by the software was at K = 2.

Within the two inferred populations were Set A which contained 131 landraces and dominated by Kalu landraces (31%), and Set B only consisted of 35% (69) of the landraces used in this study. The genetic similarity between the inferred populations was 0.082. Set A was dominated by Degalet types which have compact heads, long cycles (6 months to mature), and are late maturing and relatively high yielding landraces, whereas Set B was mainly composed of *Jigurte* types which are short cycle (4 months to mature) and early maturing and low yielding landraces. The AMOVA clearly indicated a significant difference between the two inferred populations (Table 5.4.5). The variance among the two populations was significantly higher compared to the variance accounted for by geographic domains which contributed only about 14% of the total variation. The within landrace and among landraces differentiation accounted for 16% and 70% of the total variation, respectively. Much higher fixation index estimate ($F_{ST} = 0.14$, P = 0.001) was observed when the genotypes were classified into two sub-groups. The two distinct subgroups may have resulted from farmers' selection for adaptation for specific production niches, cooking value and market orientation (Asfaw et al., 2009). The occurrence of a moderately strong genetic structure ($F_{ST} = 0.137$) between the two populations indicated that the two groups were genetically different. This will allow breeders to select and fix unique alleles between populations (Hartl and Clark, 1997). Among the numerous explanations forwarded as the cause of transgressive segregation in plants, complementary gene action is a more popular explanation (Rieseberg et al., 1999). Crosses between genetically divergent inbred lineages may result in transgressive segregation. This is because different parental lineages are often fix different sets of alleles with opposing effects. In this particular study, strategic crossing between the two divergent populations may allow breeders to identify transgressive segregants.

Table 5.4.5 Analysis of molecular variance (AMOVA) of 200 sorghum landraces based on the two inferred population using 30 SSR markers

Source	Sum of squares	Variance components	% variation	F-statistics
Among populations	81.277	1.353	13.75	$F_{ST} = 0.137$
Among landraces	17.437	6.946	70.56	$F_{IS} = 0.818$
Within landraces	1.545	1.545	15.69	$F_{IT} = 0.863$
Total	3842.88	9.844	100	

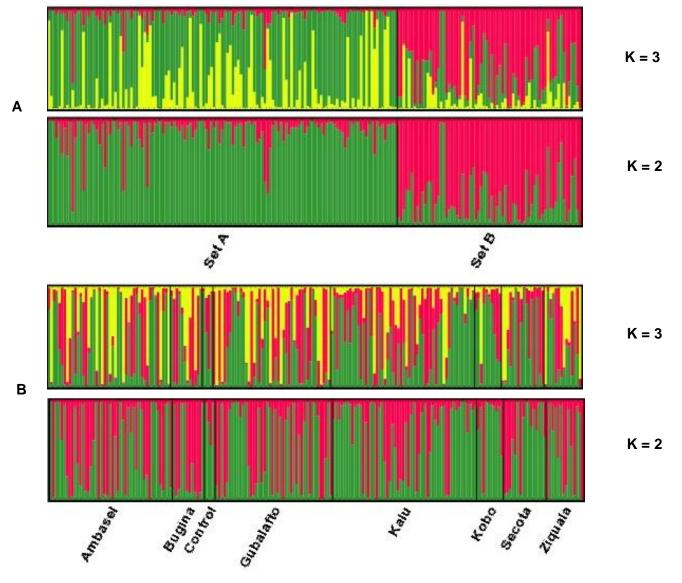


Figure 5.4.2 Population genetic structure for 200 sorghum landraces from North Eastern Ethiopia at K = 2 and K = 3 based on estimates of Q. (A) based on districts of collection; (B) based on inferred sets

The SSR markers used in this study were sampled from ten different linkage groups of the sorghum genome (Dean *et al.* 1999), which collectively had a moderate to high PIC values that provided information to uniquely identify most of the genotypes profiled in this study. All the 30 SSR markers were polymorphic, confirming that each marker would be effective and valuable for genetic analysis. The level of polymorphism in the 200 sorghum landraces used in this study was found to be moderate to high with a mean PIC value of 0.6. The degree of precision of molecular markers in estimating genetic relatedness between genotypes is strongly dependent on the type and number of markers that are used and their genome coverage (Powell *et al.* 1996; Agrama and Tuinstra, 2003; Garcia *et al.*, 2004; Menz *et al.*, 2004; Geleta *et al.*, 2006; Perumal *et al.*, 2007). The use of SSRs for the analysis of population genetics and genetic diversity in sorghum could reduce the limitations in indentifying polymorphisms and result in more complete genomic coverage (Perumal *et al.*, 2007).

The SSR analysis result suggested weak district and zonal genetic differentiation among the tested sorghum landraces. This can be attributed to high level of gene flow among districts and zones within the North Eastern Ethiopia. In the preceding chapters (Chapter 3 and 4), it was observed that farmers maintained a large number of landraces in a single plot, based on combination of selection criteria in order to cope with the diverse environmental conditions (Teshome et al., 2007). Additionally, farmers exchanged seeds through gifts and via markets, to renew old seed stocks or to acquire new varieties. Consequently, there may be a continuous exchange of genes among genotypes. Apart from gene flow, the landraces were collected in the lowland agro-ecologies where moisture stress is the major production bottleneck and farmers probably have similar selection preferences in accordance with ecological adaptation. This led to weak genetic differentiation among the districts of collection. The weak differentiation of germplasm on the bases of districts and zones, was also confirmed by AMOVA that a large proportion of the variation was found among landraces within districts. Similar results were reported from studies based on allozymes (Ayana et al., 2001), microsatellite markers (Dje et al., 2000) and RAPD (Menkir et al., 1997) and all the authors pointed out that geographic variations do not make major considerable contributions to genetic differentiation among sorghum genotypes. Aldrich and Doebley (1992) also reported the low correlation between genetic and geographic distances in cultivated sorghum. This suggested that sorghum genotypes from different geographic origins can be considered to come from roughly the same gene pool when devising germplasm collection and breeding activities.

5.5 Conclusion

The result suggested that there is considerable genetic variability among sorghum landraces found in the drought-prone lowlands of the North Eastern Ethiopia. However, this variation was not correlated with geographic origins of the genotypes. This signifies that the high phenotypic variability observed in Chapter 3 and 4 of this thesis was associated with high genetic diversity among genotypes. Additionally, low differentiation among collection sites and high variation within sites suggests that large random collection would capture most of the genetic variation within sorghum in the target area. Given the high level of gene flow among genotypes and high level of within-population variation, the Ethiopian farmers' traditional agricultural production system has played a vital role in maintaining and directing genetic diversity and evolution. Case et al. (2005) studied genetic diversity analysis of sorghum using 98 SSR markers and found that local landraces captured 86% of the total variation found in wild species. Similarly Deu et al. (2010) pointed out the role of farmers in the management and preservation of genetic diversity over time. The high genetic variability among landraces provide enough genetic plasticity to adapt to the diverse environmental conditions in the tropical areas (Manzelli et al., 2007), and allow them to circumvent crop failure by reducing vulnerability in environmental stresses (Ceccarelli et al., 1992).

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CHAPTER 6

Combining Ability and Heterosis in Lowland Sorghum [Sorghum bicolor (L.) Moench] Landraces under Moisture Stress Condition

6.1 Abstract

Moisture stress is one of the major sorghum production constraints limiting genetic gain through breeding in the arid and semi-arid tropics. Developing high yielding and stable sorghum hybrids is one of the novel approaches to alleviate the adverse effect of drought and to boost productivity. The objectives of this study were to determine heterosis and combining ability of lowland landraces for grain yield, yield components and drought tolerance and to identify suitable parents for future sorghum hybrid cultivar development. Thirty two selected lowland sorghum landraces were crossed with four Cytoplasmic Male Sterile (CMS) A-lines of different cytoplasmic sources and crossed in a line x tester mating design to generate 128 F1s. Field evaluations were conducted under moisture stress and well-watered conditions involving parents, F1s and four checks. The general combining ability (GCA) and specific combing ability (SCA) effects were estimated for ten agronomic traits using the line x tester genetic design. Highly significant GCA effects were observed among landraces for all the considered traits. while non-significant GCA effects were found among testers except for above ground biomass. The ratio of GCA to SCA was equal to unity for days to 50% anthesis and panicle length, whereas intermediate values were ontained for days to 50% maturity and stay-green scores. However, the ratio was much lower than unity for grain filling duration, plant height, panicle weight, 100-garin weight, above ground biomass and grain yield. Thus, non-additive gene action was predominant in controlling plant height, grain yield, above ground biomass, grain filling duration, 100-seed weight and panicle weight. On the other hand, predominance of additive gene action was found for days to anthesis and panicle length. Male parents 214838-A, 242039, 75454, 73056, and 242050-B and female parents ICSA 749 and ICSA 756 displayed positive and significant GCA effects for grain yield and yield components. These lines also showed highly significant SCA effects and heterosis for grain yield, which are recommended as potential parents for inclusion in sorghum hybrid breeding programme to exploit heterosis.

Key words: Combining ability, drought tolerance, heterosis, moisture stress, *Sorghum bicolor*

6.2 Introduction

Sorghum [Sorghum bicolor (L.) Moench] is one of the most important cereal crops grown in arid and semi-arid regions of the world. It is the fifth most important cereal crop worldwide after wheat, rice, maize, and barley (FAO, 2006). Sorghum has high grain yield potential comparable to other cereals such as rice and wheat under optimal conditions. The ability of the crop to withstand considerable drought stress and providing reasonable yields under adverse environmental conditions has proven its importance as a food security crop in arid and semi-arid tropics of the world. In stress environments, sorghum is the predominant crop and survives well under limited agricultural inputs than any major cereals such as maize and wheat (McGuire, 2008). The crop is mainly grown for its grain as human food, feed for livestock, for production of local beverages, while the stalk is also used for animal feed and construction material (Teshome et al., 1999; McGuire, 2000).

In the past sorghum breeding in Ethiopia has mainly relied on selecting improved inbred lines from exotic sources. In the last 10 years, with the integration of improved varieties and appropriate management practices sorghum production and yield has almost doubled (CSA, 2012). Several improved varieties have been developed and released with better drought tolerance and relatively high yield potential. However, on average, 1.5 t/ha of grain yield is obtained (CSA, 2012), which is much lower than the potential yield of 7 to 9 t/ha obtained under intensive management (Borrell *et al.*, 2000). In the country there is a growing interest of improving agricultural production and productivity for supplying food for the ever increasing population. This can only be achieved through increasing productivity per unit area. Given the advances in yield improvement in the past, further improvement through phenotypic selection of lines has not been promising under drought conditions. This challenge suggests the necessity of sorghum breeding and cultivar development with improved productivity per unit area.

Development of high yielding, locally adapted and famers-preferred hybrids is one of the approaches to alleviate the inherent low productivity of crops in the semi-arid tropics. In Africa, the use of hybrid sorghum is lagging behind as compared to a long history of hybrid production in the US and Asia. For example, in the last 30 years the use of hybrid rice in China enabled reduction of a total amount of land by 20% while yield have been increased by 48% (FAO, 2004). Similarly, India through the use of hybrid sorghum has achieved a yield increase of 80%

by reducing 37% of the total amount of land planted to sorghum during the same period (Mishra *et al.*, 1992). Further, hybrid cultivars have been reported to be more productive and stable than pure line varieties (Kenga *et al.*, 2004). Doggett (1961) reported the consistent performance of sorghum hybrids over a range of environments. Further, sorghum grain yield advantage of 50 to 100% has been reported in hybrids as compared to their parents over a range of environments (Blum *et al.*, 1990; Haussmann *et al.*, 1999).

The discovery of cytoplasmic male sterility system promoted sorghum research towards the commercial production of hybrid cultivars (Stephens and Holland, 1954). The existences of different cytoplasmic sources that induce male sterility apart from milo-kafir cytoplasmic male sterility system have become widely available (Quinby, 1980; Worstell *et al.*, 1984). This has enabled sorghum breeders to exploit the cytoplasm diversity available in different races and species of sorghum from diverse geographic locations (Pedersen *et al.*, 1998). Similarly, the continued discovery of alternative male sterile cytoplasm would effectively allow the construction of heterotic pools different from the common R/B-lines defined in terms of A₁ cytoplasm (Pedersen *et al.*, 1998).

Success of any breeding programme depends on proper characterization and quantification of the available genetic resources (Betran *et al.*, 2003). In Eastern Africa, more than 70% of sorghum is cultivated in the dry and marginal areas (Mukuru, 1993) suggesting the crop has been adapted to such adverse environments. This, in turn, increases genetic plasticity of the crop to flourish under harsh environmental conditions. A wide genetic diversity of sorghum has been developed over the years as a result of continued active selection by farmers (Gebrekidan, 1973, Ayana and Bekele, 2000). This offers breeders greater opportunity to select superior parental genotypes for the production of commercial hybrids.

The sorghum hybrid development research programme in Ethiopia is at its infant stage. Attempts have been restricted to characterizing combining ability and assessing the heterotic patterns of exotic parental lines (Adugna and Tesso, 2006; Degu *et al.*, 2009). The potential of the available genetic diversity within local landraces for hybrid cultivar development is not yet been exhaustively assessed. The genetic potential of any material is largely determined by its general and specific combing ability for yield and yield related traits under diverse environmental conditions. Therefore, this study was aimed to determine heterosis and combining ability of lowland landraces for grain yield, yield components and drought tolerance and to identify suitable parents for future sorghum hybrid cultivar development.

6.3 Materials and methods

6.3.1 Plant material and crosses

Thirty two selected landraces were crossed to four cytoplamic male-sterile lines in a line x tester mating design. Preliminary adaptation and crossing study was conducted for a total of 12 CMS (4 A1, 2A2, 3 A3 & 3 A4) lines. Among those only 4 CMS lines, which were well adapted and best in seed setting, were selected. The Cytoplasmic Male Sterile lines (CMS) (ICSA 101, ICSA 749, ICSA 743 and ICSA 756) were selected from different cytoplasmic sources (A1, A2, A3 and A4) and used as females (Table 6.3.1). The 32 lines originated from different geographic origins mainly grown in lowland drought prone areas in Ethiopia and selected from prior genetic and agro-morphological studies based on relatively better yield and yield components under moisture stress. These lines were kept homogenous through continued selfing and selection and served as male parents. The 128 F1s, 32 male, four B lines and four checks were field evaluated under two water regimes. The B lines were used in place of their male sterile counterparts. The local check (Jigurty) is an early maturing, tall plant height (~290 cm) and compact headed variety commonly grown by farmers when there is moisture stress. The standard checks (Girana1 and Miskir) are early maturing, relatively short (120 - 200 cm) and semi loose head types which were released by Sirinka Agricultural Research Center (SARC) for their moisture stress tolerance. The fourth check (ICSV 111) was developed for terminal moisture stress and obtained from the International Crops Research Institute for the Semi-Arid Tropics (ICRISAT).

6.3.2 Field experiments and data collection

Field experiments were conducted at Kobo research sub-center under irrigation and rainfed conditions. The Kobo research site is situated at $12^{0}9$ ' N latitude and $39^{0}38$ ' E longitude with an altitude of 1400 meter above sea level (masl). It receives a mean annual rainfall of approximately 650 mm with the length of growing period 60 - 120 days showing high coefficient of variability, usually >30% with regard to quantity, onset and cessation of rainfall. The soil is sandy, of shallow depth, light textured, low in fertility and organic matter.

The experiments were laid out in 12 x 14 alpha designs with two replications. Each genotype was planted in three rows of 3 m long with between row spacing of 75 cm. The plots were

separated by 1 m paths and seeds were manually drill planted into the rows at a seed rate of 10 kg/ha. At approximately 25 days after planting, the seedlings were thinned to an intra-row spacing of 30 cm leaving one plant per hill and ten plants in a row. Planting for the irrigation experiment was carried out on June 18, 2012 and irrigation was applied every week from planting to grain filling stage. Planting was delayed by 20 days for the rainfed experiment to ensure that the materials were exposed to terminal drought stress.

Table 6.3.1 List of sorghum genotypes used in the study indicating fertility status, origin and pedigree.

No.	Genotype	Fertility ^a	Origin	Pedigree
1	71160-A	R	IBC, Ethiopia	Landrace
2	72472	R	IBC, Ethiopia	Landrace
3	72482	R	IBC, Ethiopia	Landrace
4	72572	R	IBC, Ethiopia	Landrace
5	72578-B	R	IBC, Ethiopia	Landrace
6	73056-B	R	IBC, Ethiopia	Landrace
7	73059	R	IBC, Ethiopia	Landrace
8	75454	R	IBC, Ethiopia	Landrace
9	200538	R	IBC, Ethiopia	Landrace
10	200654	R	IBC, Ethiopia	Landrace
11	211239-B	R	IBC, Ethiopia	Landrace
12	214855	R	IBC, Ethiopia	Landrace
13	232036	R	IBC, Ethiopia	Landrace
14	237260	R	IBC, Ethiopia	Landrace
15	239156	R	IBC, Ethiopia	Landrace
16	239175	R	IBC, Ethiopia	Landrace
17	239208	R	IBC, Ethiopia	Landrace
18	242039-B	R	IBC, Ethiopia	Landrace
19	242047	R	IBC, Ethiopia	Landrace
20	244712	R	IBC, Ethiopia	Landrace
21	244715	R	IBC, Ethiopia	Landrace
22	244727	R	IBC, Ethiopia	Landrace
23	244733	R	IBC, Ethiopia	Landrace
24	244735-A	R	IBC, Ethiopia	Landrace
25	214838-A	R	IBC, Ethiopia	Landrace
26	214838-B	R	IBC, Ethiopia	Landrace
27	239167-B	R	IBC, Ethiopia	Landrace
28	242049-B	R	IBC, Ethiopia	Landrace
29	242050-A	R	IBC, Ethiopia	Landrace
30	244725-A	R	IBC, Ethiopia	Landrace
31	244725-B	R	IBC, Ethiopia	Landrace
32	69286-A	R	IBC, Ethiopia	Landrace
33	ICSA 101	A1	ICRISAT, India	(Ind. Syn. 89-1 x Rs/R 20-682)5-1-3
34	ICSA 749	A2	ICRISAT, India	(IS 10036 x ICSR 33)9-3-2-1-1
35	ICSA 743	A3	ICRISAT, India	[(ICSB 101 x TRL 74/C57) x PM 1746B]2-5-1
36	ICSA 756	A4	ICRISAT, India	(ICSB 6 x IS 18432)13-1-2-2-1
37	Girana1	C	SARC, Ethiopia	CR:35XDJ1195 x N-13
38	Jigurty	C	Local variety	Local variety
39	Miskir	C	SARC, Ethiopia	PGRC/E#69441 x P-9401
40	ICSV 111	С	ICRISAT, India	ICSV 111 inc

^a R = male parents with fertility restorer gene; IBC = Ethiopian Institute of Biodiversity Conservation; A1 to A4 denote Cytoplasmic Male Sterile lines with A1 to A4 cytoplasm systems; ICRISAT= International Crops Research Institute for the Semi-Arid Tropics

Fertilizer was applied at the rate of 100 kg/ha diammonium phosphate (DAP) and 50 kg/ha urea as recommended for sorghum in the lowland of Ethiopia. All the DAP was applied at the time of planting, while urea was applied in a split application, 45 days after planting. To minimize variability and maximize genetic expressions of the genotypes, the field was kept weed free by regular hand weeding and other standard agronomic practices were followed as required.

For recording the data five plants from the middle rows were tagged in every plot and covered with bird proof bags to protect them from bird damage. Ten agronomic traits were measured in all genotypes at the two environments. Days to 50% anthesis (DTA) was measured as the number of days from planting till 50% of the plants in a plot reached half bloom stage. Days to 50% maturity (DTM) was measured as the number of days from planting until 50% of panicles from the main shoot in a plot reached the black layer stage. Plant height (PH) was recorded as the average height in centimeters from the ground to the tip of the panicle at maturity. Panicle length (PL) was recorded as the average distance in centimeters from the base of a panicle branch to the tip of the panicle. Above ground biomass (BIOM) was recorded as the average fresh weight of five randomly selected plants and expressed as tons per hectare. Panicle weight (PW) was measured in grams as the average weight of five heads per plot. Hundred seed weight (HWT): measured in grams as the weight of randomly sampled 100 grains at 12.5% moisture content. Grain yield (GY) was measured as the weight of the threshed grains of five plants at 12.5% moisture content and expressed as tons per hectare. Stay-green (SG) was recorded as visual scoring using 1-5 scale where 1, as complete senescence and 5, as very slight senescent. Data was then recorded from the five selected plants except for days to 50% anthesis and maturity, which was recorded on whole plot basis. Grain filling duration (GFD) was calculated by subtracting number of days to maturity from days to anthesis.

6.3.3 Data analysis

Statistical analysis was performed using SAS statistical software systems using the GLM and PROC MIXED procedures included in SAS 9.1.3 (SAS Institute 2003). The two water treatments were considered as two environments. The combined analysis over environment was done after homogeneity test of variances using Bartlett's test for each of the studied agronomic traits.

The variance components were estimated from their respective expected mean squares as the methodologies described by Kempthorne (1957). The total genotypic variance was partitioned into variation due to lines, due to testers, and that due to the interaction between line and

testers. Genotypes and environments were considered as fixed effects and replications as random effects, then a combined analysis over environments were estimated from linear additive model expressed as:

$$Y_{ijklm} = \mu + g_i + g_j + s_{ij} + e_k + (ge)_{ik} + (ge)_{jk} + (se)_{ijk} + \varepsilon_{ijk}$$

Where Y_{ijklm} = the observed phenotypic value from each experimental unit

 μ = the population mean

 g_i = the GCA effect of the i^{th} parental line

 g_i = the GCA effect of the j^{th} tester line

 s_{ii} = the SCA effect of the ij^{th} F₁ hybrid

 e_k = the effect of the k^{th} environment

 $(ge)_{ik}$ = the interaction effect between the i^{th} line and the k^{th} environment

 $(ge)_{jk}$ = the interaction effect between the $m{j}^{th}$ tester and the $m{k}^{th}$ environment

 $(se)_{ijk}$ = the interaction effect between the ij^{th} F₁ hybrid and the k^{th} environment

 ε_{iik} = the residual effect

Adjusted means for each agronomic trait were obtained using the *Ismeans* option in the PROC GLM model procedure in SAS 9.1.3 (SAS Institute 2003). The adjusted means were then used to the subsequent calculations of general combining ability (GCA) effects of parents, specific combining ability (SCA) effects of hybrids and heterosis. The statistical significance of the GCA and SCA effects was tested using the t-test, and their corresponding standard errors were estimated using a line x tester analysis described by Kempthorne (1957). Estimates of GCA variances (δ^2_{GCA}) and SCA variance (δ^2_{SCA}) were obtained using formula suggested by (Kempthorne 1957). The relative importance of GCA was determined from the ratio of mean square components associated with GCA and SCA variances as described by Baker (1978). The closer the value to unity, the higher the predictability of progeny performance based on GCA effects alone.

General combining abilities (GCA) effects were computed as:

$$GCA_{i} = \left(\frac{y_{i.}}{rf}\right) - \left(\frac{Y}{rmf}\right)$$

$$GCA_{j} = \left(\frac{y_{.j}}{rm}\right) - \left(\frac{Y}{rmf}\right)$$

Where,

 GCA_i and GCA_j = the general combining ability of the i^{th} line and j^{th} tester, respectively. y_i and $y_{.j}$ = the grand total of the i^{th} line mated with all testers and the j^{th} tester mated with all lines, respectively. Y = the grand total of all crosses, r = the number of replication, m = the number of lines and f = the number of testers

Specific combining ability effects were calculated as:

$$SCA_{ij} = \left(\frac{y_{ij}}{r}\right) - \left(\frac{y_{i.}}{rf}\right) - \left(\frac{y_{.j}}{rm}\right) + \left(\frac{Y}{rmf}\right)$$

Where SCA_{ij} = the specific combining ability effect of ij^{th} cross, y_{ij} = the grand total for cross i^{th} line and the j^{th} tester, $y_{i.}$ = the grand total of lines for the ij^{th} cross, $y_{.j}$ = the grand total of testers for the ij^{th} cross, Y = the grand total of all crosses, r = the number of replication, m = the number of lines and f = the number of testers

Mid- parent heterosis (MPH) for the studied agronomic traits were calculated to estimate the hybrid advantage compared to the mean of the parents. This provides an estimate of the mean directional dominance of alleles for a given character. MPH values were calculated as:

$$MPH = \left(\frac{F_1 - MP}{MP}\right) \times 100$$
, where $MP = \left(\frac{P_1 + P_2}{2}\right)$, in which P_1 and P_2 are mean of parent 1 and 2, respectively

 F_1 = the mean of F_1 hybrid performance

High parent heterosis (HPH) was estimated to determine the hybrid advantage compared to that of the better parent. In the case of HPH, the assumption is that there is dispersion for dominance allele between the parents. HPH values were calculated as:

$$HPH = \left(\frac{F_1 - HP}{HP}\right) x 100$$
 Where HP = the mean of the best parent

Standard heterosis (SH) was used to estimate genetic gain or superiority of the hybrids to standard varieties in a given area. SH was computed as:

$$SH = \left(\frac{F_1 - SP}{SP}\right) x 100$$
 Where SP = mean of the standard variety

6.4 Results and discussion

6.4.1 Combined analysis of variance

Combined analysis of variance for five growth and phenological parameters that are associated with drought tolerance and five yield and yield component parameters measured across stress and non-stress environments are presented in Table 6.4.2. The coefficient of variations (CV) ranged between 2.6 and 24.7% and it was higher for biomass, panicle weight and grain yield. The CVs were higher for the non-stress environment than stress environments (data not shown) and this may be because of variability in water distribution due to uneven furrow irrigation across the field. Environment had a significant effect on the expression of all agronomic traits studied. Entries and environment by entry interactions were significant for all the traits studied indicating that the genotypes were diverse in performance and in their responses to the environments were different. This suggests that crops adapted to arid and semi-arid tropical environments, such as sorghum has genetic plasticity which adjusts their phenological growth in response to the prevailing environmental conditions.

Partitioning of the entry mean squares into variation attributed to parent, hybrids and checks demonstrated that the variation within each group and the response with the environment was highly significant for most of the traits. This suggested that in order to get reliable estimates of the main effects of the characters, it is worthwhile to carry out experiments in various environments. Variation among the checks was significant for all the traits except stay-green and panicle weight (Table 6.4.1). The checks included three improved cultivars (*Girana1*, *Miskir* and ICSV 111) developed for moisture stress environments with characteristics such as short plant stature and early maturity, while the local check (*Jigurty*) is a local pure line variety grown by most subsistence farmers for its early maturity and high biomass yield.

Further partitioning of parental mean squares into male, female and male versus female revealed that the main and the interaction effects of males were highly significant for most of the traits, while the female effects showed non-significant differences for most of the traits. This suggests that wide range of variation existed among the male parents, the landraces, and not the female parents (testers). The mean squares due to male vs female were highly significant for most traits except grain filling duration; panicle weight and grain yield (Table 6.4.1). The low

variation observed among testers is expected due to the fact that these lines have gone through successive breeding and selection for high yield, short plant stature and other desirable agronomic traits. Additionally, these testers are improved parental lines developed as seed parents with certain characters such as proper nicking and small grain filling duration for practical commercial hybrid seed production programmes. However, the paternal lines were landraces maintained by farmers for decades with no selection. Similarly, main effects of the mean square values for hybrids and parents versus hybrids were highly significant for most of the traits; however the interaction effects with the environment were not significant for most traits except days to 50% maturity and panicle weight.

The observed highly significant main effects due to hybrids for all traits studied, suggests that further partitioning of the genetic variance in to paternal and maternal combining ability effects could give more information about the superiority of the lines and testers. Variance component estimates along with analysis of variance, relative proportion of the lines, testers and hybrids to the total variance for ten agronomic traits measured across two environments are presented in Table 6.4.2. The mean squares due to GCA of the lines and testers and the SCA of the crosses were highly significant for all traits, indicating that both the additive and non-additive genetic effects controlled these traits.

6.4.2 General combining abilities

The GCA variance was highly significant among lines used as male for all the traits while it was non-significant among testers except for above ground biomass (Table 6.4.2). From this result there were clear differences in the performance of the landraces as parents in hybrid combinations. Significant interactions were observed between the environment and GCA effects of lines for all the traits except stay-green and plant height. There existed further significant interactions between the environment and GCA of testers for days to 50% anthesis, days to 50% maturity, grain filling duration, stay-green and grain yield. The high GCA x environmental interaction associated with the lines suggests the need to use different sets of parents for hybrid development at different environments. The non-significant GCA x environment effect of testers for plant height, panicle length, above ground biomass and panicle weight suggests that stable performance of the testers across environments and can be used as parents in any of the environments.

Similarly, the SCA effects for line x tester and interaction effects with the environment were found to be highly significant for all the traits (Table 6.4.2). This suggests that the environment had a significant effect on the expression of these traits and the hybrids showed differential responses to the prevailing environmental conditions. Previous reports by Clarke *et al.* (1992) and Ejeta *et al.* (1997) pointed out the presence of differential expression of drought related traits on sorghum genotypes in relation to the environmental conditions.

6.4.3 Variance components

The variance component estimates due to SCA were larger than those due to GCA for all traits except days to 50% anthesis. In addition, the ratio of the mean square components associated with variance of GCA and SCA ranged between 0.06 and 1.03. The ratio was equal to unity for days to 50% anthesis and panicle length, whereas it was intermediate for days to 50% maturity (0.52) and stay-green scores (0.48). But the ratio was much lower than unity for grain filling duration, plant height, panicle weight, thousand-grain weight, above ground biomass and grain yield (Table 6.4.2). Information on the GCA effects of parents helps breeders to estimate the genetic potential of a breeding material for many desired traits. The differences in GCA among lines are mainly due to additive genetic effects and higher order additive interactions (Falconer and Mackay, 1996). The GCA effects among the female parents were larger than those of male parents except for plant height and biomass (Table 6.4.2). This may resulted from the fact that female parents were undergone through excessive breeding and selection for many desirable traits. This, in tern, may have positive and significant effect in accumulation of many additive genes.

These results suggest that the genetic variation observed among crosses on grain filling duration, plant height, panicle weight, thousand-grain weight, above ground biomass and grain yield was mainly due to non-additive gene effects. The variation observed among hybrids on days to 50% anthesis and panicle length was mainly due to additive gene effects. However, both additive and non-additive gene effects were prominent for the variation expressed on days to 50% maturity and stay-green scores among hybrids. Similar results were reported by Kenga et al. (2004) for grain yield in sorghum. Earlier studies have indicated that additive gene effects were relatively more important in sorghum for days to 50% anthesis, plant height, percentage dry matter, grain and biomass yield (Beil and Atkins, 1967; Kirby and Atkins, 1968; Haussmann et al., 1999). On the other hand, other researchers have reported that both additive and non-

additive gene effects were important in determining plant height, panicle length, grain yield and 100-grain weight in sorghum (Toure *et al.*, 1996; Degu *et al.*, 2009).

The fact that the present findings were not in agreement with the previous studies including Beil and Atkins (1967), Kirby and Atkins (1968) and Haussmann *et al.* (1999) on the predominant additive genetic effects may be due to the differences in the genetic background of the landraces used in the present study. Most authors mentioned above either use photoperiod insensitive conversion lines or improved inbred lines as male lines for their combining ability studies. The materials used as male lines for the present study were lowland sorghum landraces as that of Kenga *et al.* (2004). They were selected from prior genetic diversity assessment for their better performance under moisture stress conditions. They were sampled from a large sorghum landrace collection to represent a wide range of genetic diversity. The highly significant line x tester interaction observed for most of the traits probably attributed to the wide genetic diversity of the male parents. It was also pointed out by Rojas and Sprague (1952) that in maize SCA effects became more prominent than GCA effects if the parental lines used had been subjected to successive prior selection for the traits of interest under study.

Table 6.4.1 Combined analysis of variance for ten phonological, yield and yield component traits of sorghum measured across two environments.

Sources of variation	d.f. ^a	DTA	DTM	GFD	SG	PH	PL	BIOM	PW	GY	HSW
Rep (Env)	1	39.1	10.8	8.8	0.7	818.5	1.0	0.1	1575.9	0.6	0.3
Rep (Block)	26	80.6	78.6	29.1	0.7	6443.1	35.1	204.6	2842.5	4.0	0.7
Environment	1	369.6**	729.8**	60.7**	2.1**	1628.4*	80.1**	7301.4**	231916.1**	283.0**	37.5**
Genotype	167	219.5**	133.7**	104.2**	0.73**	7395.6**	74.0**	228.8**	5918.2**	6.7**	0.91**
Parent	35	190.7**	273.4**	56.1**	0.95**	4873.6**	101.8**	263.3**	1212.9**	1.95**	1.09**
Male	31	130.4**	246.3**	69.7**	1.04**	1084.5**	93.8**	220.6**	1277.3**	1.95**	1.2**
Female	3	283.4**	99.6**	48.7**	0.06	135.1	60.6**	19.3	236.4	5.0	0.26
Male vs Female	1	1950.8**	1716.0**	7.5	6.5**	139806**	389.9**	3660.3**	2403.3	0.001	5.77**
Checks	3	134.5**	61.9**	38.2**	0.06	8232.2**	17.4**	47.1*	170.4	0.54*	0.59*
Checks vs rest	1	259.7	1055.1**	267.9**	2.81*	26314**	188.4**	2377.8**	10480*	11.87*	0.0023
Hybrids	127	107.6**	52.3**	90.8**	0.59**	8236.7**	45.6**	206.9**	4874.0**	6.2**	0.71**
Parent vs hybrid	1	15831.9*	5026.6**	3016.9**	9.0**	5390.0	2933.0**	499.8*	288630**	248.7**	21.7**
Env x genotype	167	369.6**	23.8**	25.5**	0.63**	574.9**	6.3**	104.3**	1309.3**	1.33**	0.17*
Env x Parent	36	22.9**	18.2**	15.7**	1.03**	514.9	4.02	142.9**	1887.4**	2.09**	0.38**
Env x Male	32	21.2*	19.5**	15.4**	1.15**	577.8	3.4	155.2**	2070.0**	2.33**	0.35**
Env x Female	4	36.6*	8.3	18.2*	0.06	11.7	8.8	44.6	426.1	0.25	0.61
Env x male vs female	2	38.0	76.4	59.6	1.44	33.4	4.0	1127.6**	20387.1**	21.8**	4.43**
Env x Hybrid	127	50.4**	26.5**	28.7**	0.54**	596.8**	7.1**	112.4*	1472.7**	1.44**	0.18
Env x Parent vs hybrid	1	87.1	2.8**	58.6	1.27	226.0	2.6	69.9	656**	3.2	0.03
Env x Checks	4	76.38**	32.4*	24.9**	0.06	278.6*	1.26	77.2**	2869.6**	2.13**	0.25
Env x Checks vs rest	2	251.9*	387.0**	37.5	1.06	947	40.2	3670.8**	113980**	142.37**	18.59**
Error	308	15.77	11.08	4.48	0.32	326.7	3.89	74.35	666.2	0.59	0.14

^a d.f.= Degrees of freedom; DTA = Days to 50% anthesis; DTM = Days to 50% maturity; GFD = Grain filling duration; SG = Staygreen; PH = Plant height; PL= Panicle length; BIOM = Above ground biomass; PW = Panicle weight; GY = Grain yield; HSW = hundred-seed weight, *, ** significantly different at p = 0.05 and p = 0.01 probability levels, respectively.

The total parental GCA (lines and testers) accounted for most of the variability observed in the hybrids including on days to 50% anthesis (57%), panicle length (63%), above ground biomass yield (60%) and stay-green scores (59%) (Table 6.4.3). Testers had a relatively higher contribution on the total variability observed in the hybrids on days to 50% anthesis, days to maturity, grain filling duration, panicle length and panicle weight. Whereas the lines had significant contribution to GCA for plant height, biomass and grain yield. Assessing the genetic potential of selected local landraces for their hybrid performance was one of the main objectives of this study. Thus the estimation of general combining ability (GCA) of parental genotypes is vital and assisted in the identification of superior lines with favorable traits. High and negative GCA estimates for days to 50% anthesis, days to 50% maturity and grain filling duration indicates that parents were earlier or non-senescent than the general performance in the crosses. However, positive and high GCA estimates are desirable for the rest of the traits.

Plant height in the testers in general was shorter than the male lines, because the testers had undergone successive selections for shorter plant height to make commercial hybrids fitting to high input and mechanized systems. In addition, the testers grew faster and completed their life cycles before the onset of drought, as they were early maturing types with drought escape (Stout *et al.*, 1978). The estimates of GCA effects for the ten agronomic traits of the 36 parents are presented in Table 6.4.3. Non-significantly detectable variation was observed for GCA estimates for above ground biomass and panicle weight among males and panicle weight among females. Lack of significant GCA effects for panicle weight and biomass among male lines and biomass among female lines may have resulted from strong selection pressure that increases genetic similarity in a gene pool (Barbosa *et al.*, 2003). Traditionally farmers' select well-matured, large seeded, big and healthy panicles in the field to use as a seed for the next cropping season. Similarly the tester lines were developed to fit to certain commercial characteristics such as shorter plant height, big panicle and low fresh biomass yield.

Table 6.4.2 Variance component estimates from analysis of variance for ten agronomic traits in sorghum measured across two environments.

Sources of variation	d.f.	DTA	DTM	GFD	SG	PH	PL	BIOM	PW	GY	HSW
Env	1	367.6**	728.9**	61.2**	2.06**	1632.6**	80.11**	7298.69**	231915.6**	283.11**	37.51**
Line (GCA)	31	200.4*	194.8**	107.9**	1.34**	16002.9**	123.19**	587.61**	6290.7**	5.61**	1.18**
Tester (GCA)	3	3740.2*	1316.1**	664.3**	3.14**	5342.0**	997.09**	64.63	51182.0**	47.56**	3.73**
Line*Tester (SCA)	93	116.83**	83.4**	87.5**	0.52**	5346.3**	34.30**	148.93**	4545.1**	5.84**	0.76**
Env*Line	31	26.96**	15.9**	22.9**	0.42	480.2	4.22*	107.11**	1254.7**	1.22**	0.13**
Env*Tester	3	74.15**	28.2*	50.9**	0.99*	824.1	4.42	105.4	1190.9	1.67*	0.12
Env*Line*Tester	93	49.1**	26.0**	25.4**	0.68**	600.6**	6.95**	105.65**	1332.0**	1.38**	0.19*
Error	234	16.80	11.50	5.00	0.32	376.90	3.96	73.35	665.50	0.59	0.14
Variance component e	stimates										
δ ² _{GCA}		51.49	18.67	8.29	0.05	147.95	14.61	4.92	671.98	0.58	0.05
δ^2_{SCA}		50.01	35.95	41.25	0.10	2484.70	15.17	37.79	1939.80	2.63	0.31
$\delta^2_{GSA}/\delta^2_{SCA}$		1.03	0.52	0.20	0.48	0.06	0.96	0.13	0.35	0.22	0.15
Proportional contribut	ion to the	total varianc	es								
Line		8.92	20.14	4.83	42.11	34.89	26.89	58.37	7.56	0.87	12.86
Tester		48.36	27.86	17.07	16.82	0.03	36.40	1.40	25.24	19.72	11.36
Line*Tester		42.72	52.00	78.11	41.08	65.08	36.71	40.23	67.20	79.41	75.78
Mean		83.11	130.78	47.68	4.41	2.45	27.41	34.54	142.07	4.70	3.77
R-square (%)		89	88	93	69	91	91	73	87	90	82
CV (%)		4.90	2.56	4.65	12.74	8.36	7.23	24.72	18.31	16.33	10.11

^a d.f.= Degrees of freedom; DTA = Days to 50% anthesis; DTM = Days to 50% maturity; GFD = Grain filling duration; SG = Staygreen; PH = Plant height; PL = Panicle length; BIOM = Above ground biomass; PW = Panicle weight; GY = Grain yield; HSW = thousand-seed weight; CV = Coefficient of variation; R² = Coefficient of determination, *, ** significantly different at p = 0.05 and p = 0.01 probability levels, respectively.

The females ICSA 749 and ICSA 756 had high positive GCA estimates for grain yield. ICSA 749 showed high negative GCA effects for days to 50% maturity, grain filling duration and 100-seed weight and high positive GCA effects for plant height. On the other hand, ICSA 756 had high negative GCA estimates for days to 50% anthesis, plant height, panicle length and biomass and high positive GCA estimates for thousand-seed weight. From this result, although the two female parents had a comparable grain yield, they were different in that ICSA 749 is relatively tall and it took longer time to flower but it matured earlier and had a very short grain filling duration. However, ICSA 756 was significantly shorter and had low above ground biomass, it took a shorter time to reach 50% anthesis. The former shows drought escape due to its short growth duration (Pantuwan *et al.*, 2002; van Oosterom *et al.*, 2006) while the later was efficient in balancing assimilate translocation between the developing grain as expressed by its high 100-seed weight (Kouressy *et al.*, 2008). Therefore, parents ICSA 756 and ICSA 749 are good for dwarf stature, low biomass and early maturity in hybrid combination.

Among the male parents, 214838-A, 242039-B, 75454, 73056-B and 242050-A had high and positive GCA estimates for grain yield (Table 6.4.3). The line 214838-A had highly significant and positive GCA estimates for stay-green, panicle length and thousand-seed weight, whereas 73056-B had highly significant and negative for the traits mentioned. The line 242039-B showed positive and highly significant GCA effects for days to 50% anthesis, plant height and panicle length, while negative and highly significant GCA effects for grain filling duration were detected. The line 242050-A had highly significant and negative GCA effects for panicle length, and days to 50% maturity, but highly significant positive GCA estimated for thousand-seed weight. The high GCA estimates for grain yield observed on line 214838-A may be associated with its compact and medium panicles length with longer grain filling duration and high 100-seed weight. This line may be used to develop high yielding, late maturing and medium height compact panicle hybrids with long grain filling duration. However, the highly significant and positive GCA effects for grain yield observed on line 242039-B may be associated with short grain filling duration and long panicle length. This provides the opportunity to develop high yielding and tall hybrids with long panicle length and short grain filling duration. Lines 242050-A, 75454 and 73056-B may be useful in developing high yielding and early maturing hybrids with small to medium panicles and early expression of stay-green character (delayed senescence). In general, lines which had high general combining ability estimates for grain yield also showed high combing ability for one or two traits such as plant height, thousand-seed weight, panicle weight or early maturity.

Table 6.4.3 General combining ability (GCA) effects of 32 sorghum landraces and 4 CMS testers for ten agronomic traits across two environments

Parent	DTM ^a	DTA	GFD	SG	PH	PL	BIOM	PW	GY	HSW	GY per se
	(days)	(days)	(days)	(scores)	(cm)	(cm)	(t/ ha)	(g/plant)	(t /ha)	(g)	(t/ha)
Male											
71160-A	-1.8	1.97	3.77**	0.21**	11.41	0.4	-0.65	-20.2	-0.70**	0.04**	4.30
72472	-2.36	-1.53	0.83	-0.11**	-28.96	0.28	-7.02	-28.31	0.19	0.13**	3.61
72482	3.64	1.91	-1.73*	-0.36**	5.38	-2.93**	-1.39	-27.67	0.04	-0.21**	3.96
72572	3.45	0.16	-3.29**	0.08*	23.13	0.28	8.75	24.35	0.12	0.15**	2.90
72578-B	8.14**	3.03*	-5.11**	-0.11*	32.97	3.07**	3.1	16.52	0.00	0.23**	3.30
73056-B	-1.61	-1.84	-0.23	-0.36**	-22.34	-1.62**	-5.75	-0.67	0.67**	0.33**	3.93
73059	-1.48	-1.9	-0.42	0.14**	2.11	-0.39	-1.38	8.02	0.17	-0.19**	3.21
75454	3.14	2.53	-0.61	0.21**	9.91	-1.33**	2.38	26.52	0.72**	0.01	3.14
200538	-2.61	3.22*	5.83**	-0.04	9.79	-1.54**	1.39	-9.89	-0.11	-0.34**	4.64
200654	0.7	-0.47	-1.17	0.08**	14.94	4.17**	2.82	38.32	-0.04	-0.01	2.57
211239-B	-3.86	-0.65	3.21**	-0.11**	-13.84	2.57**	-6.82	-32.93	-0.67**	-0.43**	3.27
214855	1.33	0.03	-1.29*	0.02	-50.15*	-2.88**	-4.32	11.15	0.48**	-0.35**	4.71
242036	2.95	0.47	-2.48**	-0.42**	-55.9**	-0.14	-1.25	16.32	-0.26**	-0.16**	1.61
237260	-3.67	-1.15	2.52**	-0.11*	20.35	-0.22	-0.25	14.6	0.42**	0.01	3.67
239156	0.89	2.97*	2.08**	0.33**	22.1	-0.68	6.81	-20.6	-0.52**	-0.15**	3.28
239175	2.14	2.78*	0.64	0.14**	21.46	4.91**	3.81	18.36	0.01	0.00	2.74
239208	-3.36	-2.03	1.33*	-0.23**	1.66	0.38	4.1	2.65	0.01	0.01	3.34
242039-B	7.70**	0.97	-6.73**	0.02	55.41**	3.33**	6.55	8.71	0.74**	-0.09**	3.53
242047	-1.67	-0.72	0.96	0.02	21.85	0.9	0.14	-25.01	-0.92**	0.09**	2.35
244712	2.02	-0.78	-2.79**	-0.36**	-30.9	-3.50**	-4.25	-17.92	-0.29**	0.19**	3.27
244715	-2.55	-4.59**	-2.04**	-0.36**	13.6	-0.93	-1.41	5.34	-0.04	0.01	3.57
244727	-5.36**	-1.9	3.46**	-0.36**	-22.84	1.35**	-10.22	-43.74	-0.96**	-0.16**	3.96
244733	-1.8	-2.15	-0.36	0.02	20.35	-0.03	-1.88	-4.86	0.44**	-0.09**	3.94
244735-A	1.77	2.28	0.52	-0.11**	35.82	1.40**	5.74	16.52	-0.64**	-0.17**	3.27

214838-A	-2.67	1.72	4.39**	0.27**	28.97	1.13*	4.3	17.68	0.90**	0.12**	5.03
214838-B	-1.48	0.6	2.08**	0.52**	-18.53	-1.03*	4.83	6.92	0.39**	-0.16**	4.03
239167-B	1.45	0.03	-1.42*	0.14**	20.36	0.85	2.61	20.16	0.28**	0.15**	3.64
242049-B	-1.61	-1.4	0.21	0.14**	-49.53*	0.88	-3.28	1.46	-0.13	0.26**	4.09
242050-A	-3.92	-3.03*	0.89	-0.17**	-27.03	-1.53**	-1.05	6.04	0.55**	0.50**	3.12
244725-A	-1.48	1.22	2.71**	0.27**	3.76	-0.63	-3.33	-21.36	-0.55**	0.45**	4.27
244725-B	-1.17	-4.40**	-3.23**	0.58**	-48.59*	-4.72**	-6.15	-5.99	-0.16	-0.16**	3.58
69286-A	5.14*	2.66	-2.48**	0.02	-6.74	-1.82**	3.07	-0.5	-0.13	0	4.22
S.E.	0.99	0.83	0.53	0.14	4.52	0.49	2.16	6.45	0.19	0.09	
LSD (0.05)	1.45	1.20	0.79	0.20	6.86	0.70	3.03	9.12	0.27	0.13	
Female											
ICSA 101	0.59**	0.13	-0.47**	0.03*	2.49	0.20**	-0.77	-6.65	-1.02**	0.03**	4.07
ICSA 743	0.28*	0.31**	0.03	0.12**	2.06	1.13**	1.46*	1.95	0.04**	-0.06**	2.62
ICSA 749	0.05	-0.51**	-0.55**	-0.18**	2.6	0.57**	0.59	2.06	0.48**	-0.05**	5.07
ICSA 756	-0.92**	0.06	0.99**	0.03*	-7.16**	-1.91**	-1.27*	2.63	0.50**	0.08**	3.25
S.E.	0.35	0.29	0.19	0.05	1.60	0.17	0.76	2.28	0.07	0.03	
LSD (0.05)	0.50	0.42	0.26	0.07	2.26	0.25	1.08	3.23	0.10	0.05	

^a DTA = Days to 50% anthesis; DTM = Days to 50% maturity; GFD = Grain filling duration; SG = Stay-green; PH = Plant height; PL = Panicle length; BIOM = Above ground biomass; PW = Panicle weight; GY = Grain yield; HSW = thousand-seed weight *, ** significantly different at p = 0.05 and p = 0.01 probability levels, respectively.

Comparison of the actual mean values of parents for the grain yield revealed marked variation exists among them. The difference in mean grain yield among male parents was widely ranging from 1.61 to 5.03 t/ha with the average yield of 3.56 t/ha. Inconsistencies were observed between yield *per se* and GCA effects of few lines, i.e. lines that have lower grain yield *per se* were showed moderate to high GCA effects and the vice versa (Table 6.4.3). This may be resulted from the fact that different lines were crossed to testers with different cytoplasm sources. Additionally, environment x line x tester interaction effects were significant for all yield and yield components. This suggests that genotypes respond differently to environmental variables and some crosses combinations may have developmental plasticity to overcome the effects of low moisture stress.

Lines 244715, 244727, 244725-B, 214838-A, and 72578-B had highly significant and negative GCA estimates for either days to 50% anthesis or days to 50% maturity or grain filling duration. These lines are good combiners for early maturity but with low yield potential. On the other hand, lines 214855, 232036, 242049-B and 244725-B had highly significant negative GCA estimates for plant height and they offer an opportunity for developing short statured hybrids without significantly affecting grain yield. In general, the other lines improve one or two of the characters under study, but they had significantly low and negative GCA effects for grain yield. Thus they have low breeding value for hybrid production under drought conditions. From this particular study, it is also clear that most of the tall lines produced relatively higher yield in hybrid combination than shorter genotypes. This may be associated with high stem reserves with the tall genotypes (Blum et al., 1997) and the genotypes probably have better efficiency in remobilization of stem reserves to the grain as suggested by Borrell et al. (2000) and Blum (2011). In some of the lines, the significant positive GCA effect for grain yield may be associated with higher seed weight (Blum et al., 1997), with the presence of stay-green trait (van Oosterom et al., 1996) or early flowering (Blum, 1970) under the drought condition. Based on the present study, lines 214838-A, 242039-B, 75454, 73056-B and 242050-A and testers ICSA 749 and ICSA 756 were found to be the best combiners and would be useful for the future hybrid development programme for moisture stress agro-ecologies in Ethiopia or other similar environments.

6.4.4 Specific combining abilities

None of the cross combinations were found to have high and desirable SCA effects for all the combined traits under study, thus, grain yield was singled out and used as important selection

criterion for hybrid performance. Grain yield is the most important target trait in most breeding programmes (Blum, 2011). Superior cross combinations were selected based on both hybrid performance and SCA effects of hybrids. The estimates of SCA effects and mean grain yield performance *per se* of the top 30 crosses are presented in Table 6.4.4. The estimates of SCA effects provide important information about the non-additive gene effects which are the result of inter-allelic (dominance) and intra-allelic (epistasis) interactions (Falconer and Mackay, 1996). Among 128 crosses 59, 56, 55, 51, 50, 47, 33, and 30% of the hybrids had significant SCA effects for plant height, panicle weight, biomass, panicle length, grain yield, days to 50% anthesis, and days to 50% maturity, respectively in a desirable direction.

The cross combinations including ICSA 743 x 72482, ICSA 101 x 200654, ICSA 749 x 75454, ICSA 756 x 242036 were the best for lowest and significant SCA effects for short growth duration. Similarly, crosses ICSA 101 x 214855, ICSA 756 x 244725-B, ICSA 749 x 244725-B, ICSA 101 x 72472 and, ICSA 743 x 242036 showed high SCA effects for plant height and above ground biomass yield. These crosses also had good SCA effects for stay-green and medium to long growth cycle but low in their yield potential. Habyarimana *et al.* (2004) also reported that stay-green, tall stature, and medium to late maturity provide useful indirect selection criteria for improving biomass yield under moisture stress. They may be released as good hybrid varieties for areas where high biomass yield is required. In Ethiopia, the stalk is an important product and overall the value of the stalk accounts for about 40% of the value of grain produced (Wortmann *et al.*, 2006). It is used as firewood for cooking, fodder, and construction material. Crosses ICSA 749 x 242039-B, ICSA 756 x 242049-B, ICSA 756 x 75454, ICSA 756 x 73059 and ICSA 756 x 214855 exhibited the highest positive SCA effects and also provided the maximum grain yield across the two environments.

Among the selected top crosses with positive and highly significant SCA effects, 8 crosses resulted from high x high GCA combinations, 7 had high x low GCA, 3 low x high and 12 low x low GCA combinations for grain yield. For the other growth, phenological and yield component traits, crosses with high SCA effects involved either one or both of the parental lines with good, average or low GCA estimates. Parents with the best GCA estimates may not always produce the best hybrid combination due to non-additive gene interactions involved in the expression of the traits. In general, high x high, high x low, low x high and low x low general combiner parents produced good specific cross combinations for grain yield indicating that both additive, dominance and interaction (additive x additive, additive x dominance, dominance x dominance) gene effects were important for controlling the traits. The SCA effect *per se* has limited value in

assessment of parental line (Marilia *et al.*, 2001). Hybrid means coupled with favorable SCA and GCA effects should be used for extensive and meaningful selection of parental lines for the development of drought tolerant hybrids (Rosenow *et al.*, 1983; Henzell *et al.*, 1992). Thus, parents with high and favorable GCA effects may increase the concentration of desirable alleles; this in turn may increase the chance of identifying the best cross combinations (Kenga *et al.*, 2004).

Table 6.4.4 Specific combining ability effects of the top 30 selected crosses in sorghum for ten agronomic traits across two environments

Hybrids	DTA	DTM	GFD	SG	PH	PL	BIOM	PW	GY	HSW	GY pe
	(days)	(days)	(days)	(score)	(cm)	(cm)	(t/ha)	(g/plant)	(t/ha)	(g)	se (t/ha)
ICSA 101 x											
244725-A	-0.22	-2.06	-1.66	0.04	3.04	0.17	-3.00	14.55	1.62**	-0.13**	4.75
71160-A	5.84	3.87	-1.97	0.10	-27.24	2.12**	-3.01	0.46	1.32**	0.53**	4.30
244725-B	1.72	-3.75	-5.47**	-0.03	-61.74*	7.23**	-7.51	20.79	1.30**	-0.27**	4.82
244727	-0.09	0.38	0.34	0.16*	27.76	0.02	3.50	27.51	1.24**	0.05	3.96
200538	-3.84	-0.74	2.47*	-0.65**	-1.87	5.76**	3.21	16.41	1.07**	-0.27**	4.64
69286-A	-7.09	-3.94	3.03*	-0.21**	9.42	3.36**	3.17	19.63	0.98**	0.35**	4.53
ICSA 749 x											
242039-B	-4.86	0.87	6.37**	-0.01	3.40	-1.63	2.02	-40.07	3.32**	0.82**	9.23
200654	5.39	3.37	-1.45	-0.32**	2.37	-0.97	3.86	-45.89	1.49**	-0.15**	6.63
72572	-1.11	-1.19	0.43	0.18*	38.05	2.02*	-1.47	-46.53	1.29**	-0.53**	6.58
239175	1.95	2.44	0.49	0.37**	-25.02	-2.91**	-8.16	-28.85	1.22**	-0.08*	6.41
237260	-3.23	6.38*	10.12**	-0.38**	-22.04	-0.74	-4.06	-61.02*	0.97**	0.45**	6.56
72578-B	-1.05	-2.38	-0.76	0.12	17.58	-0.37	3.31	-16.58	0.87**	-0.07*	6.05
244733	-4.61	-2.00	3.24**	-0.01	-22.79	2.58**	-5.11	-12.52	0.74**	0.36**	6.36
242050-A	-1.98	-2.25	0.24	0.18*	-102.92**	3.58**	-0.04	25.38	0.73**	0.04	6.46
214838-A	-0.73	-2.81	-1.51	0.24**	51.33*	-0.04	5.16	35.36	0.64**	0.24**	6.71
ICSA 743 x											
237260	8.03	-3.94	-12.22**	0.07	1.50	-1.34	4.44	33.47	1.87**	0.09*	7.02
244735-A	-1.41	1.12	2.28	0.07	15.27	1.45	3.22	-44.09	1.16**	-0.08*	5.25
242047	8.53*	1.62	-7.16**	-0.06	0.50	5.09**	4.82	16.51	0.90**	-0.08*	4.72
214838-B	-0.16	-0.44	-0.28	-0.31	31.62	-0.25	-4.94	-13.02	0.84**	-0.78**	5.96
244727	-3.53	-0.50	2.84*	0.32**	34.19	-4.10**	-1.44	-36.19	0.74**	-0.33**	4.52
239208	-4.78	-1.13	3.47**	-0.06	1.44	-1.79	-1.03	-15.81	0.68**	-0.52**	5.43
244712	-5.16	2.76	7.09**	0.32**	47.50	0.20	7.74	8.63	0.68**	0.06	5.13
200538	-3.28	-3.31	-0.28	0.51**	-14.19	1.28	-4.99	1.91	0.63**	-0.31**	5.25
239167-B	-4.34	3.56	7.72**	0.32**	-14.01	-0.35	2.68	-12.80	0.60**	0.81**	5.62
ICSA 756 x											
242049-B	-5.33	-3.94	1.64	-0.10	44.10	-0.10	-0.53	50.37	2.48**	0.39**	7.54
75454	-3.83	-0.13	3.95**	0.34**	36.66	0.86	4.18	-3.19	2.41**	0.33**	8.32
73059	-3.45	0.13	3.01*	-0.10	5.71	-1.03	0.70	-9.15	2.31**	-0.03	7.67
214855	2.23	3.44	0.64	0.53**	-67.78	-1.34	-3.59	8.60	1.88**	0.02	7.56
242036	-8.39*	-8.06**	0.58	-0.28**	-69.78*	-0.53	-8.41	3.64	1.71**	-0.11**	6.64
239167-B	-1.89	-5.31	-3.99**	-0.35**	-12.54	4.78**	-11.18*	-36.29	0.80**	0.38**	6.27
S.E.	1.99	1.67	1.06	0.28	9.04	0.99	4.27	5.96	0.38	0.02	
SLSD (0.05)	2.90	2.36	1.58	0.40	13.73	1.40	6.04	<i>8.4</i> 3	0.54	-0.04	

^a DTA = Days to 50% anthesis; DTM = Days to 50% maturity; GFD = Grain filling duration; SG = Stay-green; PH = Plant height; PL= Panicle length; BIOM = Above ground biomass; PW = Panicle weight; GY = Grain yield; HSW = thousand-seed weight;*, ** significantly different at p = 0.05 and p = 0.01 probability levels, respectively.

6.4.5 Heterosis

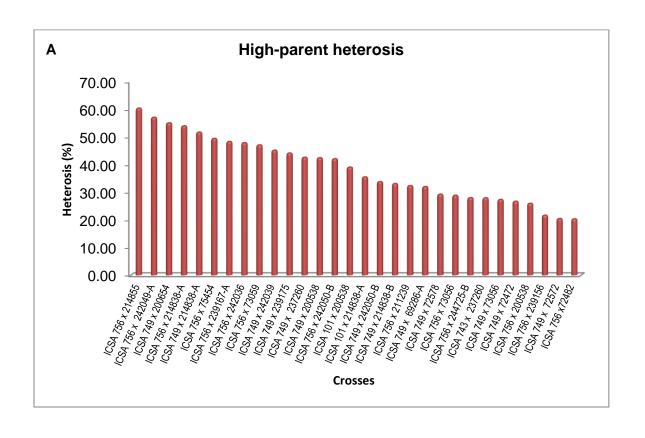
Comparison of the mean values for the hybrids and their respective parents under both stress and non-stress environments provides useful information on the relative superiority of the hybrids. The relative superiority of the hybrids over the mid-parents values is presented in Table 6.4.5. In the non-stress environment both the lines and hybrids showed a relaxed growth than the stress environment. Both days to 50% anthesis and days to 50% maturity at each environment showed similar patterns. However, the hybrids showed 10 and 6, and 2 and 4 days earlier to anthesis and maturity over their parents in the stress and non-stress environments, respectively. These two traits are the most important attributes that need to be considered in selecting genotypes for drought prone environments. Grain fill duration and panicle length were not affected, however, hybrids showed a fairly higher grain filling duration under stress condition. The stress environment highly hampered the performance of the lines for plant height, panicle weight, thousand-seed weight, above ground biomass and grain yield. However, the hybrids showed a relatively stable performance for the described traits. Thus, the hybrids significantly outperform the lines for plant height at 26%), panicle weight (63%) and above ground biomass (43%) under stress conditions. Hybrids displayed generally a significantly superior performance for panicle length, panicle weight, grain yield and thousand-seed weight across environments.

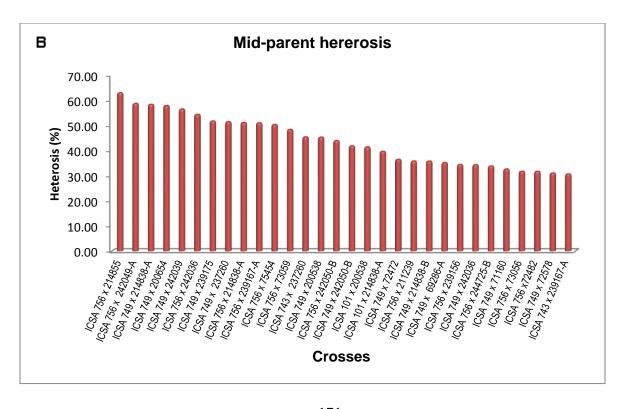
Evaluation of a large number of hybrid combinations in multi-location trials facilitates verification of hybrid yield potential and heterosis (Jordan *et al.*, 2003; Gabriel, 2005). For days to 50% anthesis, days to 50% maturity and grain filling duration negative heterosis is desirable but for other growth parameters, yield and yield components positive heterosis is a prerequisite for selection. Except for the cross ICSA 749 x 237260, all other crosses had negative heterosis for days to 50% anthesis. Negative high parent heterosis ranged from -0.56 to -40.6%,-0.4 to -18.4% and -1.1 to -23.5% for days to anthesis, maturity and grain filling duration, respectively. Additionally, none of the crosses selected based on high SCA showed heterosis for the above mentioned traits. This suggests additive gene action appeared to be more important than non-additive gene action in controlling the two traits. Earlier studies have also indicated that GCA is relatively more important for days to anthesis and maturity (Dequ *et al.*, 2009).

Table 6.4.5 Means of sorghum lines and hybrids, relative hybrid superiority (%) for ten agronomic traits measured at two environments

		Stress	5		Non st	ess	Combined		
Traits	Line	Hybrid	Hybrid superiority	Line	Hybrid	Hybrid superiority	Line	Hybrid	Hybrid superiority
DTA	88.8	79.7	-10.2	92.8	86.7	-6.6	92.8	80.7	-13.0
DTM	131.2	128.5	-2.1	136.4	130.8	-4.1	136.3	129.6	-4.9
GFD	42.4	48.7	14.9	44.6	49.1	10.1	43.9	48.8	11.2
SG	4.6	4.5	-2.2	4.0	4.4	10.0	4.12	4.5	9.2
PH	197.9	249.6	26.1	240	243.9	1.6	246.6	245.1	-0.6
PL	25.3	28.2	11.5	23.8	29	22.1	23.2	28.6	23.3
BIOM	23.2	33.3	43.3	37.1	38.4	3.5	34.2	35.6	4.1
PW	82.6	134.6	63.0	119.7	172.6	44.2	104.1	152.5	46.5
GY	3.2	4.4	35.7	4.1	5.7	39.0	3.6	5.0	38.9
HSW	2.9	3.6	23.7	3.7	4.5	21.6	3.5	4.0	14.3

^a DTA = Days to 50% anthesis; DTM = Days to 50% maturity; GFD = Grain filling duration; SG = Stay-green; PH = Plant height; PL = Panicle length; BIOM = Above ground biomass; PW = Panicle weight; GY = Grain yield; HSW = thousand-seed weight





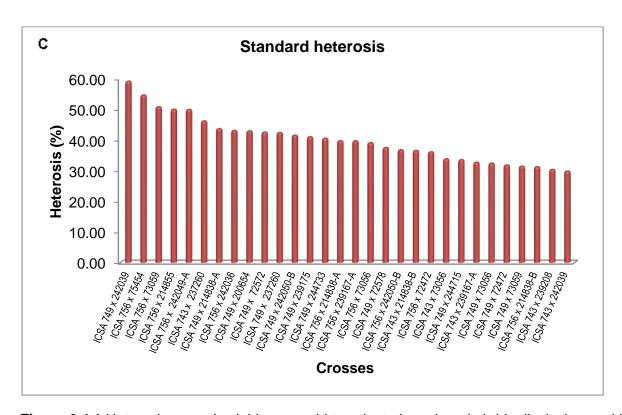


Figure 6.4.1 Heterosis on grain yield among thirty selected sorghum hybrids displaying positive high-parent heterosis (A), mid-parent heterosis (B), and standard heterosis (C).

Both negative and positive high-parent, mid-parent and standard heterosis was observed for all the traits studies. Negative heterosis was desirable for days to 50% anthesis, days to 50% maturity and grain filling duration, but positive heterosis was required for the other traits. General for all the hybrids, positive high parent heterosis ranged from 0.1 - 32.9%, 0.5 – 35.2%, 0.5 – 94.4%, 1.9 – 188.3%, 0.8 – 60.2% and 0.2 – 66.5% for plant height, panicle length, above ground biomass, panicle weight, thousand-seed weight and grain yield, respectively (data not shown). The maximum heterosis for grain yield was recorded from cross ICSA 756 x 214855 at 60%. Of the crosses 46, 33, 30, 27, 24, and 23% were selected on the basis of their high SCA effects and high heterosis for panicle length, panicle weight, plant height, thousand-seed weight, grain and above ground biomass, respectively. Quinby (1963) reported heterosis of 39 to 80% for grain yield and Blum *et al.* (1990) reported heterosis of 23.9% to 39.6% for grain yield. Blum et al. (1990) also observed significant heterosis for biomass, grain yield per plant, and grain number per panicle. Haussmann *et al.* (1999) reported a relative hybrid superiority of 47.1%. Overall, hybrids had higher grain yields and their earliness to flower and mature than parents may be explained by high productivity associated with hybrid vigour or heterosis.

Thirty hybrids displayed positive high, mid and standard heterosis of up to 60% grain yield (Figure 6.4.1). Eighty seven percent of the hybrids with positive high-parent heterosis involved ICSA 756 and ICSA 749 as female parents. In most of the breeding programmes, the A1 system is the most commonly used as compared to the A2, A3, A4 and A5 systems (Sleper and Poehlman, 2006). These CMS systems have distinct genetic features within and among species. Cytoplasmic male sterility is a maternally inherited trait and fertility restoration is conferred by nuclear-encoded fertility restorer gene (Rf) (Rooney and Smith, 2000; Sleper and Poehlman, 2006). In some cases additional modifier genes are required for full expression of the restorer gene. However, in this particular study, A2 (ICSA 749) and A4 (ICSA 756) systems were more stable and displayed the highly significant positive SCA effect and high parent-heterosis, suggesting their utmost value in future sorghum hybrid programme in this and similar environments.

The importance of landraces from semi-arid tropics as valuable genetic resources for drought tolerance is well documented (Blum and Sullivan, 1986; Reynold *et al.*, 2007). High genetic variability was observed among the male parents for the various traits studied. The high SCA estimates were greatly associated with high level of heterosis that may have resulted from the high genetic diversity available among the parental lines. Previous reports by Li and Li (1998) and Rattunde *et al.* (2001) indicated that high level of heterosis have been attained from diverse germpalsm of sorghum.

6.5 Conclusion

Results from the current study indicated that most of the hybrids obtained from crosses among selected lowland landraces showed better performances than their parents for most of the traits studied. Thus, it can be concluded that there is high potential for developing hybrids with superior grain yield and improved drought tolerance. Predominantly additive genes effects were responsible in controlling days to anthesis, maturity and panicle length, whereas non-additive effects were more important governing grain yield, thousand-seed weight, panicle weight, plant height and biomass. Male parents 214838-A, 242039-B, 75454, 73056-B, and 242050-A and female parents ICSA 749 and ICSA 756 were selected displaying positive and significant GCA effects for grain yield and yield related traits. Crosses including ICSA 756 x 214855, ICSA 756 x 242049-B, ICSA 749 x 200654 and ICSA 756 x 214838-A were identified as promising hybrids showing highly significant SCA effects and heterosis for grain yield and yield components. The lines are recommended as potential parents for inclusion in the future breeding programme.

Further stability analysis at representative growing environments and farmers-preferences of the selected experimental hybrids should be established before large scale production.

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CHAPTER 7

An Overview of the Research Findings

7.1 Introduction and objectives of the study

Sorghum is the dominant crop grown in the North Eastern Ethiopia with considerable genetic diversity for the most important traits. Crop productivity is the function of the genetic potential of the crop and of the environment in which the crop are growing. Thus, understanding the genetic diversity and its interaction with the prevailing environmental condition has paramount importance in developing improved sorghum cultivars for food security. In most breeding programmes landraces have been used as sources of genes for many desirable agromorphological traits. Likewise, yield and yield stability can be improved in sorghum through exploiting the available genetic diversity and genetic potential of landraces through well designed and systematic genetic characterization, and taking into consideration farmers-preferred traits. The objectives of the present study are highlighted and achieved from which inferences and suggestions can be drawn based on results obtained. The summary of the core findings and their implications for the development of high-yielding and drought tolerant sorghum varieties is presented in this chapter. This information may be utilized for future improvement and genetic conservation efforts of sorghum in the North Eastern Ethiopia or similar envioenmnts.

To summarize, the objectives of this study were:

- To evaluate sorghum production system and pattern, major production constraints and cropping mechanisms, varietal diversification, farmers' criteria for choosing varieties over time and space, technology generation and adoption of improved varieties;
- To assess the agro-morphological and molecular diversity and population structure of lowland sorghum landraces collected from different geographic origins using morphological and SSR markers;
- To assess the performance of sorghum landraces under moisture stress conditions and identify promising lines;
- To determine heterosis and combining ability of lowland landraces for grain yield, yield components and drought tolerance and to identify suitable parents for future sorghum hybrid cultivar development.

7.2 Research findings in brief

7.2.1 Analysis of the sorghum production systems in the north Eastern Ethiopia: breeding priorities and implications on varietal adoption

- The study demonstrated that the performance of sorghum is generally poor in the the north Eastern Ethiopia which is mainly accounted for by moisture stress, pests, diseases, weeds, farmland fragmentation due to demographic pressure, poor soil fertility, and poor performance of the local varieties.
- The study documented that in the low and mid-altitudes of North Eastern Ethiopia, enormous amount of genetic diversity of sorghum landraces existed as the result of decades of farmers' selection with regard to the environmental stress that growers have been facing over time. Over 70 landraces which are under production were identified varying in a number of morphological and socio-economic characters such as grain colour, head morphology, plant height, growth cycle, earliness, drought resistance, total plant biomass, market value and grain yield.
- Farmers identified indispensable selection criteria in sorghum that include: yield potential, early maturity, drought tolerance, good baking and brewing quality, resistance to insects, high biomass yield and stalk palatability. On top of all, early maturity and drought tolerance were considered as the most important selection criteria as they allowed the crop to escape drought stress and ensure good harvest under the prevailing moisture stress conditions.
- The study found that about forty improved sorghum varieties have been released to drought prone areas of Ethiopia. The varieties were mainly released for their early maturity, drought resistance, resistance to *Striga hermonthica* and stalk borer, and relatively high yield potential. Despite the greater availability of improved varieties with considerable yield potential (2 6 t/ha), they have not yet been adequately adopted by smallholder farmers. Farmers were reluctant to use improved varieties because of their short stature and reduced biomass, making them less valuable for livestock feed. Eary maturity and white seed color of these varieties make them liable to bird attack.

7.2.2 Genetic diversity assessment of lowland sorghum [Sorghum bicolor (L.) Moench] landraces of Ethiopia using qualitative traits

 A wide range of variability was observed among the landraces for qualitative traits. High Shannon-weaver phenotypic diversity indices ranging from 0.98 to 1.00 and an average

- diversity index of 0.91 was observed for most of the characters studied. Additionally, the accessions showed very high polymorphism for leaf rolling and stay-green which are important traits for selection under drought stress conditions.
- The proportion of total diversity obtained among the collection districts was less than that of within collection regions, maybe because all the accessions of each district were collected from the lowland agro-ecology. It would be possible to make selection for any of the traits within a particular region, presuming that a significant portion of the phenotypic variation is genetic.
- Stay-green, leaf rolling, awns, head shape, panicle exsertion, inflorescence compactness
 and leaf orientation were important traits in differentiating accessions of different regions.
 This implies that these traits are vital for the variation in sorghum landraces collected from
 different regions.

7.2.3 Genetic diversity assessment of lowland sorghum [Sorghum bicolor (L.) Moench] landraces under moisture stress environments using quantitative traits

- Landraces exhibited highly significant variations for ten quantitative characters assessed in this study. The extent of variation was highly influenced by location and moisture stresses.
- Landraces designated as 214838-A, 72476, 73090, 244733, 239150, 200113, 214855, 239255-B, and 69251 were identified as suitable novel genotypes with high grain yield and head weight, increased panicle length, medium maturity and long stalk.
- Unique landraces such as 244745, 244732-A, 72439-C, 69251, and 72493 were selected for short stalk, reduced biomass yield and relatively high grain yield.
- Landraces 239167-B, 244725-a, 242049-B, 242050-A, 244711-A, 202508, 244729-A, 242046, 75454, 73056-B and 72457 were recognized as early maturing with relatively medium grain yield and intermediate stalk.
- High levels of trait repeatability values were recorded for days to 50% anthesis at 77%, panicle length (65%) and plant height (71%). Repeatability estimates of traits were influenced by the environment and genotype by environment interaction.

7.2.4 Molecular genetic variation in lowland sorghum [Sorghum bicolor (L.) Moench] landraces of Ethiopia assessed by simple sequence repeats (SSRs)

- The morphological variability examined above was complemented by high molecular markers diversity. A total of 316 putative alleles were recorded with an average of 10.5 alleles per locus and the PIC values ranged from 0.26 to 0.88 with a mean value of 0.61.
- The total gene diversity was 0.69, which partitioned 86% among and 14% within landraces. The first was largely due to variation among genotypes within geographic origins. No genetic variation was observed between geographic origins.
- Landraces from different collection sites appeared to cluster together indicating the
 existence of high level of gene flow among regions. Gene flow could be attributed to the
 local seed system, where framers commonly exchange planting material.
- The model-based analysis clustered the landraces into two distinct sub-groups with relatively high genetic differentiation.

2.2.5 Combining ability and heterosis in lowland sorghum [Sorghum bicolor (L.) Moench] landraces under moisture stress condition

- The study found non-additive gene action to be predominant in controlling plant height, grain yield, above ground biomass, grain filling duration, thousand-seed weight and panicle weight. However, predominance of additive gene action was found for days to anthesis and panicle length.
- The study identified candidate landraces 214838-A, 242039-B, 75454, 73056-B, and 242050-A and female CMS parents ICSA 749 and ICSA 756 displaying positive and significant GCA effects for grain yield and yield components.
- Candidate sorghum hybrids: ICSA 749 x 242039-B, ICSA 756 x 242049-B, ICSA 756 x 75454, ICSA 756 x 73059 and ICSA 756 x 214855 were idetified exhibiting the highest positive SCA effects and maximum grain yield across the two environments.
- In this study two CMS systems: A₂ (ICSA 749) and A₄ (ICSA 756) were found to be more stable and displayed highly significant positive SCA effects and high parent-heterosis for grain yield.

7.3 Implications of the research findings for breeding sorghum to moisture stress tolerance

- The importance of addressing the missing link in the research-extension-farmer linkages for better diffusion and impact of improved technologies cannot be under-estimated. To benefit the most from the available improved technologies, farmers have to be part of the breeding process right from the very beginning.
- Large genetic diversity examined for grain yield potential and drought tolerance in the lowland sorghum landraces collections of North Eastern Ethiopia implied that there is huge potential for selection of source germplasm for breeding for moisture stress tolerance/resistance.
- The low level of genetic differentiation observed and the high level of gene flow between the different geographic origins indicates that a relatively small random collection of landraces would capture the bulk of genetic diversity in the target area. A random sample may result in the loss of some genes. It is advisable to determin a core collection in order to capture the entirety of the variation.
- Two distinct sub-groups observed from model-based analysis, would enable breeders to select distinctive alleles and exploit the potential of transgressive segregation between the two sub-groups.
- Sorghum being cultivated in marginal and risk-prone production systems, it would be
 appealing to associate genetic diversity with drought tolerance and adaptation potential in
 future research activities.
- The importance of both additive and non-additive effects in controlling grain yield and its components and drought tolerance suggested that that breeding gain can be realized through hybridization and selection.
- Therefore, selection of parents with high GCA and hybridizing them and further stability analysis at representative growing environments and farmers-preferences of the selected experimental hybrids should be established before large scale production.