

# **Breeding Tef [*Eragrostis tef* (Zucc.)Trotter] for Drought Tolerance in Northern Ethiopia**

By

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## Thesis Abstract

Tef is the leading crop in Ethiopia for human food and animal feed. Worldwide tef is becoming a popular health food due to its gluten-free property. Productivity of tef is low in Ethiopia, with a mean of  $1.6 \text{ t ha}^{-1}$ , which is well below potentially attainable yields of over  $4 \text{ t ha}^{-1}$ . In the past the national tef improvement program has developed improved tef varieties. However, adoption of these varieties in the moisture-stressed tef growing areas has been limited because of their late maturity and the frequent occurrence of terminal drought. Development of tef varieties with high yield potential and adaptation to moisture stress is the overriding consideration in Ethiopia. Therefore, the overall goal of the study was to contribute to food security in Ethiopia, through improving the yield and productivity of tef. To achieve this, a research project was conducted aiming to develop drought tolerant tef varieties with farmer preferred traits, wide adaptation and better performance under moisture stress. The specific objectives of the study were: (1) to assess tef production constraints, management practices, input use, farmers' trait preferences and breeding priorities in northern Ethiopia, Tigray Region; (2) to determine the genetic diversity of tef genotypes for drought tolerance using phenotypic and molecular markers, and to identify promising tef genotypes for improving moisture stress tolerance and grain yield; (3) to estimate the level of genetic variation, inheritance and trait association of tef genotypes; and (4) to determine gene action and inheritance of drought tolerance in tef and to identify better performing populations in moisture-stressed and non-stressed environments, and to advance these through the single seed descent method. Different research activities were conducted that contributed to these objectives.

Participatory rural appraisal research was conducted in northern Ethiopia in four selected districts of three administrative Zones of the Tigray Region, namely, Laelay-Maichew, Medebay-Zana, Ahferom and Alamata. Primary data was collected from a total of 240 sampled farmers and secondary data was collected from the Bureau of Agriculture and Rural Development (BoARD). Data was coded and analysed using SPSS software. Results indicated that improved varieties were used by the majority of respondents in all districts and fertilizers in all except Alamata, where severe moisture stress dictated the choice of variety and reduced fertilizer application. High grain yield, increased panicle length and straw yield were the most important farmer-preferred traits. In Alamata District, early maturity was the third preferred trait while straw yield ranked fourth. Transplanting maximized the yield of tef, but a cost-benefit analysis showed that direct row sowing was more profitable. Moisture stress (reported by 60.4% respondents, weeds (61.2%), shoot fly (58.2%), lodging (53.3%) and seed shattering (60.8%) were rated as

severe tef production constraints in the study areas. Farmers estimated that moisture stress caused yield losses of 37%, 49%, 48% and 60%, in the Laelay-Maichew, Medebay-Zana, Ahferom and Alamata districts, respectively. This study prioritized production constraints and farmers'-preferred traits useful in the breeding of tef to enhance its productivity in drought-prone environments in northern Ethiopia.

One hundred forty four tef genotypes were evaluated using four experiments representing optimum moisture and moisture-stressed environment in the 2014 main and off seasons. Genotypes, DZ-Cr-387, DZ-01-787, DZ-01-3186, 9432, 9403, 9415, 205917, 205896, 215678, 213237, Jano, Kaye-Agachew, Purpurea, Kaye-Murri and Dschanger were selected as promising parents with superior grain yields, reduced lodging, tall plant height and long panicles in a non-stressed environment. Conversely, genotypes, DZ-Cr-385, DZ-Cr-37, HO-Cr-136, DZ-01-2053, Dabbi, 207832, Zagure and Shawa-Gemerra were selected as superior parents for their early maturity and high yield performances under moisture-stressed conditions. Main shoot panicle seed weight had high genotypic coefficients of variation (GCV) of 22.4% and 25.9% under non-stressed and stressed conditions, respectively. Grain yield had GCV values of 17.6% and 20.0% in the corresponding environments. Heritability was higher under optimum conditions than moisture stressed conditions. Days-to-maturity, plant height, panicle length and panicle seed weight were positively correlated with grain yields under non-stressed conditions, while these traits negatively correlated with grain yield under moisture stressed conditions. A path-coefficient analysis indicated that direct selection for high biomass, harvest index and late maturity could increase grain yield under optimal condition, while under moisture stressed conditions early maturity, high biomass and harvest index were important direct selection criteria for tef breeding aiming for drought tolerance.

The genetic diversity of 60 selected tef genotypes was determined using 10 selected diagnostic and polymorphic simple sequence repeat (SSR) DNA markers. The number of alleles per locus varied from 10 to 23 with a mean of 16. The polymorphic information content (PIC) ranged from 0.64 for marker CNLTS11 to 0.94 (CNLTS136A/B) with a mean of 0.84, suggesting sufficient discrimination power of the markers for the tested genotypes. The analysis of molecular variance showed that 63% and 35% of the total variability could be attributed to differences within and among tef genotypes, respectively. Overall, the SSR analysis identified distinct genotypes such as DZ-Cr-385, 222076 and 213237, which are known for their early maturity and good yields under moisture stress. The analysis also identified genotypes DZ-Cr-387, 205896, 205917 and Dschanger, which was consistent with their unique agronomic attributes

such as late maturity, high grain yields, relatively good plant heights and long panicles under optimum rainfall conditions. The identified agronomic complementary tef genotypes are valuable genetic resources for further breeding.

Seventeen crosses along with their eight parents were evaluated in the F<sub>2</sub> generation under moisture-stressed and non-stressed conditions in northern Ethiopia during 2015 and 2016. Additive gene action predominantly controlled the inheritance of grain yield, biomass yield, harvest index, days-to-panicle emergence, days-to-maturity, panicle length, plant height, main shoot panicle seed weight, number of productive tillers per plant and relative water content under both the moisture-stressed and non-stressed conditions. Non-significant general combining ability (GCA) and specific combining ability (SCA) effects were detected for biomass yield, relative water content and peduncle length under non-stressed conditions. Under both test conditions genotypes, DZ-Cr-387 and 9415 were the best general combiners for increased grain yield and yield components, except for days-to-maturity. Conversely, genotype 222076 was the best general combiner for reduced maturity period only. The families of DZ-Cr-387 x 207832 and DZ-Cr-387 x 222076 were high grain yielders with early maturity under both test conditions. There were marked genotypic and phenotypic variations, among the crosses in the F<sub>2</sub> generation, allowing for successful selection and genetic advancement. Overall, the study identified valuable tef genotypes with high combining ability for yield and yield components in drought-stressed and non-stressed environments. It also initiated the development of novel tef families that will be further evaluated for cultivar development and release.

## Declaration

I, Mizan Tesfay Abraha, 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.
4. This thesis does not contain other persons' writing, unless specifically acknowledged as being sourced from other researchers. Where other written sources have been quoted, then:
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Signed

.....  
Mizan Tesfay Abraha

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

.....  
Prof. Hussien Shimelis (Supervisor)

.....  
Prof. Mark Laing (Co-Supervisor)

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## **Dedication**

This thesis is dedicated to my family.

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## Publications Pertaining to This Thesis

### Chapter two

Abraha, M.T., H. Shimelis, M. Laing, and K. Assefa. 2016. Diagnostic assessment of the production status, constraints and farmer-preferred traits of tef [*Eragrostis Tef* (Zucc.) Trotter] under drought-prone environments. *Transylvanian Review* 24:926-939.

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### Chapter three

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### Chapter five

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### Chapter six

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# Thesis Introduction

## Background

Agriculture contributes to 42% of the gross domestic product (GDP) and employs more than 85% of the population in Ethiopia (CSA, 2015). The majority of farmers in Ethiopia are smallholders and practice a mixed crop-livestock farming system. Input use (e.g. improved seeds and fertilizers) is low leading to poor crop productivity (Yu and Nin-Pratt, 2014). Cereal crops predominate grain production with 80% area coverage and 87% of the total harvest (CSA, 2016). Of the main cereal crops grown in the country, tef [*Eragrostis tef* (Zucc.) Trotter] takes first rank with a production area of 2,866,052.99 ha with mean productivity of 1.6 t ha<sup>-1</sup> (CSA, 2016). Tef is grown for food, feed and as a source of cash income. It is mainly used for making the popular pancake-like bread called 'injera' (Ketema, 1997). Tef contains 11% protein, 80% complex carbohydrates and 3% fat (Piccinin, 2002). Grain of tef is known for its high mineral contents than other cereals such as maize, wheat, sorghum and millets. 100 gram of tef grain contains 180, 0.8, 7.6, 184, 9.2, 429, 3.6, 427, 12 and 4.4 mg of calcium, copper, iron, magnesium, manganese, phosphorus, zinc, potassium, sodium and selenium, respectively (Bultosa and Umata, 2013). In terms of vitamins, tef grain contains 0.39, 0.27 and 3.4 mg of Thiamin, Riboflavin and Niacin, respectively, which is higher than reported in other cereals (Bultosa and Umata, 2013). Because of its nutritional balance, tef is increasingly becoming popular in Europe and North America as a health food for persons of gluten intolerance (Spaenij-Dekking et al., 2005; Hopman et al., 2008; Saturni et al., 2010). Tef seeds can store for a long time, even under local storage conditions. Furthermore, diseases and pests are not a serious problem in the major tef-growing areas of Ethiopia (Ketema, 1997).

The major tef producing regions in Ethiopia, include Oromia with 48% production, Amhara (38%), Southern Nations Nationalities and Peoples (SNNP) (7%), Tigray (6%), and Benishangul-Gumuz (1%). Of the major cereals produced in each region, tef shares an area of 22, 32, 29, 17 and 25% with productivity of 1.2, 1.6, 1.6, 1.2, 1.3 t ha<sup>-1</sup>, in that order (CSA, 2016). The human population of Ethiopia is estimated at 99 million (World Bank, 2016). Due to the ever increasing population, productivity of tef and other food security cereal crops needs to be enhanced to meet food demands.

Tef grows under a wide range of ecological conditions that can range from sea level up to 2800 m above sea level (m.a.s.l). The crop performs best from 1800 to 2100 (m.a.s.l). The annual

rainfall requirement of the crop ranges from 950 to 1500 mm (FAO, 2011). Several authors (Ketema, 1997; Tefera and Ketema, 2001; Assefa et al., 2011) have noted the wide adaptation of tef and its remarkable drought tolerance. It grows relatively well under low rainfall condition, unlike other cereals that fail to provide yield. Moderate yields can be harvested even under low rainfall conditions of 450 to 550 mm (Ketema, 1997). Furthermore, tef can grow fairly well under water lodging conditions and it is suitable for double or relay cropping or intercropping with sorghum and safflower. Tef requires temperatures of 10 to 27°C. The crop is very sensitive to day length and performs best with a 12 hours day length (Ketema, 1997).

### **Constraints to tef production in Ethiopia**

The mean tef yield in Ethiopia is estimated at 1.6 t ha<sup>-1</sup> (CSA, 2016), while attainable yield potential is 6 t ha<sup>-1</sup> (Ketema, 1993). The current low yield levels can be attributed to different production constraints such as susceptibility to lodging, moisture stress, water logging, weeds, seed shattering and poor pre- and post-harvest agronomic management practices. The crop requires a well-ploughed field for successful establishment and to suppress weed competition (Yadeta et al., 2001). Ploughing four times has been recommended for improved tef yields (Tsefahunegn, 2014). It has been argued that more efficient agronomic management could double the yield of crop plants (Mueller et al., 2012). However, this is challenging in tef, given that the crop is primarily grown by subsistence farmers using low input agronomic systems (Altieri, 2009; Yu and Nin-pratt, 2014). Currently, the majority of the farmers practise broadcast sowing, which is associated with a high incidence of lodging, reduced plant growth and yield (Asargew et al., 2014; Abebe and Workayehu, 2015). The small seed size of tef makes it difficult to control population density and accurate seed distribution during sowing, which also has an impact on nutrient use efficiency of the crop and increases susceptibility to lodging. At harvesting and threshing time, there is much yield loss because of the unavailability of post-harvest machinery suitable for small-scale farmers. Farmers mow the crop using hand-held sickles and thresh the crop using oxen, and winnow the seed manually to separate the seeds from the chaff. During winnowing much seed is lost with the chaff because of the small size and light weight of the seed (Ketema, 1997).

Tef has a weak stem and a shallow root system, leading to lodging (Ayele et al., 2001; Van Delden et al., 2010), which is one of the main production problems, causing yield losses of 11 to 25% (Ketema, 1993). Lodging severely affects grain yield and straw quality (Assefa et al., 2011). Lodging of tef is also a main constraint limiting fertilizer application. Use of nitrogenous

fertilizers enhances lodging (Assefa et al., 2014; Tesfay and Gebresamuel, 2016). Presently row sowing of tef seeds is being introduced to farmers in order to reduce plant populations and hence reduce lodging. However, drilling of the small sized tef seed is difficult, tedious and expensive, as it requires much human labour, and there are no small-scale matching machines available for row sowing.

Numerous grass and broad-leaved weed species affect tef productivity, especially during early crop establishment (Assefa, et al., 2008; Tesfahunegn, 2014). About 35% yield loss has been reported to be caused by weed competition (Addisu, 2016). The weeds also harbour insect pests and make harvesting operations difficult (Zewde and Damte, 2013). Repeated hand weeding is necessary where both grass and broad-leaved weeds are present, making tef production expensive.

Tef is less sensitive to disease and insect pests than other cereals cultivated in Ethiopia (Ketema, 1997; Assefa et al., 2011). The diseases that affect tef production includes tef rust (*Uromyces eragrostidis*), head smudge (*Helminthosporium miyakei*), damping-off (*Drechslera spp*) and helminthosporium leaf spot (*Helminthosporium spp*). Tef rust and head smut are relatively important (Badebo, 2013). Insect pests affecting tef productivity include grasshopper (*Aiolopus longicornis*), shoot fly (different species), red worm (*Mentaxya ignicollis*), Wello bush cricket (*Decticoidea brevipennis Rag.*), termites (*Macrotermes subhyalinus* and *Odontotermes sp.*) and black beetle (*Eelangerius niger*) (Damte, 2013). Tef shoot fly is an insect pest that is more important in the Tigray Region (where there is limited moisture) than other major tef growing regions of Ethiopia. Shoot fly causes 16-20% yield losses in Tigray Region (Damte, 2013). To date there is limited genetic variability reported for shoot fly resistance. Scheduled sprays using the chemical, 'Karate' (cyhalothrin), reduce its impact (AxARC, 2011).

Tef is a relatively drought tolerant crop compared to other cereal crops. However, about 25.5 to 51% grain yield reductions have been reported for tef due to moisture stress (Admas and Belay, 2011; Shiferaw et al., 2012). Farming systems studies by Yizengaw and Verheye (1994) in the Adda District found that poor rainfall distribution was one of the major tef production constraints. Yihun et al. (2013) also reported 1 t ha<sup>-1</sup> grain yield reduction due to 25% soil moisture deficit at the mid-growth stage of the crop. This indicates that mild moisture stress could lead to a substantial yield loss in tef. In the semi-arid tef growing areas of Ethiopia, such as the northern Tigray, a mild to severe moisture stress is a common phenomena during the grain filling stage (i.e. during September) of tef and other crops, which leads to high yield losses. Considering the

largest area coverage by tef, such a yield loss, would have a high implication in supporting the food self sufficiency program in the country. Moisture stress also limits fertilizer application for tef (Molla and Muhie, 2011) dictating application of high inputs for increased output strategy of tef. Therefore, breeding for moisture stress-tolerance could be a good option to improve food security in the face of frequent droughts in Ethiopia.

### **Rationale underlying breeding tef for tolerance of drought stress**

Moisture stress is one of the main factors limiting crop yields in areas with low and erratic rainfall. Due to climate change the negative effect of moisture stress on crop yields is increasing. Most resource constrained farmers in Ethiopia face food insecurity due to recurrent drought increased by climate change (Endalew et al., 2015). Agricultural activities are influenced by a wide range of environmental factors related to altitude, soil conditions, rainfall duration and temperature. The semi-arid areas of Ethiopia such as the Tigray region receives limited and erratic rainfall ranging from 400 to 700 mm year<sup>-1</sup> (Gebrehiwot et al., 2011; Teklu, 2014). Because of the rainfall variations from year to year farmers use various crops, and plant varieties with varied maturity periods as a coping strategy. Due to its flexible environmental adaptation, tef grows in high rainfall and drought-prone areas (Assefa et al., 2011; Assefa et al., 2015). In such areas, tef serves as a food security crop enabling farmers to have good harvest in seasons with good rainfall and to attain a moderate yield during drought spells, without complete total crop failure, as experienced with other crops such as maize and sorghum (Reda, 2015).

Developing moisture stress tolerant varieties with high yield potential is one of the main goals of the national tef breeding program (Assefa et al., 2011). A tef variety locally referred to as Tseday (DZ-Cr-37), released in 1983, is the most widely grown improved variety in the moisture stressed areas. Other improved varieties such as DZ-Cr-387 (RIL-127) (Gemechis), DZ-Cr-385 (RIL 295) (Simada) and Boset (DZ-Cr-409, RIL-50d) were released in 2007, 2009 and 2012 (MoA, 2014), in that order, for moisture-stressed areas. However their adoption is limited, because of their poor grain yield performance, despite their early maturity. Recently released tef varieties such as Quncho (DZ-Cr-387) released in 2006 and Kora [DZ-Cr-438 (RIL No. 133B)] released in 2014 (MoA, 2014) are suitable for areas with optimum moisture but do not perform well in moisture limited and variable rainfall conditions. This indicates the need for the continuous development of improved varieties with high grain yield and early maturity, which could sustain grain yield both in the moisture stressed and non-stressed areas and across

seasons. Ethiopia is believed to be the center of genetic diversity of tef (Vavilov, 1951). Local tef germplasm grown by farmers are known for their good performance in harsh environments affected by moisture stress and low soil fertility, which makes them suitable as a source of genes for moisture stress tolerance breeding. Therefore, proper evaluation of the landraces and improved tef varieties in the targeted environments could be helpful to exploit the source of genes for moisture stress tolerance in the landraces and high grain yield in the improved varieties, aiming to breed high yielding varieties with better adaptation to drought-affected environments.

Farmers are the main actors in agriculture in that they have a wealth of knowledge about their crops, farming systems and constraints, and have their own skills to manage crops in their environment (Altieri and Koohafkan, 2008). Before the start of any crop improvement activities, detailed knowledge of the farming system, and the variety and trait preference of farmers of the target environment is essential. This can be achieved through collection of primary data from farmers which will have a great role in the breeding of a successful variety or development of novel agricultural technology and thereby in their ultimate adoption by farmers. There is little or no up-to-date information on the production potential, constraints, input adoptions and farmer-preferred traits of tef under marginal and moisture-stressed growing environments in Ethiopia. Identification of the farmers' trait and variety preferences in these areas is crucial to devise breeding strategies. This will allow for the breeding of tef varieties both for optimum rainfall and for drought- prone environments.

### **Research objectives**

The overall goal of the study was to contribute to food security strategy in Ethiopia, through improving yields and productivity of tef. To achieve this, a research project was conducted aiming to develop drought tolerant tef varieties with wide adaptation and better performance in moisture stressed areas.

The specific objectives of the study were:

1. To assess tef production constraints, management practices, input use, farmer's trait preferences and breeding priorities in northern Ethiopia, Tigray Region.
2. To determine the genetic diversity of tef genotypes for drought tolerance using phenotypic and molecular markers and identify promising tef genotypes useful for improving moisture stress tolerance and grain yield.

3. To estimate the level of genetic variation, inheritance and trait association of tef genotypes.
4. To determine gene action and inheritance of drought tolerance in tef, and to identify better performing families for moisture-stressed and non-stressed environments, and then advance these through the single seed descent method.

### **Research hypotheses**

The current study was based on the following test hypotheses:

1. Farmer's perceptions and their indigenous knowledge on trait preference for variety selection in moisture stressed areas have great implication for breeding moisture stress tolerant tef varieties with better yield performances.
2. There is valuable genetic diversity in landraces and released tef varieties for breeding for moisture stress tolerance and high grain yield.
3. Additive gene action is predominant in the control of inheritance of drought tolerance in tef.
4. The single seed descent method of selection is an effective method to advance the selection of drought tolerant genotypes in tef.

### **Thesis outline**

The thesis consists of six chapters, which are outlined below. The referencing system used in this thesis is based on the referencing style of the Journal of Crop Science. The thesis is in the form of discrete research chapters, each following the format of a stand-alone research paper (whether or not the chapter has already been published). This is the dominant thesis format adopted by the University of KwaZulu-Natal. As such, there is some unavoidable repetition of references and some introductory information between chapters. The research outcomes covered in Chapter Two have been published in the Journal of Transylvanian Review (Volume XXIV, No. 7, Special Issue, 2016) and in the International Journal of Agricultural Sustainability (DOI:10.1080/14735903.2016.1173990). Chapter two have provided information on the current production and productivity status, farmers' agronomic management practices, inputs use, farmers trait and variety preferences, cultivated landraces and production constraints of tef in the semi-arid tef growing areas of northern Ethiopia. Chapter Three have been published in the Journal of Crop Science (DOI: 10.2135/cropsci2015.07.0449). This chapter provided information on the genetic diversity and yield and yield component performance of 144 tef genotypes under

moisture stressed and non-stressed conditions, from which parents were selected for high grain yield and moisture stress tolerance breeding. Chapter Four has been accepted in the Australian Journal of Crop Science. In this chapter, traits with higher variability, heritability and highly correlated with grain yield under moisture stressed and non-stressed condition were indentified, which would be useful to increase response to selection. Chapter Five has been published in the South African Journal of Botany (DOI:10.1016/j.sajb.2015.12.009). Out of the 144 tef genotypes, 60 genotypes selected based on their morphological diversity were subjected for DNA level genetic diversity and this enabled to select diverse tef genotypes to be used in breeding tef varieties with high grain yield for the moisture stressed areas. Chapter six has been submitted to the Crop Science Journal. This chapter has provided information on the gene action of yield and yield components of tef under moisture stressed and non-stressed conditions and crosses were selected with high genetic variation for targeted breeding of tef varieties with high grain yield and yield component performance under moisture stressed and non-stressed conditions.

The structure of the thesis is outlined below:

Chapter	Title
-	Thesis introduction
1	Review of literature
2	Assessment of production status, constraints, management practices and farmer-preferred traits of tef [ <i>Eragrostis tef</i> (Zucc.) Trotter] under drought-prone environments: Implications for future research directions
3	Performance of tef [ <i>Eragrostis tef</i> (Zucc.) Trotter] genotypes for yield and yield components under drought-stressed and non-stressed conditions
4	Genetic variation and trait association of tef [ <i>Eragrostis tef</i> (Zucc.) Trotter] evaluated under optimal and moisture stressed environments
5	Assessment of the genetic relationship of tef ( <i>Eragrostis tef</i> ) genotypes using SSR markers
6	Gene action and early generation selection of yield and related traits among tef populations under moisture-stressed and non-stressed environments
--	An overview of the research findings

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# Chapter 1: Review of Literature

## 1.1 Introduction

Tef [*Eragrostis tef* (Zucc.) Trotter] is an allo-tetraploid ( $2n=4x=40$ ), small cereal grain crop that belongs to the family Poaceae, sub-family Eragrostoideae, tribe Eragrostidae and genus *Eragrostis* (Ketema, 1997). Its origin and diversity is in Ethiopia (Vavilov, 1951). Tef is the most important cereal crop in Ethiopia for its largest area coverage and its high value for food, feed, but with low grain yield of  $1.6 \text{ t ha}^{-1}$  (CSA, 2016). Drought is one of the main contributing factors for its low yield level.

Most resource constrained farmers in sub-Saharan Africa including those in Ethiopia, face food insecurity due to regular droughts that are often exacerbated by climate change. In Ethiopia, agricultural activities and crop productivity are influenced by a wide range of environmental variables, including altitude, soil conditions, rainfall and temperature (Kassahun, 2013). As a coping strategy to deal with the risk of drought, smallholder farmers cultivate various crops and landrace varieties with different maturity periods. Due to its versatile environmental adaptation, tef grows in high rainfall as well as in drought-prone agro-ecologies. The crop yields relatively well in seasons with good rainfall, but also provides limited yields during drought periods, without complete crop failure as would occur with other cereal crops such maize and sorghum.

Drought tolerance is a complex quantitative trait controlled by many genes and affected by the environment and genotype by environment interactions (Xoconostle-Cazares et al., 2010). Breeding for drought tolerance depends on the accumulation of additive genes, a controlled stress screening environment and high throughput selection methods to maximize selection gains (Blum, 2011). This review summarizes the literature on tef breeding in Ethiopia, farmer's selection criteria, morphological and DNA level genetic variation in tef, heritability and trait association of tef, yield losses and the implication of drought, mechanisms of drought tolerance, genetic diversity and environments in breeding for drought tolerance, drought tolerance adaptive traits, gene action of drought tolerance and selection method for developing drought tolerant varieties. The aim of the review is to provide perspectives on current status and future research directions of the genetic management of drought in tef for reducing losses incurred by moisture stress. Literature from related cereal crops is adapted wherever necessary because of the scarcity of literature that deals specifically with breeding tef for drought tolerance.

## 1.2 Breeding tef in Ethiopia

Tef breeding program in Ethiopia started in late 1950s and since then about 35 improved tef varieties have been released (MoA, 2014). The objectives of the national breeding programs include developing lodging tolerant, high grain yield, moisture-stress tolerant and good quality tef varieties. Tef accessions acquired from Ethiopian Biodiversity Institute (EBI), collected from farmers field or market, core-collection and released varieties conserved at Debre-Zeit Agricultural Research Center (DZARC) and other regional centers are used as source of genetic variation either for direct selection or to be used in hybridization for improving traits of interest. Before the major findings on the floral biology of tef (Berhe, 1975), breeding was mainly focused on mass and pure line selection from landraces (Ketema, 1997; Assefa et al., 2011a) and about ten varieties have been released using these breeding methods. After the discovery of tef flower, effective hybridizations were achieved involving intra-specific crosses and some inter-specific crosses especially with *Eragrostis pilosa*. Due to the difficulty of tef emasculation and pollination which are conducted under the microscopy and the limited skilled manpower and infrastructure, tef crossing is conducted only at DZARC which is the national tef improvement center. Tef is strictly a self-fertilizing (02.-1% cross fertilization) crop with very small flower, in which its emasculation and pollination needs to be done under stereo-microscope (x15 magnification). Naturally, tef flower opening and pollination happens early in the morning (6:45 to 7:45 h). Flower opening can be delayed till noon by putting the plant in a complete dark place with a temperature of 14 to 22°C (Ketema, 1997).

Following crossing, segregating materials were selected using the modified pedigree or bulk methods of breeding (Assefa et al., 2011a) and presently the single seed decent method is commonly used. After initial evaluations and selections, the Ethiopian national breeding and germplasm testing procedure mandates multi-location preliminary and national variety trials in tef variety development program. Selected candidate and best performing lines from the national variety trials need to be tested at research stations and farmers' fields for performance verification. The national variety release committee evaluates the candidate lines for agronomic performance, distinctness, uniformity and stability using the data received from the breeder (Setimela et al., 2009; Assefa et al., 2011a). Additionally the committee has to do field evaluations of the candidate varieties at research station and farmers field conditions. After getting a positive feedback from the committee the variety is multiplied for seed increase and release (Assefa et al., 2011a).

Despite the limited resources, tef breeding has been successful in developing farmers desired varieties such as DZ-Cr-387 (released in 2006), which has been widely adopted in optimum tef growing environments (Assefa et al., 2011b; Awesa et al., 2015; Berhe, 2015). This had led in changing the livelihoods of millions of farmers. Tef variety named, DZ-Cr-438 (RIL No. 133B), was released in 2014 for its better lodging tolerance and high yield potential. The performance of both varieties in moisture stressed conditions is poor, due to their late maturity. This demands for the development of high yielding varieties with better adaptation to the moisture stressed areas in the country.

### **1.3 Farmers variety selection criteria**

Conventionally researchers develop technology at research stations and deliver the end product for adoption and expansion to farmers. This top down process, results in less adoption of the technology, especially in the resource poor farmers and marginal environments (Morris and Bellon, 2004; Ponzio et al., 2013). This challenge is getting attention and driving the assumption that participatory research could be the lead to development of technology which would address the demand of the farmers (Ceccarelli and Grando, 2007). Farmers are primary source in getting reliable information regarding the crop they produce. Therefore participation of farmers in identifying and prioritizing major crop production constraints is important in having a good start of successful research.

Participatory rural appraisal (PRA) is one of the participatory research methods helpful for identification and prioritization of production constraints and farmers' indigenous knowledge which can be useful to develop problem solving scientific research. This approach is becoming popular for its contribution for agricultural scientists to learn the real productivity problems of farmers and accordingly to develop scientific research (Henman et al., 2001; Abdullah et al., 2012). Chambers (1994) described PRA as an approach and method to enable local people to share, enhance and analyze their knowledge of life and conditions, to plan and act. Abay et al. (2008) emphasized the advantage of PRA method for breeders to capture indigenous farmer's knowledge and incorporate in their breeding program for developing the best performing varieties. Ethiopian farmers have been growing tef for a very long time. Therefore they have rich indigenous knowledge systems in managing crop production and its production problems. PRA would be helpful to exploit this indigenous knowledge and understand the farmer's choice of traits of a variety be considered in the tef breeding.

In Ethiopia 35 tef varieties are thus far released (MoA, 2014). Of these varieties, namely DZ-01-354, DZ-01-196, DZ-Cr-974, DZ-Cr-37 and DZ-Cr-387 are the most widely adopted by farmers (Assefa et al., 2011a). This is because the other varieties did not meet the farmers' selection criteria. DZ-Cr-387, the most popular improved variety which has the farmers' most preferred traits such as high yield, white seed colour and good *injera* making quality, which contributed to its high adoption (Belay et al., 2008; Assefa et al., 2011b; Gemechu and Alemu, 2016). Farmers in different agro-ecology do have varied varietal selection criteria. Wale and Chianu (2015) reported that farmers in the marginal areas prefer tef varieties with early maturity, yield stability, and adaptable to local environments while the rich farmers in the optimum tef growing areas prefer high yielding varieties. Market and social value, storability, grain colour, lodging tolerance, straw yield, threshability and resistance to diseases and pests were also described as farmers preferred traits in the selection of tef varieties (Fentie et al., 2012; Melkam et al., 2014). Farmer's selection criteria for a variety could be different depending on the environment and market condition, in which breeders should be aware of and consider accordingly. Therefore, knowledge on the desired varieties and their attributes should be updated continuously depending on the social, economic and environmental condition of the society where the variety is going to be used.

#### **1.4 Morphological and DNA level genetic variation in tef**

Characterization and quantification of genetic variation for valuable traits of crop germplasm is the main task of plant breeding programs. Genetic variation can be assessed using morphological traits, biochemical and molecular markers. Morphological traits have been the most widely used traits to determine genetic variation in many crops (Ali et al., 2009; Ahmadi et al., 2012; Hossain et al., 2015), and their use will be continued, despite their limitations due to the genotype and environment interaction. Tef is grown in a wider agro-ecology and farmers in the different agro-ecology apply different selection criteria such as selection for seed colour, grain yield, straw yield and palatability for animals' feed, *injera* making quality and, maturity which resulted in evolution of landrace with diverse traits. Previous studies indicated the existence of substantial genetic variation of different morphological traits (Assefa et al., 1999, 2001; Admass and Belay, 2011; Ayalew et al., 2011; Shiferaw et al., 2012; Plaza-Wuthrich et al., 2013) and physiological traits such as osmotic adjustment, leaf water relations and root length (Ayele et al., 2001; Degu et al., 2008) of tef genotypes tested under different environments. Most of the studies were targeted at optimum tef growing environments. Genetic variation of genotypes is subject to the testing environmental condition. Evaluation of tef

genotypes for their yield and yield component performance in moisture stressed environments would be important to exploit the tef genetic diversity for breeding high yielding and moisture stress tolerant varieties.

Molecular markers are widely used to determine DNA level genetic diversity for many crop plants. Molecular markers are identifiable DNA sequences, found at specific locations of the genome and thus can be used for identification of a germplasm by a specific pattern of polymorphisms, to assess diversity and determine genetic relationships. Unlike morphological markers, molecular markers are detectable and stable regardless of the growth stage of the plant and the environment (Collard et al., 2005; Agarwal et al., 2008). Use of molecular markers in tef breeding is limited. Earlier studies by Bai et al. (1999) and Ayele et al. (1999) using amplified fragment length polymorphism (AFLP) markers reported low polymorphism of 1-17.5% and 85-90%, respectively. Similarly Bai et al. (2000) reported low polymorphism of 84–96% among tef accession using random amplified polymorphic DNA (RAPD) markers. While genetic diversity study using inter simple sequence repeat (ISSR) indicated a similarity coefficient of 26-86%, which showed the existence of higher polymorphism in tef (Assefa et al., 2003). SSR markers developed by Zeid et al. (2011) and validated by Zeid et al. (2012) revealed genetic similarity coefficient ranging from 21 to 99 % indicating high level of genetic diversity in tef.

### **1.5 Inheritance and traits association in tef**

Heritability is the degree of resemblance of individuals and it can be defined as the ratio of the genotypic variance to the total phenotypic variance (Falconer and Mackay, 1996). Traits with greater heritability can be fixed more easily by selection than traits with lower heritability. Selection for yield must target on traits with sufficient additive genetic variance, which is related to heritability. Days to anthesis and days to maturity are among the easily measured morphological traits which are mostly indicated for their high heritability in tef (Ayalneh et al., 2012; Shiferaw et al., 2012; Jifar et al., 2015) and in other crops such as wheat, sorghum, and rice (Mondal et al., 2013; Ogunbayo et al., 2014; Abraha et al., 2015). In tef, other yield component traits such as plant height and panicle length (Assefa et al., 2001; Dargo, 2016), panicle seed weight and harvest index (Ayalneh et al., 2012) were also reported for their higher heritability than grain yield. In order to have information on how much of the heritable trait is achieved in selection, it is important to know the progress attained from selection and this is measured as genetic advance. Genetic advance depends on the genetic variability of the base population, heritability of a trait under selection and selection intensity. High genetic advance

translates into more progress from selection. Previous studies in tef showed that grain yield has a relatively higher genetic advance (Tefera et al., 2003; Chanyalew et al., 2009; Admass and Belay, 2011; Ayalew et al., 2011; Mewa et al., 2013). Trait for selection should combine both high heritability and genetic advance.

The yield of a crop plant is a complex trait influenced by the performance of associated yield components. The direction and strength of correlations of traits of a crop plant is useful in planning, evaluating and setting selection criteria for the desired characters in a breeding program. In absence of well-equipped laboratories and technologies to measure physiological traits, selection for high yielding genotypes in moisture stressed conditions could be done through indirect selection for easily measurable traits which are highly correlated with grain yield (Tuberosa et al., 2012). Grain yield of tef is reported for its strong positive correlation with panicle seed weight (Tefera et al., 2003), biomass yield and harvest index (Lule and Mengistu, 2014; Jifar et al., 2015). Under moisture stressed and non-stressed conditions, Shiferaw et al. (2012) mentioned a strong positive correlation of grain yield with panicle seed weight, plant height, harvest index and biomass yield. Days to maturity is reported for its negative association with grain yield (Tefera et al., 2003; Plaza-Wuthrich et al., 2013; Jafar et al., 2015). Ayele et al. (2001) reported positive association of osmotic adjustment with delayed wilting and higher leaf relative water content, but not associated with root length.

Information on the direct and indirect influence of yield components on grain yield is essential to prioritize for traits with strong direct influence. Path analysis is a statistical tool that helps to identify the most influential predictor variable useful for simultaneous selection (Singh and Chaudhary, 1977; Dabholkar, 1992). For their strong direct effect, selection for increased harvest index and biomass could lead to increase grain yield of tef (Ayalew et al., 2011; Debebe et al., 2013).

## **1.6 Drought and yield loss in tef production**

Drought is one of the main challenges facing world agriculture (Fang and Xiong, 2015), even though its effects may vary from region to region. Ethiopia is one of the sub-Saharan African countries facing recurrent droughts leading to low crop productivity or crop failure and food insufficiency (Deressa and Hassan, 2009). Declining levels and high variability of rainfall is among the main causes for low crop productivity in different parts of Ethiopia (Tilahun, 2006; Abrha and Simhadri, 2015; Fekadu, 2015). Farming systems study indicated moisture stress as one of the main contributors for yield reduction in tef (Yizengaw and Verheye, 1994). Drought is

predicted to cause reduction of 24% of the current suitable land for tef production (Yumbya, 2014) and can significantly impact on reduction of tef yield (Araya et al., 2015). Yihun et al. (2013) reported 1 t ha<sup>-1</sup> grain yield reduction due to 25% soil moisture deficit at mid growth stage of the crop. Similarly, 25.5% and 51% grain yield reduction was reported to be caused by moisture stress during pre and post-anthesis period, respectively (Admas and Belay, 2011; Shiferaw et al., 2012). Studies on impact of drought on tef yield are limited.

## **1.7 Breeding for drought tolerance**

### **1.7.1 Mechanisms of drought tolerance**

Plants respond to drought with different mechanisms such as drought escape, dehydration avoidance and dehydration tolerance (Blum, 2011; Shashidhar et al., 2013; Tardieu, 2013). Drought escape is a mechanism in which plants mature early before drought occurs. This mechanism is advantageous in environments with frequent terminal drought stress (Tuberosa, 2012; Krannich et al., 2015). Dehydration avoidance can be defined as the plant's ability to retain a relatively higher level of hydration under stress conditions. It is a mechanism where plants maintain their water potential through controlling the stomata opening and developing a deep and prolific root system (Mitra, 2001). Dehydration tolerance describes the ability of plants to continue metabolizing at low leaf water potential and to maintain growth despite dehydration of the tissue, or to recover after release from stress conditions (Oliver et al., 2010).

### **1.7.2 Traits for selection to drought tolerance**

#### *1.7.2.1 Phenological and morphological traits of drought tolerance*

Among the different morphological traits, early crop establishment and vigor, early flowering and maturity, leaf rolling and a deep and extensive root system are the most commonly evaluated ones for selection of drought tolerant genotypes (Gowda et al., 2009). In a dry climate, vigorous growth and efficient utilization of plant growth resources are desirable crop traits for good yield performance (Lidon and Cebola, 2012). Early and vigorously developed leaf biomass of a genotype enables storage of water that can be used in later growth stages when soil moisture is less available. This reduces the inhibition of stomata conductance (Monneveux et al., 2012).

Flowering and maturity period are two easily measured traits to be considered in screening trials. In optimum environments, the number of days to flowering and maturity has a positive correlation with yield while under moisture stress the correlation is negative. Therefore,

selection should be targeted for early flowering and maturity genotypes (Kilic and Yagbasanlar, 2010; Blum, 2011). Plasticity of these traits enables the crop plants to have wide adaptation to environmental fluctuation by adjusting their growth duration to the specific environmental situation (De Rouw and Winkel, 1998). In many cereal crops, early maturity has been among the targeted major drought avoidance mechanisms (Fischer and Fukai, 2003; Allah et al., 2010; Mondal et al., 2013; Manga et al., 2015; Mondal et al., 2016). Moisture stress during the grain filling stage is a critical period which leads to significant yield loss in tef (Tsegay et al., 2012), indicating the importance of selection for early flowering and maturity. Drought tolerance study on recombinant inbred lines (RIL) of tef along with varieties, Key Murri, Cross-37 (a widely grown drought tolerant improved variety) and *Eragrostis pilosa* showed that, some of the RIL populations, DZ-Cr-37, and *Eragrostis pilosa* were all early maturing genotypes that could escape late season drought events and yielded better under these conditions than the other varieties (Admas and Belay, 2011). Similarly, Fentie et al. (2012) recommended an early maturing tef variety, Cross-37, for its high yield in north western Ethiopia. Tef landraces are reported for their wider maturity periods leading to wide adaptation in different environments (Assefa et al., 2015), while the majority of high yielding tef genotypes are late maturing (Ayele et al., 2001; Bedane, 2015). Genes for earliness could be introgressed into the high yielding varieties through a planned hybridization and selection, to combine both high yield and early maturity.

Leaf rolling is a dehydration avoidance mechanism. It assists in the reduction of transpiration water loss and often occurs if the plant leaf water potential is reduced (Kadioglu and Terzi, 2007; Kadioglu et al., 2012). This trait is also easily measured and can be evaluated in large number of genotypes grown under moisture stress. Another morphological trait that mitigates drought events is deep and extensive root systems that enable plants to exploit moisture from deeper soil layers. Breeding and selection for root characteristics can, therefore, provide an indirect selection for drought tolerance in crop plants (Chloupek et al., 2010). Drought tolerant tef genotypes showed longer root (Ayele et al., 2001; Degu et al., 2008) indicating the importance of this trait for drought tolerance selection. However, this trait is difficult and costly to evaluate (Babu, 2010). Consequently it is desirable to look for other traits that are correlated with root architecture to undertake indirect selection, especially when screening large numbers of genotypes.

### 1.7.2.2 *Physiological drought tolerance traits*

Physiological traits such as chlorophyll content, stomata conductance, canopy temperature and osmotic adjustment are among the important parameters found to be well-correlated with yield performance of genotypes under stressed environmental conditions. Crop growth is dependent on photo-assimilates of the whole plant, and proper functioning of photosynthetic pathways depends on continued transpiration and gas exchange through opened stomata (Arve et al., 2011). In tef, a 95% reduction of stomatal conductance was reported, when plots were watered at 25% field capacity (severe moisture stress level) (Mengistu, 2009). This reflects the sensitivity of stomata openings in tef to moisture stress that has adverse effects on yield through reduced rates of photosynthesis.

Plants keep their leaves at an optimum temperature through transpiration. Under moisture stress they avoid loss of water by closing their stomata, which leads to increased leaf temperatures. High temperatures impact on enzyme efficiencies in the leaves (Gonzalez-Dug et al., 2006). Drought susceptible genotypes that showed greater yield losses under drought tended to have warmer canopies. Lower canopy temperature of plants can be useful as an indicator of drought tolerance and its measurement is recommended at the vegetative growth stages (Monneveux et al., 2012).

Osmotic adjustment is a process that lowers osmotic potential and increases leaf water potential due to the accumulation of soluble compounds in plant cells. This enables maintenance of cell turgor, improved soil water extraction and enhanced water use efficiency. The accumulation of organic compounds such as proline provides stabilization and protection of cell membrane systems (Zivcak et al., 2009). High osmotic adjustment enabled tef genotypes to delay wilting and hence retain higher leaf water content (Ayele et al., 2001; Degu et al., 2008)

### 1.7.3 **Genetic diversity and environment in breeding for drought tolerance**

Breeding for drought tolerance largely depends on the existence of genetic diversity within the target and related crop species. The initial population for screening must be large and diverse enough to ensure that adequate genetic variability is represented to achieve strong selection response (Fischer et al., 2003). Selected cultivars, landraces, wild species, mutants and transgenic plants are some of the sources of diversity in breeding for drought tolerance. Crossing of locally adapted varieties with improved varieties is recommended in developing drought tolerant varieties (Fischer et al., 2003). Often, the local varieties are a good source of

genes for drought tolerance whereas the improved varieties are mostly known for their high yield (Shashidhar et al., 2013; Haddadin, 2015). Tef productivity study under terminal drought in northern Ethiopia showed that, under severe moisture stress, the local tef varieties, Abat-Nech, Kobo and the widely grown drought tolerance improved variety, DZ-Cr-37 had comparative grain yields of 1.43, 1.42 and 1.48 t.ha<sup>-1</sup>, respectively. In contrast, the improved varieties, namely; DZ-Cr-358, Dz-01-1281, DZ-01-1681 and DZ-01-99 yielded 0.93, 1.33, 0.60 and 0.31 t.ha<sup>-1</sup>, respectively, indicting the sensitivity of the improved varieties to terminal drought stress (Mengistu and Mekonnen, 2011).

High throughput phenotyping for drought tolerance requires adequate control of the stress environment. Among other factors, site homogeneity and controlled drought conditions are two major factors. The site should be uniform in topography, soil fertility status, previous season cropping, and moisture status (Cattivelli et al., 2008). For a managed drought tolerance study, different methods can be employed to control unwanted rainfall such as the use of dry season environment, delayed planting, rain-out shelters and greenhouses (Blum, 2011). In using delayed planting, it is important to know the planting time of the area at which the trial is to be conducted and the likelihood of unwanted rainfall to avoid its confounding effect on deliberate moisture stress. In semi-arid environments where the rainfall is erratic and unpredictable, it is difficult to conduct drought tolerance studies using delayed planting. Rain-out shelters offer the possibility of investigating the adaptive response of crops to a desired level of drought stress through avoiding the unpredictable rainfall events. An off-season study of drought tolerance is a common approach in the semi-arid tropics (Blum, 2011). But during the off-season the climatic conditions may be different from the main season; so it is important to understand the off-season conditions of the location where the trial is to be conducted. Greenhouse pot experiments are also an option to study drought tolerance in an artificial environment. This is mostly used in studying root characteristics of crop plants under drought conditions. It is easy to manage the stress conditions but the greenhouse soil and environmental conditions will be different from what occurs in the field (Passioura, 2012).

#### **1.7.4 Gene action of drought tolerance**

Drought tolerance is a complex trait, in which its expression depends on the action and interaction of different genes, and the environment, which control morphological, physiological and biochemical characters. The success of any breeding program for developing drought-tolerant varieties depends on precise estimates of genetic variance components of the traits of

interest that mainly consist of additive, dominant and epistatic genetic effects (Nouri et al., 2011). Additive gene action is associated with the occurrence of heterozygotes performing intermediate between the two homozygotes, with respect to a particular character. Dominance refers to a type of gene action where the heterozygote provides the same performance as dominant homozygotes. Epistasis refers to a condition where two or more gene loci interact to determine the performance of a genotype (Chahal and Gosal, 2002; Sleper and Poehlman, 2006). Information on the mode of gene action of targeted traits of a specific crop in specific environment is helpful for deciding the selection methods to apply (Acquaah, 2007). In self-pollinated species, if additive gene action is predominant, then the breeder can effectively select at various levels of inbreeding and result in substantial improvement of the trait of interest, because additive effects are fixable and readily transmissible from one generation to another. On the other hand, sufficient non-additive gene action may warrant the production of hybrids as it maximizes heterosis.

In wheat, studies indicated the predominance of additive genes in controlling drought tolerance (Farshadfar et al., 2013; Rad et al., 2013; Farshadfar and Amiri, 2015). On the contrary, Yogameenakshi and Vivekanandan (2015) reported the predominance of dominance gene action for the majority of the drought tolerance traits. There is no information on gene action and inheritance about *tef* under moisture stress conditions. While under non-stressed conditions, Tefera and Peat (1997) reported dominance gene action for grain yield and panicle weight and a large portion of additive gene action controlled plant height, panicle length, days to heading and days to maturity. Similarly, Tefera (2002) reported largely additive gene effects controlling plant height, kernels per spikelet, primary panicle branches, panicle weight and spikelets per panicle, while there were small additive effects for grain yield.

#### **1.7.5 Selection of drought tolerant genotypes using single seed descent**

Single seed descent (SSD) is one of the most important selection methods for complex quantitative traits such as drought tolerance. It is a procedure of advancing early segregating generation through a single seed derived from each plant of diverse crosses. In this method, selection of progenies and families starts at the  $F_6$  or  $F_7$  generations, enabling the accumulation additive genetic variance effects. From the  $F_8$  onwards, multi-location preliminary and national variety trials can be conducted as per national testing requirements (Chahal and Gosal, 2002; Acquaah, 2007). This procedure was developed with the aim of advancing large numbers of  $F_2$  plants from diverse cross combinations to yield trials to assess their potential for producing lines

with a full spectrum of desirable characteristics. In self-pollinated crops such as tef, additive genetic variance along with additive x additive gene action can be exploited through the use of SSD selection method. To exploit the total additive genetic variance, continuous selfing is required to attain homozygosity. Other selection methods such as pedigree selection or pure line selection begin at earlier generations leading to a loss of additive genetic variance (Sleper and Poehlman, 2006) while at the F<sub>2</sub> generation, only half of the additive genetic variance is present and could not guarantee high selection response.

The SSD method has been used by various researchers to fix additive genetic variance. Lalic et al. (2003) compared barley genotypes developed by pedigree and SSD methods. The authors reported that genotypes developed through SSD selection showed superior grain yield performance to the genotypes developed through the pedigree method. Knott and Kumar (1975) also reported the advantages of SSD methods for developing superior yielding wheat genotypes and reduced the cost of handling segregating generations relative to the pedigree selection method. Similarly better yielding inbrelines and hybrids were developed through modified SSD than pedigree and double haploid methods in maize (Jumbo et al., 2011) and using SSD in rice (Kanbar et al., 2011). In tef breeding, pedigree and bulk selection methods were used in handling segregating populations (Assefa et al., 2011a) but presently SSD has become the dominantly applied selection method in tef. This method was successfully used in the development of the widely adopted tef variety, "DZ-Cr-387 (Quncho)", which was selected for its high yield and yield components performance (Assefa et al., 2011b). Tefera and Peat (1997) and Tefera (2002) also recommended leaving selection of tef segregants to advanced generations to increase homozygosity to enable the exploitation of additive gene action for quantitative traits such as yield and drought tolerance.

#### **1.7.6 Pre-breeding and breeding of drought tolerant tef in Ethiopia**

There is limited information on the genetic basis of drought tolerance in the Ethiopian tef landraces, cultivars and breeding lines despite the significance of the crop and occurrence of frequent droughts in the country. Understanding the present information on genetic management of drought tolerance in related cereal crops should aid in the identification of an appropriate breeding strategy. This will enable tef breeders to routinely introduce candidate genes into drought susceptible breeding lines for cultivar release. These genes should be introgressed into the widely used tef cultivars as a means to increase drought tolerance.

Given the need to breed tef for drought tolerance and other relevant agronomic traits, the Ethiopian Biodiversity Institute and the Ethiopian Tef Breeding Project based at Debre-Zeit have collected a wide genetic pool of tef germplasm. These genetic resources can be explored to combat drought by searching for novel drought tolerant genes aimed at developing germplasm with drought tolerance. Phenotypic field selection can be complemented with genomic techniques to identify and diagnose effective drought tolerance genes. Marker-assisted selection (MAS) has been proven to be a powerful tool to aid in the development of cultivars with drought tolerance in other crops such as wheat, rice and pearl millet (Jongdee et al., 2006; El Ameen, 2013).

There is a need to develop and identify diagnostic molecular markers that are linked to effective drought tolerance genes in tef for routine applications of MAS in breeding programs. The markers linked to drought tolerance genes could be used to predict the presence of specific genes and might help in the transfer several genes into adapted materials. Presently, low cost and next-generation sequencing platforms are becoming available. Consequently, more diagnostic markers may be made available for high-throughput screenings and application of MAS in tef breeding for drought tolerance. Genome sequence of tef is available (Cannarozzi, 2015) which is going to be a useful for discovery of genes responsible for important traits and to improve efficiency of the tef breeding. To date, there is no research on gene cloning in tef aimed to incorporate drought tolerant genes into suitable genetic backgrounds. Tef breeders should consider using the doubled haploid (DH) breeding technique, which has the potential to reduce the number of selfing generations required for maximum homozygosity and release of new varieties (Gugsa and Kumlehn, 2009; Tadesse et al., 2013). Overall, pre-breeding and breeding efforts should be strengthened to develop drought tolerance cultivars in Ethiopia. With advancements in high throughput technology and MAS, tef breeding programs can integrate new sources of drought tolerance into the existing gene pool to sustainably manage drought in water limited agro-ecologies.

## **1.8 Conclusions**

Tef is an important staple food grain crop in Ethiopia, and it is internationally gaining popularity as a health food. Despite its great economic and nutritional importance, the current tef yield is low, about 1.6 t ha<sup>-1</sup> (CSA, 2016). Moisture stress is a major contributor for its low yield. Breeding drought tolerance tef varieties would provide a genetic management approach to enhance and sustain tef productivity, especially in the drought-prone regions.

Breeding for drought tolerance remains the most feasible and economical approach to drought management. Tef breeders need to continuously search new sources and introgress these genes into susceptible cultivars. Germplasm screening using both phenotypic and genotypic data is important to identify drought resilient breeding lines. Furthermore, the national breeding project should strengthen capacity development efforts for effective breeding for drought tolerance in the country. The need for international collaborative initiatives needs to be emphasized to adapt new and advanced technology. This initiative will assist in the release and promotion of new cultivars with drought tolerance genes aligned with rapid seed multiplication systems and institutional coordination. There is a need for an intensive study and understanding of the genetics and variability of drought tolerance which will require research funds to be allocated to breed better tef cultivars.

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## Chapter 2: Assessment of Production Status, Constraints, Management Practices and Farmer-Preferred Traits of Tef [*Eragrostis tef* (Zucc.) Trotter] Under Drought-Prone Environments: Implications for Future Research Directions

### Abstract

Tef [*Eragrostis tef* (Zucc.) Trotter] is an ancient small grain crop being advocated globally as a health food for people with gluten intolerance and a major staple food in Ethiopia. Yet, tef has low national average yield level of 1.6 t ha<sup>-1</sup>. Up-to-date information on the production potential, constraints, crop management practices, input use and farmers-preferred traits of the crop under marginal and moisture stressed environments is scanty. This study was, therefore, carried out to assess the current production and productivity status, crop management practices and farmers' trait and variety preferences of tef in moisture stressed growing areas in order to devise optional breeding strategies. A two-stage sampling was followed to select a total of 240 farmers from four districts within three administrative Zones of Tigray Region, northern Ethiopia. Primary data through individual structured interview and secondary data from the Bureau of Agriculture and Rural Development (BoARD) were collected and subjected to analyses. The results indicated that more than 95% of the interviewed farmers grew tef in 2013 cropping season and about 57%, 61%, 50% and 51% of the farmers' households lands were allocated to tef production in the four districts of Laelay-Maichew, Ahferom, Alamata and Medebay-Zana, respectively. High grain yield, increased panicle length and straw yield were the most important farmer-preferred traits. In Alamata district, early maturity was the third preferred trait while straw yield ranked fourth. Improved varieties were used by the majority of respondents in all districts and fertilizers in all districts except Alamata where severe moisture stress dictated the choice of variety and reduced fertilizer application. Transplanting maximized the yield of tef, but a cost-benefit analysis showed that row sowing was more profitable. Moisture stress (60.4%), weed (61.2%), shoot fly (58.2%), lodging (53.3%) and seed shattering (60.8%) were rated as severe tef production constraints in the study areas. Estimated tef yield reductions of about 48%, 60%, 37% and 49% were reported by farmers due to moisture stress in Ahferom, Alamata, Laelay-Maichew and Medebay-Zana districts, respectively. This study prioritized production constraints and farmers'-preferred traits useful in the breeding of tef to enhance its productivity under drought-prone environments in northern Ethiopia.

**Key-words:** Farmer preferred traits, moisture stress, participatory rural appraisal, tef

## 2.1 Introduction

Tef [*Eragrostis tef* (Zucc.) Trotter] is the major staple food in Ethiopia; it is also now being grown in other countries because of its gluten free properties (Assefa et al., 2011a). Its most common use is for the preparation of fermented tef flour for the staple food known as “*injera*”. The attraction for growers is tef’s high flour productivity, high market price, and adaptability to wide range agro-ecologies. Pests and diseases are considered to have a relatively small impact on crop health both in the field and in storage. Tef straw is a preferred animal feed because of its high palatability and feed quality (Ketema, 1997).

Tef has a wide range of ecological adaptation in Ethiopia and can be cultivated from sea level up to 2800 m above sea level (m.a.s.l.); however best performance occurs between 1800 to 2100 m.a.s.l. It is well-adapted to a temperature range of 10 to 27°C and performs well in day lengths of around 12 h (Ketema, 1997). The annual rainfall requirement of the crop ranges from 950-1500 mm (FAO, 2011). Tef has wide area coverage of 2,866,052.99 ha with 1.6 t ha<sup>-1</sup> productivity (CSA, 2016), which is low. This low yield of tef is mainly attributed to lodging, moisture stress, shattering, and poor pre- and post-harvest agronomic management (Assefa et al., 2011a). Ketema (1993) estimates that tef has a potential productivity of 6 t ha<sup>-1</sup>.

Yield losses of tef due to low moisture are estimated to reach up to 40% during severe stress (Ayele, 1993). Furthermore, yield reduction of up to 77% has been reported to have occurred as a result of drought at the anthesis stage of tef (Takele, 2001). Haile Selassie et al. (2011) studied tef production constraints in Tigray Region of northern Ethiopia, and found that in the highlands, poor soil fertility, and in the lowlands, low moisture stress, were the major constraints affecting tef production.

The use of agricultural inputs is a basic way to improve crop productivity (Muzari et al., 2012). However, in sub-Saharan African countries such inputs are limited due to socio-economic reasons (Salami et al., 2010; Van den Berg, 2013). Similarly in Ethiopia, the adoption of fertilizer practices and use of improved varieties is low (Ayele and Bosire, 2011; Fufa et al., 2011). The government of Ethiopia has set strategies to promote the adoption of inputs for improving crop productivity to attain food security (Chanyalew et al., 2010). Tef is one of the crops which have received considerable attention in this respect. There has also been a reluctance of farmers to adopt improved varieties and apply fertilizers, particularly in areas affected by moisture stress.

It has been argued that more efficient agronomic management could double the yield of tef (Mueller et al., 2012), however, this is a large expectation given that the crop is primarily grown by subsistence farmers using low input and output agronomic systems (Altieri, 2009; Yu and Nin-pratt, 2014). To date, little research and technological development has been directed towards helping these farmers deal with issues like managing planting populations, cultivation and weed control, and harvesting and threshing (Fufa et al., 2011); currently, the majority of the farmers practise broadcast sowing which is associated with high susceptibility to lodging (Asargew et al., 2014). To address these problems, the Ethiopian Agricultural Transformation Agency (EATA) promotes activities such as how to determine optimum seed rate, and also transplanting seedlings into rows, a practice that is expected to result in increased crop productivity (Ehsanullah et al., 2000; Lafarge and Tubana, 2008) because of increased tillering and reduced lodging (Axum Agricultural research Centre (AxARC), 2013). As yet, there is limited information on the efficiency and acceptance of these technologies in solving the production constraints.

Farmers are the main actors in agriculture in that they have a wealth of knowledge about their crops, farming systems and constraints, and have their own means to adapt their environment (Altieri and Koohafkan, 2008). This indicates that before the start of any crop improvement activities knowing the farming system, and the variety and trait preference of the target environment is essential. This can be achieved through collection of primary data from farmers which will have great roles in the selection of a successful variety or development of other agricultural technologies and thereby in their ultimate adoption. The development of demand based tef varieties has led to the increased adoption of improved tef varieties. An improved tef variety known as 'Quncho' has received good acceptability for its high yield, white seed colour and relatively lodging resistance, which are farmers preferred traits in the optimum rainfall areas (Assefa et al., 2011a). However, the recently released varieties such as 'Gemechis' and 'Simada' for moisture stressed were not successfully adopted because they did not satisfy the farmers preferred traits in the area, indicating the need for identifying the targeted preferred traits of tef for strategic breeding.

There is little or no up-to-date information on the production potential, constraints, input adoptions and farmer-preferred traits of tef under marginal and low moisture stress growing environments in Ethiopia. Identification of the farmers' trait and variety preferences in these areas is crucial to devise breeding strategies. This will allow for the breeding of tef varieties both for optimum rainfall and drought- prone environments. This study was, therefore, undertaken to

assess the current production and productivity status, level of adoption of inputs, farmers' trait and variety preferences of tef in moisture stressed growing areas, and thereby to point out the implications for breeding.

## **2.2 Materials and methods**

### **2.2.1 Description of the study areas**

The study was conducted in 2014 in four major tef growing districts (viz. Laelay-Maichew, Medebay-Zana, Ahferom and Alamata) in the Tigray Region of northern Ethiopia (Figure 2.1). These four districts have wide altitudinal ranges (Table 2.1); the greatest is in Alamata which spans 2000 m elevation, although 75% of the area is at < 1800 m above sea level (m.a.s.l.).

The rainfall in Alamata is typically bi-modal in distribution; in the other districts it is mono-modal. Mean annual rainfall (2000-2013) ranges between 695 mm at Alamata to 961 mm at Medebay-Zana (Ethiopian Meteorology Agency, 2014). Thus in spite of the bimodal pattern at Alamata, this district has the lowest rainfall. Mean daily temperatures were also the highest at Alamata. Because the topography is mountainous in Tigray region, erosion can occur during the high rainfall events, although participatory soil and water conservation practices are currently being adopted to reduce the effects of erosion.

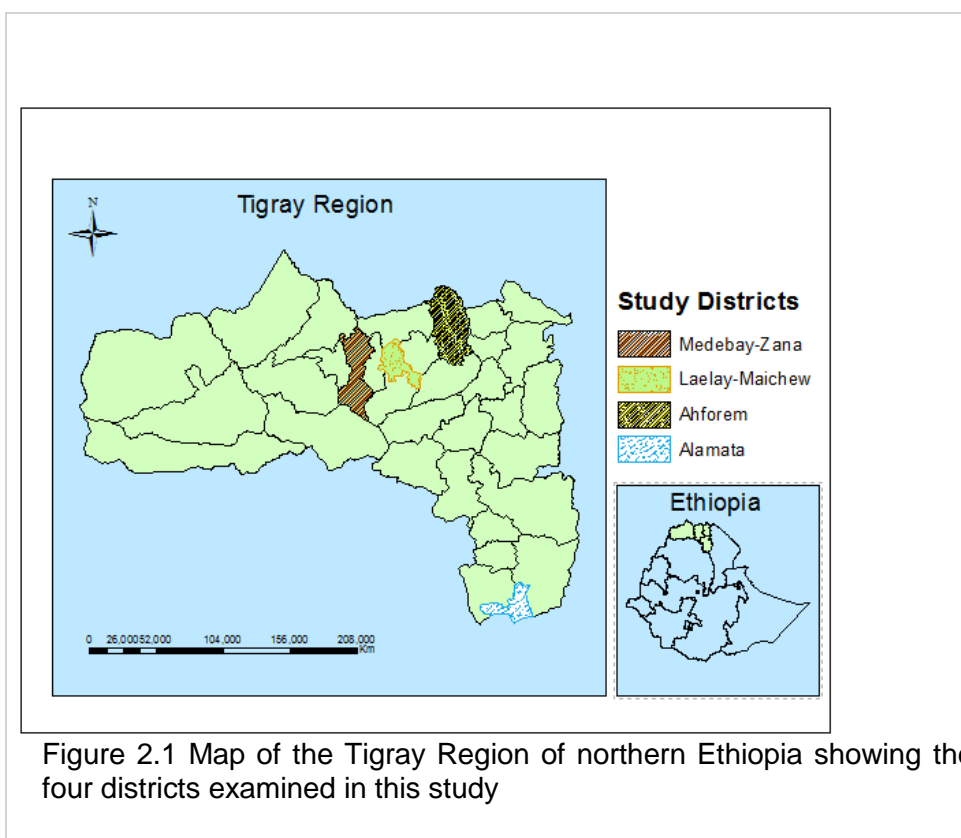


Figure 2.1 Map of the Tigray Region of northern Ethiopia showing the four districts examined in this study

Table 2.1 Altitudinal, weather (2000-2013) and climatic characteristics of the study districts

District	Altitude (m above sea level)	Daily mean temperature ranges (°C)	Mean annual rainfall (mm/year)	Agro-ecological Zone	Rainfall Pattern
Medebay-Zana	1500-2300	10.7 -26.5	961	Semi-arid	Mono-modal
Laelay-Maichew	1450-2050	11.1-27.8	703	Semi-arid	Mono-modal
Ahferom	1332-2921	14.0-22.0	716	Semi-arid	Mono-modal
Alamata	1178-3148	14.1-29.4	695	Semi-arid	Bi-modal

Source: Ethiopian Meteorology Agency and Bureau of Agriculture and Rural Development (BoARD) of each district

### 2.2.2 Sampling procedure

A purposive sampling procedure was followed to select three major tef-growing administrative zones within the Tigray Region; these were the Northern Western, Central and Southern Zones (Table 2.2). Two districts, Laelay-Maichew and Ahferom were selected from Central Zone and in the Southern and North-Western Zone, only one district each, Alamata and Medebay-Zana respectively, were sampled.

Two Peasant Associations (PA) from each district were selected from the 16, 18, 18 and 10 PAs presently found in Laelay-Maichew, Medebay-Zana, Ahferom and Alamata districts,

respectively. The PAs were selected based on their accessibility for transport and tef production coverage. Based on the degree of homogeneity of the population, time and resource availability, 30 households from each PA were randomly sampled providing a total of 240 respondents of which 72 (30%) were women-headed households (Table 2.2).

Table 2.2 Administrative zones, districts, Peasant Associations (PAs) and number of households by gender of household heads (respondents) sampled for the study

Administrative Zone	District	Name of Peasant Association	Number of farmers' households		
			Male-headed	Femal-headed	Total
North Western	Medebay-Zana	Hakfen	21	9	30
		Adi-kemalik	21	9	30
Central	Laelay-Maichew	Hastebo	21	9	30
		Debrebirhan	21	9	30
		Tahtaymegariastebri	21	9	30
		Sero	21	9	30
Southern	Alamata	Gerjelle	21	9	30
		Timuga	21	9	30
Total			168	72	240

### 2.2.3 Data collection

The data were collected in 2014; both primary and secondary data were collected. For the primary data a formal survey was employed in which household heads were interviewed using a structured questionnaire. The answers were based on their 2013 farming experience. The developed questionnaire was pre-tested on three farmers (10% of the sample) in each peasant association (PA). These farmers were not among those subsequently surveyed. The questionnaire was then modified to exclude unwanted questions and include others that were relevant to the study areas. Enumerators were trained to create a clear awareness to the interviewees of the subject matter of the questionnaire. The primary data collected covered the demographic characteristics of the households, tef area cultivated and productivity status, tef landraces grown and farmers preferred traits of tef varieties, uses of fertilizers and improved varieties, seed systems of tef, and different planting practices and their acceptability, production constraints and effect of moisture stress on tef and its coping mechanisms,.

Secondary data were collected from the district's Bureau of Agriculture and Rural Development (BoARD) and the Ethiopian Meteorology Agency.

## 2.2.4 Data analysis

The collected data were subjected to statistical analyses using the Statistical Package for Social Sciences (SPSS Inc., 2009). Descriptive statistics such as percentages, mean, standard deviation, minimum, maximum and ranges were calculated. The non-parametric Kruskal–Wallis procedure was used to test for significance differences between data sets for the four districts.

## 2.3 Results

### 2.3.1 Demographic characteristics of households

Table 2.3 summarizes the demographic features of the households of the study areas. The mean family sizes of the respondents were 5.8, 6.3, 6.6 and 5.5 for Laelay-Maichew, Medebay-Zana, Ahferom and Alamata, respectively. The majority of the respondents in Laelay-Maichew and Medebay-Zana had completed elementary education while in Ahferom (24%) and Alamata (37%) were illiterate. Mean ages of the respondents were 43.3, 46.7, 50.8 and 41.1 years in Laelay-Maichew, Medebay-Zana, Ahferom and Alamata, respectively (Table 2.3).

Table 2.3 Sex, family size, level of education and age of the sample households as percentage of respondents in four districts of Tigray Region in northern Ethiopia.

Description	Districts			
	Laelay-Maichew	Medebay-Zana	Ahferom	Alamata
<b>Sex</b>				
Male	70.0	70.0	70.0	70.0
Female	30.0	30.0	30.0	30.0
<b>Family size (number/household)</b>				
Minimum	2.0	2.0	2.0	2.0
Maximum	11.0	11.0	10.0	11.0
Mean	5.8	6.3	6.6	5.5
<b>Education level</b>				
Illiterate	21.7	23.3	40.0	61.7
Read and write	10.0	6.7	8.3	21.7
Elementary (grades 1-4)	26.7	36.7	33.3	8.3
Elementary (5-8)	31.7	25.0	15.0	5.3
Secondary (9-10)	8.3	8.3	3.3	0.0
Higher education	1.7	0.0	0.0	0.0
<b>Age (years)</b>				
Minimum	24.0	22.0	35.0	25.0
Maximum	74.0	68.0	68.0	68.0
Mean	43.3	46.7	50.8	41.1

### 2.3.2 Land ownership systems of households

The various forms of land ownership and land sizes of households across districts are presented on Table 2.4. The mean land size per family was < 1 ha in Laelay-Maichew, Medebay-Zana and Ahferom and close to 1 ha in Alamata. About 50, 55, 40 and 38%, of respondents in Laelay-Maichew, Medebay-Zana, Ahferom and Alamata, districts shared-in land in addition to their landholdings. The percentage of farmers who shared-out their lands was minimal (Table 2.4). About 1.7% and 3.3% of respondents in Laelay-Maichew and Alamata districts, respectively did not own land. Farmers who rent land were minimal in Laelay-Maichew, Medebay-Zana and Ahferom, while were highest in Alamata (Table 2.4).

Table 2.4 Forms of land ownership and mean land size (hectare) of households in the four study districts of Tigray Region in northern Ethiopia.

Land ownership	Laelay-Maichew		Medebay-Zana		Ahferom		Alamata	
	Respo ndents (%)	Land size (ha)	Respon dents (%)	Land size (ha)	Respon dents (%)	Land size (ha)	Respond ents (%)	Land size (ha)
Own land	98.3	0.7	100.0	0.7	100.0	0.8	96.7	1.0
Shared- in	50.0	0.4	55.0	0.6	40.0	0.4	38.3	0.8
Shared-out	5.0	0.2	3.3	0.3	3.3	0.6	3.3	0.5
Rented-out	1.7	0.1	-	-	-	-	3.3	0.2
Rented-in	3.3	0.3	13.3	0.4	-	-	23.3	0.7
No land	1.7	-	-	-	-	-	3.3	-

Some households have multiple land ownerships causing percentage of respondents >100%.  
Dash (-) indicates zero response

### 2.3.3 Farming systems

#### 2.3.3.1 Main types of crops grown: area coverage and productivity of tef

Figure 2.2 summarises household land allocated to the main crops they grow. Sorghum (*Sorghum bicolor* L. Moench), tef [*Eragrostis tef* (Zucc.) Trotter], bread wheat (*Triticum aestivum* L.), maize (*Zea mays* L.), barley (*Hordeum vulgare* L.) and finger millet (*Eleusine coracana*) were the dominant cereal crops cultivated followed by pulses such as field pea (*Pisum sativum* L.), fababean (*Vicia faba* L.), fenugreek (*Trigonella foenum-graecum* L.), chickpea (*Cicer arietinum* L.) and grass pea (*Lathyrus sativus* L.). Among the cereals, tef is the principal with mean share of 57%, 61%, 50% and 51% of the total household land in the Laelay-Maichew, Ahferom, Alamata and Medebay-Zana districts, respectively. In Medebay-Zana, tef was followed by maize, finger millet, pulses, barley and sorghum in terms of percentage of area cultivated (Figure 2.2). In Laelay-Maichew wheat, pulses, barley, maize, finger millet and sorghum

occupied 17.6%, 13.9%, 8.3%, 2.7%, 1.9% and 0.4% of the production area, respectively. In Ahferom district, the relative share was 20.1%, 11.5%, 2.8%, 2%, 1.6% and 1.4% for wheat, finger millet, maize, pulses, barley and sorghum, respectively. In Alamata, the area planted to sorghum, maize and barley were 28.1%, 12.0% and 0.3%, respectively (Figure 2.2).

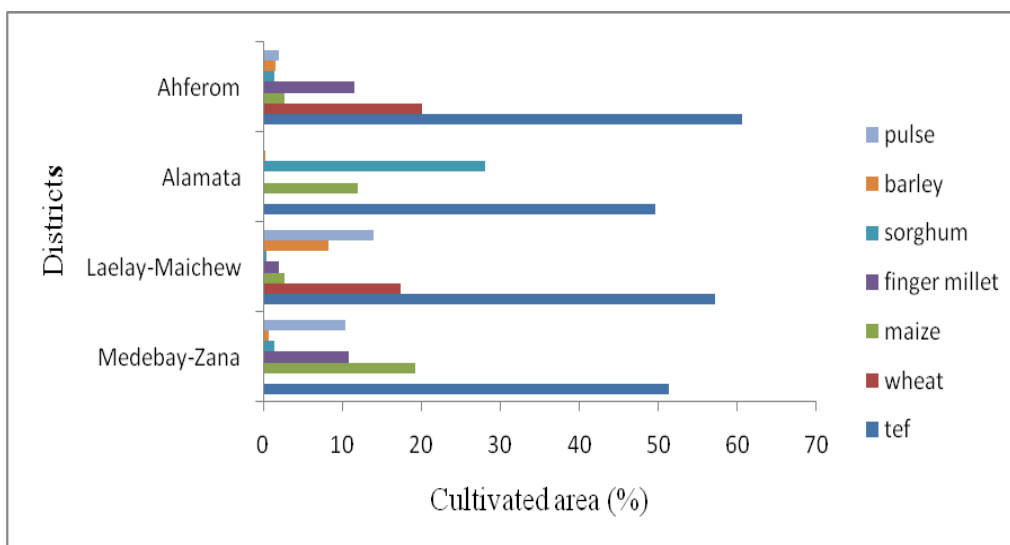


Figure 2.2 Percentage of households land allocated to different crops across four districts of Tigray Region in northern Ethiopia during 2013 cropping season.

Table 2.5 summarizes the percentage of respondents who grow different crops and the mean yield of each crop. As shown in Table 2.5 about 95%, 100%, 100% and 98.3% of the respondents in Laelay-Maichew, Medebay-Zana, Ahferom and Alamata, respectively, cropped tef in 2013 growing season. The mean grain yields of tef were highest in the Laelay-Maichew district at  $1.1 \text{ t ha}^{-1}$  followed by Medebay-Zana, Ahferom and Alamata each with  $1.0$ ,  $0.9$  and  $0.8 \text{ t ha}^{-1}$ , respectively (Table 2.5). In Laelay-Maichew, maize ( $1.9 \text{ t ha}^{-1}$ ) and wheat ( $1.7 \text{ t ha}^{-1}$ ) were highest yielders followed by finger millet, sorghum, faba bean, grass pea, barley, chickpea, tef, field pea and fenugreek with grain yields of  $1.6$ ,  $1.4$ ,  $1.3$ ,  $1.3$ ,  $1.3$ ,  $1.2$ ,  $1.1$ ,  $1$  and  $0.9 \text{ t ha}^{-1}$ , respectively.

In Medebay-Zana, maize yielded the highest with  $2.1 \text{ t ha}^{-1}$  followed by sorghum, finger millet, faba bean, barley, tef, chickpea and grass pea with  $1.9$ ,  $1.5$ ,  $1.3$ ,  $1.2$ ,  $1$ ,  $0.9$  and  $0.8 \text{ t ha}^{-1}$ , respectively. In Ahferom, maize was reported to be the highest yielder with  $1.8 \text{ t ha}^{-1}$  followed by sorghum, wheat, finger millet, faba bean, tef and chickpea with yields of  $1.5$ ,  $1.4$ ,  $1.4$ ,  $1.1$ ,  $0.9$

and 0.4 t ha<sup>-1</sup>, respectively. In Alamata, sorghum was the highest grain yielder at 2.0 t ha<sup>-1</sup> followed by maize, barley and tef with respective yields of 1.9, 1.0 and 0.8 t ha<sup>-1</sup> (Table 2.5).

Table 2.5 Percentage of households growing different crops, with their corresponding productivity (t ha<sup>-1</sup>), across four districts of the Tigray Region in northern Ethiopia during the 2013 cropping season.

Crop	Districts							
	Laelay-Maichew		Medebay-Zana		Ahferom		Alamata	
	Respondents (%)	Yield (t ha <sup>-1</sup> )	Respondents (%)	Yield (t ha <sup>-1</sup> )	Respondents (%)	Yield (t ha <sup>-1</sup> )	Respondents (%)	Yield (t ha <sup>-1</sup> )
Tef	95.0	1.1	100.0	1.0	100.0	0.9	98.3	0.8
Sorghum	1.7	1.4	8.3	1.9	6.7	1.5	86.7	2.0
Wheat	56.7	1.7	-	-	60.0	1.4	-	-
Barley	35.0	1.3	35.0	1.2	6.7	1.0	3.3	1.0
Finger millet	18.3	1.6	60.0	1.5	38.3	1.4	-	-
Maize	20.0	1.9	83.3	2.1	13.3	1.8	53.3	1.9
Fababean	30.0	1.3	38.3	1.3	11.7	1.1	-	-
Field pea	3.3	1.0	-	-	-	-	-	-
Chickpea	16.7	1.2	18.3	0.9	1.7	0.4	-	-
Fenugreek	5.0	0.9	-	-	-	-	-	-
Grass pea	21.7	1.3	8.3	0.8	-	-	-	-

Some households grow multiple crops leading percentage of respondents >100%.

Dash (-) indicates zero response

#### 2.3.4 Relative importance of tef

For all uses except animal feed and crop rotation, there were significant differences among districts for the relative importance of tef (Table 2.6). Thus, for all districts, ≥ 80.0% of respondents indicated the high importance (HI) of the crop for animal feed. For crop rotation, differences in importance were not well-defined, hence the lack of significance; for all districts it was ranked by 40-61.7% of the respondents as of low importance (LI).

For food, HI was ≥ 90.0% in all districts except Alamata (65%) (Table 2.6). For income generation, in Ahferom district HI was 96.7%; for the other districts HI was between 76.7 and 80.0%. Differences in importance between districts for drought, water-logging, pest and disease tolerance varied. For drought tolerance, 53.3% of respondents in Alamata indicated high importance; for the other districts HI was ≤ 26.7% and in Medebay-Zana it was only 10.0%. Both Alamata and Ahferom rated water logging tolerance of low importance (LI = 71.7% and 75.0%, respectively). For pest tolerance, 83.3% indicated low importance in Medebay-Zana whereas for other districts LI was ≤ 41.7%. For disease tolerance, HI varied between 43.3 and 70.0% (Table 2.6).

Table 2.6 Proportion of respondents (%) that rated the relative importance of tef for different uses as high, medium or low, in four districts of Tigray region in northern Ethiopia

Importance	Districts												Kruskal–Wallis test
	Laelay-Maichew			Medebay-Zana			Ahferom			Alamata			
	HI (%)	MI (%)	LI (%)	HI (%)	MI (%)	LI (%)	HI (%)	MI (%)	LI (%)	HI (%)	MI (%)	LI (%)	
Food	100.0	-	-	91.7	8.3	-	90.0	10.0	-	65.0	35.0	-	0.000**
Animal feed	90.0	10.0	-	90.0	10.0	-	80.0	20.0	-	88.3	11.70	-	0.302ns
Income generation	76.7	23.3	-	76.7	23.3	-	96.7	3.3	-	80.0	20.00	-	0.010*
Drought tolerance	26.7	36.7	36.7	10.0	25.0	65.0	23.3	68.3	-	53.3	28.30	18.30	0.000**
Water logging tolerance	28.3	25.0	46.7	38.3	30.0	31.7	10.0	15.0	75.0	-	28.30	71.70	0.000**
Pest tolerance	31.7	35.0	33.3	-	16.7	83.3	10.0	41.7	48.3	33.3	33.30	33.30	0.000**
Disease tolerance	43.3	26.7	30.0	56.7	33.3	10.0	70.0	30.0	-	63.3	25.00	11.70	0.001**
Early maturity	50.0	31.7	18.3	65.0	30.0	5.0	68.3	31.7	-	91.7	8.30	-	0.000**
Yield	43.3	33.3	23.3	25.0	40.0	35.0	23.3	28.3	48.3	15.0	36.70	48.30	0.002**
Adaptation to environment	81.7	15.0	3.3	66.7	33.3	-	70.0	30.0	-	56.7	20.00	23.30	0.006*
Crop rotation	21.7	38.3	40.0	28.3	30.0	41.7	26.7	31.7	41.7	18.3	20.00	61.70	0.110ns

HI = High importance; MI = Medium importance; LI = Low importance

HI + MI + LI = 100% of the respondents

\* and \*\* denote significant differences at  $P < 0.5$  and  $P < 0.01$  probability levels, respectively; ns=non-significant difference.

Dash (-) indicates zero response

Alamata indicated very high importance (HI = 91.7%) for early maturity; for the other districts HI ranged between 50.0% and 68.3%. However for yield, HI = 15.0% in Alamata, in the other districts, HI = 23.3– 43.3%. For better adaptability of tef, HI varied between 56.7% in Alamata and 81.7% in Laelay-Maichew (Table 2.6).

### **2.3.5 Landraces of tef grown in the study areas**

Table 2.7 presents current and historic tef landraces and improved varieties grown by farmers during and prior to 2013. Farmers identified the following landraces: Taf-Shewa, Birgida and Zezew in Lealay-Maichew; Taf-Mussie and Zezew at Medebay-Zana; Sergen and Saeda-Taf in Ahferom and Magna in Alamata. These landraces are known for their high yields under optimum environments, high market prices, white seed colour, relative lodging and shattering resistance, and high straw yields across the study areas. In addition, farmers listed other important landraces including Halengay in Laelay-Maichew; Medebay, Barkay, Keyih-Taf in Medebay-Zana; Keyih-Taf in Ahferom and Tikure in Alamata, which are known for their intermediate maturity, high yield levels and brown seed colour (Table 2.7). Landraces such as Zagure, Wafey, Gerima in Laelay-Maichew and Medebay-Zana; Wedikonanit in Medebay-Zana; Goyayito, Wafey at Ahferom and Buni in Alamata were described as having early maturity, brown seed color, better yield potential in moisture stressed and low fertile soils, and possession of palatable straw for cattle.

Table 2.7 Tef landraces and modern varieties grown in the study areas listed by farmers with their desirable and non-desirable traits.

Varieties	Districts				Characteristics	
	Laelay-Maichew	Medebay-Zana	Ahferom	Alamata	Desirable	Undesirable
Landraces	Taf-Shewa Birgida Zezew	Taf-Mussie Zezew	Sergen Seada-Taf	Magna	High yield in optimum season, high straw yield, good <i>injera</i> quality, high market price, tolerant to waterlogging, lodging and shattering resistant	Late maturity, low yield under moisture stress
	Halengay	Medebay Barkay Keyih-Taf	Keyih-Taf	Tikure	High grain and straw yield, good <i>injera</i> quality, intermediate maturity	poor market price
	Zagure Wafey Gerima	Wedikonanit Zagure Wafey Gerima	Goyayito Wafey	Buni	Early maturity, high grain yield during moisture stress, palatable straw yield, preferred <i>injera</i> , good yield in low fertile soil	Low yield, lodging susceptible poor market price
Improved Varieties	Quncho DZ-01-974	Quncho DZ-01-974	Quncho	Quncho	High yield in optimum rainfall, lodging tolerant, shattering resistant, high market price	Low yield in moisture stressed season, late maturity
	-	DZ-Cr-37	DZ-Cr-37	DZ-Cr-37	high yield under moisture stress, early maturity, high market price	Lodging susceptible, Easily shattering

### 2.3.6 Farmers-preferred traits of tef under drought prone environments

Table 2.8 summarises farmer-preferred traits of tef across four drought-prone districts of Tigray Region. Farmers in all the districts described that high grain yield is the first preferred trait, followed by a long panicle length (Table 2.8). The third preferred trait was high straw yield in Laelay-Maichew, Medebay-Zana and Ahferom districts. In Alamata, early maturity was the third and straw yield the fourth preferred trait. Seed color is the fifth preferred trait in all the study districts, except in Alamata where it ranked sixth.

Table 2.8 Farmer-preferred traits of tef with their mean scores across four drought-prone districts of Tigray Region of northern Ethiopia.

Traits	Laelay-Maichew		Medebay-Zana		Ahferom		Alamata	
	Mean Score	Rank	Mean Score	Rank	Mean Score	Rank	Mean Score	Rank
Grain yield	1.0	1	1.0	1	1.1	1	1.5	1
Straw yield	3.2	3	3.9	3	2.9	3	4.0	4
Plant height	6.2	5	7.8	9	6.8	8	8.0	8
Panicle length	2.2	2	3.0	2	2.7	2	3.3	2
Disease and pest tolerance	7.2	9	8.0	8	8.3	9	8.2	9
Drought tolerance	7.1	8	6.5	7	5.7	6	4.1	5
Earliness	6.2	5	4.4	5	5.1	5	3.6	3
Seed colour	4.7	4	4.1	4	4.3	4	5.0	6
Lodging resistance	7.0	7	6.2	6	6.2	7	7.4	7

Ranks are based on 1-9 scores, 1 shows the highest and 9 the least preference.

### 2.3.7 Uses of fertilizers and improved varieties

In Laelay-Maichew, Medebay-Zana, Ahferom and Alamata, 33.3%, 66.7%, 71.7% and 71.2% of respondents, respectively, indicated that their land allocated to tef is poor in fertility; conversely, only a low percentage, 15.8%, 11.7%, 5.0% and 11.9%, respectively, had land of high fertility (Table 2.9).

Table 2.9 Proportion of respondents (%) that rated their tef land fertility as high, medium or low in four districts of Tigray region of northern Ethiopia

Districts	Sample	Land fertility (%)			Total
		High	Medium	Low	
Laelay-Maichew	57	15.8	50.9	33.3	100
Medebay-Zana	60	11.7	21.7	66.7	100
Ahferom	60	5.0	23.3	71.7	100
Alamata	59	11.9	16.9	71.2	100

For inorganic fertilisers,  $\geq 95\%$  of respondents in Laelay-Maichew, Medebay-Zana and Ahferom districts indicated their use; in Alamata, 61.7% indicated use of both di-ammonium phosphate (DAP) and urea (Table 2.10). In Ahferom, 45.0% and 33.3% used animal manure and compost, respectively. In the other districts, use of organic fertilizers was  $\leq 25.0\%$  (Table 2.10).

The mean amounts of urea applied were 94.6, 102.2, 101.2 and 87.3 kg ha<sup>-1</sup> for Laelay-Maichew and Medebay-Zana, Ahferom and Alamata, respectively. Similarly, in the same order of the districts, 90.9, 102.2, 99.6 and 90.0 kg ha<sup>-1</sup> DAP fertilizers were applied. There was greater variation in the mean amounts of organic animal manure (1053-2028 kg ha<sup>-1</sup>) and compost (820-2966 kg ha<sup>-1</sup>) applied; the highest amounts were in Ahferom and Alamata

districts. However, for all fertilizers, the standard deviations indicate that the variation between respondents within all districts was large (Table 2.10).

Table 2.10 Type and amount (kg ha<sup>-1</sup>) of inorganic and organic fertilizers and corresponding proportions of respondents who applied these for tef production in the four study districts of Tigray Region in northern Ethiopia during the 2013 cropping Season

Fertilizers	Statistics	Districts			
		Laelay-Maichew	Medebay-Zana	Ahferom	Alamata
Urea	% of respondents	95.0	100.0	100.0	61.7
	Mean (kg ha <sup>-1</sup> )	94.6	102.2	101.2	87.3
	Standard deviation (SD)	32.7	20.3	39.0	31.8
DAP	% of respondents	95.0	100.0	100.0	61.7
	Mean	90.9	102.2	99.6	90.0
	Standard deviation (SD)	28.3	20.3	39.4	32.8
Animal manure	% of respondents	16.7	8.3	45.0	11.7
	Mean (kg ha <sup>-1</sup> )	1053	1563	2103	2029
	Standard deviation (SD)	465	687	656	1830
Compost	% of respondents	25.0	15.0	33.3	5.0
	Mean (kg ha <sup>-1</sup> )	1578	820	2056	2966
	Standard Deviation (SD)	1439	2422	656	2793

Standard deviations and means are based on N=60

The majority, 85-96.7%, of respondents in all districts had adopted the use of one or more of three improved varieties, DZ-Cr-387 (Quncho), DZ-01-974 and DZ-Cr-37. The most common ( $\geq 80.0\%$ ) in all districts except Alamata was DZ-Cr-387 (Quncho). In Alamata, only 30% of respondents grew DZ-Cr-387 (Quncho) because of its late maturity and need for high rainfall, and DZ-Cr-37 was the preferred variety (Table 2.11). As a consequence, landrace varieties were less commonly grown, and across districts varied between 29.0%-55.0% of respondents.

Table 2.11 Percentage of respondents who grow landraces and improved tef varieties across four districts of Tigray Region in northern Ethiopia during the 2013 cropping season

Districts	Varieties and proportion (%) of respondents		Improved varieties and proportion (%) of respondents		
	Landraces	Improved varieties	DZ-Cr-387 (Quncho)	DZ-01-974	DZ-Cr-37
Laelay-Maichew	30.0	88.3	85.0	5.0	-
Medebay-Zana	29.0	96.7	80.0	20.0	36.7
Ahferom	55.0	96.7	86.7	-	23.3
Alamata	32.0	90.0	30.0	-	85.0

Dash (-) indicates zero response

### 2.3.8 Seed system

The majority, 69.0%-94.3%, of respondents accessed improved varieties through farmers' cooperatives. The second most common source of improved seed across districts was farm-saved, and varied between 7.4%-13.8%; however no farm-saved seed was used in Laelay-Maichew. In Ahferom and Alamata; 17.2% and 16.7% of the respondents, respectively, used seeds of improved varieties from other farmers. For landrace seed,  $\geq 53.1\%$  of respondents in all districts used farm-saved seed (Table 2.12).

Table 2.12 Seed sources of tef and corresponding proportion of respondents across four districts of Tigray region in northern Ethiopia during the 2013 cropping season

Seed sources	Districts							
	Laelay-Maichew		Medebay-Zana		Ahferom		Alamata	
	No.	%	No.	%	No.	%	No.	%
<b>Improved varieties</b>								
Farmers' cooperative	50.0	94.30	51.0	87.9	40.0	69.0	41.0	75.9
Research centres	3.0	5.70	-	-	-	-	-	-
Farm saved	-	-	7.0	12.1	8.0	13.8	4.0	7.4
Neighbouring farmers	-	-	-	-	10.0	17.2	9.0	16.7
<b>Landraces (local varieties)</b>								
Local market	6.0	20.0	6.0	20.7	6.00	18.2	8.0	25.0
Farm saved	17.0	56.7	18.0	62.1	21.00	63.6	17.0	53.1
Neighbouring farmers	7.0	23.3	5.0	17.2	6.00	18.2	7.0	21.9

### 2.3.9 Cropping patterns and cultural management

In Alamata, Ahferom, Laelay-Maichew and Medebay-Zana, 46.7%, 96.7%, 100% and 93.3% of respondents, respectively, used hand weeding at least two times, whereas 53.3%, 3.3% and 6.7% used both hand weeding and the herbicide 2-4-D in Alamata, Ahferom and Medebay-Zana, respectively. More than half of the respondents in Alamata used both herbicide and hand weeding (Table 2.13).

In all the study districts except in Laelay-Maichew, tef was the sole crop planted on a given piece of land (monoculture); 15.0% of respondents in Laelay-Maichew mixed tef with oil crops such as safflower and mustard. More than 60% of the respondents in all districts practised crop rotation in tef production. The percentage of respondents who use crop rotation was highest (91.7%) in Laelay-Maichew and lowest in Alamata (63.3%) (Table 2.13).

Table 2.13 Common crop management practices in tef production across four districts of Tigray region in northern Ethiopia during 2013 cropping season

Districts	Crop Management						
	Weeding methods			Planting pattern		Crop rotation	
	Hand weeding <sup>#</sup>	Chemical	Hand weeding and chemical	Sole	Sole and mixed	Yes	No
Alamata	46.7	-	53.3	100.0	-	63.3	36.7
Ahferom	96.7	-	3.3	100.0	-	88.3	11.7
Laelay-Maichew	100.0	-	-	85.0	15.0	91.7	8.3
Medebay-Zana	93.3	-	6.7	100.0	-	90.0	10.0

<sup>#</sup>at least twice in a season

### 2.3.10 Planting methods

Respondents indicated that transplanting resulted in highest grain yield, seed weight and flour productivity, least seed cost, reduced lodging, reduced moisture stress, most responsive to fertilizer application and easy weed control. Row sowing was also indicated to be good for increased tef productivity but not as good as transplanting (Table 2.14). Even though transplanting improved yield and other desirable traits, its high labour cost and difficulty of application were among the main drawbacks limiting its acceptance by respondents.

Table 2.14 Perception of respondents on benefits and drawbacks of different tef planting methods

Planting methods	Benefits	Drawbacks
Transplanting	High grain yield Highest seed weight, flour productivity and straw yield Better stack thickness and strength Increased plant height, panicle length and tiller capacity Reduced lodging Least seed cost Reduced moisture stress Responsive to fertilizer application Easy weed control	Very high labour cost Complex and difficult to apply
Row sowing	High grain yield Higher seed weight, flour productivity and straw yield than broadcasting Reduced seed cost Reduced lodging Easy cultivation and weed control	Higher labour cost than broadcast sowing
Broadcasting	Easy application Low labour cost	Difficult to control plant population High seed cost High susceptibility to lodging Low yield

Average tef yields from transplanting, row sowing and broadcasting were, respectively, 2.1, 1.3 and 0.9 t ha<sup>-1</sup>. However, the production cost of transplanting was two times that of row sowing

and almost three times that of broadcasting. As a result, the net profit was highest from row planting (Table 2.15).

Table 2.15 Cost benefit analysis of different planting methods in tef

Planting method	Grain yield (t ha <sup>-1</sup> )	Income (grain) (ETB ha <sup>-1</sup> )	Income (straw) (ETB ha <sup>-1</sup> )	Total income (ETB ha <sup>-1</sup> )	Total production cost (ETB ha <sup>-1</sup> )	Profit (ETB ha <sup>-1</sup> )	Cost-benefit ratio
Transplanting	2.1	37800	3113	40913	30039	10874	1.4
Row sowing	1.3	23400	3067	26467	15013	11454	1.8
Broadcasting	0.9	16200	2790	18990	11732	7258	1.6

### 2.3.11 Constraints to tef production

The Kruskal-Wallis tests showed that the relative severity of the constraints varied among districts (Table 2.16). In Laelay-Maichew, the majority of the interviewees' identified lodging as the major production constraint followed by high cost of fertilizers, weeds, poor soil fertility and pest (shoot fly) in that diminishing order. Only 33.3% and 18.3% of respondents in Laelay-Maichew and Medebay-Zana responded that tef is a low yielder as compared to other crops, while in Ahferom 46.7% and in Alamata 41.70% of the interviewed farmers responded tef is a low yielding crop. In Medebay-Zana 70%, 68.3% and 58.3% of the respondents suggested, that a pest (shoot fly), moisture stress and weeds were the most severe tef production constraints. In Ahferom 73.3%, 71.7%, 60% and 51.7% of the respondents in that order described weeds, moisture stress, lodging and poor soil fertility as most severe production problems (Table 2.16). In Alamata 86.7%, 63.3%, 58.3% and 53.3% of the interviewees indicated that respectively moisture stress, weeds, shoot fly and lodging were the most severe constraints to tef production. As indicated on Table 2.16, more than 50% of the respondents in all the districts except in Laelay-Maichew claimed moisture stress and shattering to be severe tef production constraints. In all the study districts, diseases and input shortage are tef production problems though with relatively low importance (Table 2.16). Leaf rust is the only reported tef disease with minor occurrence, but under epidemic conditions it may cause high yield losses as reported by farmers (Table 2.16).

Table 2.16 Major constraints to tef production across four Districts of Tigray Region in northern Ethiopia during the 2013 cropping season

Constraints	Districts												Kruskal - Wallis test
	Laelay-Maichew			Medebay-Zana			Ahferom			Alamata			
	Severity			Severity			Severity			Severity			
	HS (%)	MS (%)	LS (%)	HS (%)	MS (%)	LS (%)	HS (%)	MS (%)	LS (%)	HS (%)	MS (%)	LS (%)	
Poor soil fertility	50.0	28.3	21.7	28.3	48.3	23.3	51.7	28.3	20.0	21.7	46.7	31.7	0.057ns
Low yield	33.3	33.3	33.3	18.3	40.0	41.7	46.7	21.7	31.7	41.7	25.0	33.3	0.028*
Inputs shortage	-	-	100.0	15.0	50.0	80.0	-	3.3	96.7	-	-	100.0	0.000**
Lodging	66.7	20.0	-	43.3	33.3	23.3	60.0	40.0	-	53.3	26.7	20.0	0.000**
High cost of fertilizers	55.0	26.7	18.3	41.7	50.0	8.3	45.0	26.7	28.3	45.0	30.0	25.0	0.56ns
Moisture stress	33.3	38.3	28.3	68.3	31.7	-	71.7	28.3	-	86.7	13.3	-	0.000**
Pests (short fly)	50.0	26.7	23.3	70.0	30.0	-	45.0	41.7	13.3	58.3	28.3	13.3	0.000**
Disease (leaf rust)	35.0	25.0	40.0	23.3	36.7	40.0	46.7	25.0	28.3	36.7	33.3	30.0	0.076ns
Weeds	55.0	25.0	20.0	58.3	28.3	13.3	73.3	13.3	13.3	63.3	18.3	18.3	0.236ns
Seed shattering	66.7	20.0	13.3	55.0	25.0	20.0	56.7	26.7	16.7	65.0	21.7	13.3	0.448ns

HS = highly severe; MS = moderately severe; LS = Less severe

\*and \*\* denote significant differences at P<0.5 and P<0.01 probability levels, respectively; ns=non-significant difference

Dash (-) indicates zero response

### 2.3.12 Moisture stress and its effect on growth and productivity of tef

In the present study areas, moisture stress in all the districts was severe at the grain filling stage (Table 2.17). In Laelay-Maichew, Medebay-Zana, Ahferom and Alamata in that order 46.7%, 35%, 31.7% and 36.7% of the farmers believed that the occurrence of moisture stress during grain filling stage of tef, severely affected yields (Table 2.17). About 26.7%, 36.7%, 25% and 26.7% of the respondents in Laelay-Maichew, Medebay-Zana, Ahferom and Alamata, respectively, noted that moisture stress is critical both during crop emergence and grain

Table 2.17 The relative importance (% of farmers reporting) of moisture stress at different tef growth stages as reported by farmers in tef production across four districts of Tigray Region in northern Ethiopia.

Stage of moisture stress	Districts			
	Laelay-Maichew	Medebay-Zana	Ahferom	Alamata
At emergence	1.7	13.3	18.3	13.3
At heading	20.0	11.7	13.3	15.0
Grain filling	46.7	35.0	31.7	36.7
At emergence and heading	5.0	3.3	11.7	8.3
At emergence and grain filling	26.7	36.7	25.0	26.7

Table 2.18 summarises the effect of different moisture regimes on tef yields in the four studied districts. Yield reductions due to moisture stress were 36.7, 49.0, 48.2% and 60.3%, in Ahferom, Alamata, Laelay-Maichew and Medebay-Zana, respectively (Table 2.18).

Table 2.18 Yield at optimum rainfall and moisture stress condition and percentage of yield reduction due to moisture stress in tef in four study districts during the 2013 cropping season.

Districts	Yield (t ha <sup>-1</sup> )		Yield reduction (%)
	Optimum rainfall	Moisture stress	
Laelay-Maichew	1.3	0.81	36.7
Medebay-Zana	1.1	0.57	49.0
Ahferom	1.0	0.50	48.2
Alamata	1.18	0.47	60.3

### 2.3.13 Farmers' coping mechanisms against moisture stress in tef production

Table 2.19 highlights the coping mechanisms adopted by tef farmers in the study areas. In all the study districts, more than 75% of the respondents valued the use of early maturing varieties during moisture stress seasons. About 41, 43, 48 and 66.7% of farmers adopted soil and water conservation in Laelay-Maichew, Medebay-Zana, Ahferom and Alamata respectively.

Table 2.19 Coping mechanisms (% of farmers reporting) of moisture stress in tef production across four districts of Tigray Region in northern Ethiopia.

Coping mechanisms	Districts			
	Laelay-Maichew	Medebay-Zana	Ahferom	Alamata
Growing early maturity tef varieties	81.7	86.7	76.7	80.0
Use of irrigation	8.3	6.7	5.0	28.3
Replacing tef by other crops	0.0	0.0	0.0	0.0
Soil and water conservation	41.7	43.3	48.3	66.7

## 2.4 Discussion

### 2.4.1 Demographic characteristics of households

The average family size of respondents of the study districts ranged from 5.5 to 6.6. Family size has implication on the farming system and food security attainment. Tefera (2009) and Kahsay and Mulugeta (2014) mentioned that large family size is one of the factors that contribute to food insecurity. Most of the land size in Tigray region is less than 1 ha, which makes difficult to meet food and other necessities of large families. The majority of the respondents in Laelay-Maichew and Medebay-Zana completed elementary education while in Ahferom and Alamata were illiterate. The education level of household head is a determinant in affecting family livelihood, as the one with higher education level seeks more for better livelihood through adoption of modern technologies and farming practices. Mean ages of the respondents ranged from 41.1 to 50.8 years. Household age affects adoption of agricultural technologies, in which most of the time the young aged households are fast adopters (Yuet al., 2011). While, in the case of genetic diversity conservation the elder's prefers to preserve their landraces and were also best source of information on indigenous farming practices from their long term experiences.

### 2.4.2 Land ownership systems of households

The mean land size a household was within a range of 0.7-1 ha. Previous studies also reported an average land size of less than 0.9 ha in Tigray region (Nega et al., 2003; Headey et al.,

2014). Of the respondents, 38-55% shared-in land in addition to their land holdings. Share cropping is a system of agriculture in which a land owner allows a tenant to use the land in return for a share of the crops produced from the land. In Laelay-Maichew and Alamata districts about 1.7% and 3.3% of respondents, respectively, did not own land. These households grow crops by sharing-in land from other farmers. Due to the ever increasing population size, farm land is becoming scarce in Tigray region, as a result now days the young households are not provided with land and are forced to fulfil their family food needs though cultivating crops by sharing-in land from those who could not to cultivate their farm such as elders and female households.

### 2.4.3 Farming systems

#### 2.4.3.1 *Main types of crops grown: area coverage and productivity of tef*

Farmers were interviewed about the types of crops they grew and the area of land allocated to each crop in the 2013 cropping season to collate information on the relative importance of each crop. In all the studied districts more than 50% of the households farm land was covered by tef. Similarly, Ferede (2011) reported 52% share of tef of the total cultivated land in major tef growing areas of central Ethiopia. In Alamata district sorghum used to be the major crop, but in recent years it has been replaced by tef for its early maturity during moisture stress seasons and better yield stability.

The proportion of farmers who cultivated a crop determines its relative importance. More than 95% of the respondents cropped tef in 2013 growing season. This indicates the relative importance of tef for the households, food security and income generation. The mean grain yields of tef were from 0.8 to 1.1 t ha<sup>-1</sup>. These figures suggested that the productivity of tef in the study areas is relatively low compared to the national mean yield of 1.58 t ha<sup>-1</sup> (CSA, 2015). Farmers in all the study areas pointed out that crop productivity was reduced by moisture stress during the 2013 cropping season. Tef was a lower grain yielder than any of the other cereals. However, it is cultivated widely for its high market price, both for the grain and the straw as well as it's suitability for grain production during both the main and short rainy seasons. Besides, tef has got the relative advantages of resilience under marginal conditions, preference for consumption, and high flour and *injera* returns in utilization as food (Assefa et al., 2011a, Assefa et al., 2015).

#### 2.4.4 Relative importance of tef

More than 80% of the respondents indicated the importance of tef for food and feed. *Injera* is the daily traditional food of Ethiopia and can be prepared from tef, sorghum, wheat, barley, maize, finger millet and rice (Abiyu et al., 2013). However, injera made from tef is the most palatable and the preferred choice of Ethiopian people. Tef has higher flour productivity than the other crops (Ketema, 1997), and tef straw is more palatable to animals than straw from barley and wheat (Gebrehiwot and Mohammed, 1989). The majority of farmers in Ethiopia are small-holders who use mixed-farming systems. Food and feed source are therefore major criteria in crop selection, so tef is chosen for its quality and palatability for both human and animal consumption.

Farmers grow tef as a cash crop, and it fetches high market prices (Tefera and Ketema, 2001). Although there were significant differences, >76% of respondents in all districts rated its importance for income generation as high. Nationally, tef is second to coffee as a cash crop (Minten et al., 2013). Also, although tef has a lower productivity, it commands a higher price/kg than wheat, sorghum, maize and barley (10.2 ETB/kg v. 6.3, 7.2, 4.5 and 6.7 ETB/kg, respectively) (Tefera, 2012). Because of its high price, tef is affordable to the majority the middle- and upper-class families in Ethiopia; an added attraction for farmers is a growing export market to North America, Europe, Asia and the Middle East (Reda, 2015).

A majority of respondents in Alamata which has low and bi-modal rainfall, but not the other districts which have mono-modal rainfall, indicated the high importance of tef for drought tolerance. However, tef is still considered sensitive to moisture stress during its early and middle stages of growth ( Tefera et al., 2000; Yihun et al., 2013), but compared to other crops is drought tolerant (Ketema, 1997; Ginbot and Farrant, 2011; Assefa et al., 2011a; Plaza-Wuthrich et al., 2013). Maturity period also contributes to the drought tolerance of crop plants (Blum, 2011), and while all districts recognised the advantage of early maturity in tef, this was most important in Alamata. Tef is a versatile crop, as it grows in diverse environments and throughout Ethiopia under rain-fed conditions (Ketema, 1997; Tesema, 2013; Anteneh et al., 2014; Assefa et al., 2015; Reda, 2015). However, higher rainfall variability and lower mean annual rainfall affect tef productivity (Gebrehiwot et al., 2011; Hadgu et al., 2013). To meet this challenge, the development of early maturing but high-yielding drought tolerant varieties is required.

Although tef has better waterlogging tolerance than other cereals such as wheat and maize (Assefa et al., 2015), the majority of respondents in all districts indicated this was of medium or

low importance. This was particularly the case in Ahferom and Alamata, although all districts are considered semi-arid with rainfall amounts that are unlikely to generate frequent waterlogging events.

Disease tolerance was considered of higher importance than tolerance to insect pests in all studied districts. Tef is generally considered to have better disease resistance than other cereals (Ketema, 1997; Assefa et al., 2011a; Assefa et al., 2015). Shoot fly is an insect pest in Tigray region, though not in most other major tef growing parts of Ethiopia (Damte, 2013); in Tigray it is being effectively controlled using a chemical spray known as Karate (Cyhalothrin)(AxARC, 2011). The effectiveness of this control measure may have been the reason such a high proportion of respondents in Medebay-Zana considered resistance to insect pests as being of low importance.

#### **2.4.5 Landraces of tef grown in the study areas**

Ethiopia is the centre of diversity and origin of tef (Vavilov, 1951). Landraces are often better referred to as farmer varieties and are often excellent sources of genetic variation with desirable traits for breeding (Aremu, 2011). Furthermore, landraces have better adaptation and yield stability in marginal and stressed environments. Therefore, the present study explored historic and present day tef landraces being grown by farmers in the study areas. Accordingly, farmers in all the study areas were asked to describe the name of tef varieties that have been cultivated during and before 2013.

Farmers identified different kinds of landraces which differs for their maturity (early, intermediate and late), seed colour (white, white and brown mixed and brown), grain yield, biomass yield and resistance to lodging. Respondents indicated that, Majority of the early maturing landraces, are brown seeded with low grain yield, low biomass yield and highly susceptible to lodging. On the other hand the majority of the late maturing landraces are white seeded with high grain yield, high biomass yield and relatively lodging tolerant. In terms of market preference also the white seed colour fetches high price, while in terms of adaptation to harsh environments such as moisture stress and poor soil fertility the brown seeded landraces are preferred. Previous studies have also identified tef landraces with their respective uses grown in other parts of Ethiopia (Anteneh and Mekbib, 2013; Anteneh et al., 2014).

The majority of the respondents mentioned that the landraces are being replaced by high yielding improved varieties such as DZ-01-974 and DZ-Cr-387 (Quncho) in the optimum

environments and DZ-Cr-37 in the moisture stressed environments, which has significantly reduced the genetic diversity of the landraces in these study areas. This suggests the need for strategic conservation of tef landraces for future breeding. Further evaluation of the landraces identified in this study for targeted traits would be essential to improve productivity and yield stability of tef in the region, without creating excessive loss of genetic diversity.

#### 2.4.6 Farmers-preferred traits of tef under drought-prone environments

Tef is the most preferred cereal grain in Ethiopia to prepare quality flat bread locally known as '*injera*', an esteemed daily food for millions of people in Ethiopia. Thus far, *injera* is the only major food product being prepared from tef flour in the country. Presently, the cultivation of tef as a cereal has been started in the USA, the Netherlands and Israel (Assefa et al., 2011a). In these countries, tef flour is processed to make various products such as breakfast cereal, porridge, bread, pancake, waffles and juices.

Depending on their farming systems, growers select a crop variety using multiple criteria. Understanding farmers' variety preferences is fundamental to breeders in order to identify important traits to be considered in their breeding program. High grain yield followed by long panicle and straw yield were the first, second and third preferred traits respectively in all the districts except in Alamata where early maturity was the third preferred trait. High straw yield is valued because straw is a key source of feed for livestock. Farmers in all districts practice mixed farming system (crop-livestock production) where palatable tef straw is the main feed source for animals (Yami, 2013).

Seed colour was also farmers most referred trait. White seeded tef grain fetches a higher market price at all times than brown and brown and white mixture grain. White seeded tef is preferred by consumers in Ethiopia for its better *injera* making quality (personal observation), but brown seeded tef is reported to be better in nutritional quality such as high iron content (Baye, 2014). Minten et al., (2013) found that, in the past, farmers demanded brown coloured tef seed for its cheap price and affordability. Recently, white seeded varieties are preferred by farmers due to the high market price of white grain. Fentie et al., (2012) reported that, farmer's trait preferences in the Amhara region were grain yield, maturity date and seed color, in that order, and this is similar to the results of this study. In the present study, disease and pest resistance were not important traits, because tef is less susceptible to pests and disease than other crops (Damte, 2013).

### 2.4.7 Inputs

Most agricultural land of Tigray Region is undulating with slopes that are associated with high soil erosion. Haileselassie et al. (2011) reported low soil fertility as the major constraint on crop production, especially in the highlands of Tigray. The agricultural soils in Tigray region are deficient of the major nutrients nitrogen, potassium and calcium and some micronutrients (Kebede and Yamoah, 2009). In this study, most of the land allocated to tef in three districts was of low soil fertility; in Laelay-Maichew, half the soils were of medium fertility. In Alamata district respondents allocated the fertile land to sorghum and the less fertile to tef production because tef is better adapted to less fertile soils. The fertility status of agricultural land determines the production inputs and subsequent management practices that are required to optimise the yield of tef. The medium to low fertility of the soils used suggests that application of fertilizer will lead to higher yields.

Most (95%) or all respondents in Laelay-Maichew, Medebay-Zana and Ahferom districts used chemical fertilisers. In Alamata, where only 60% of respondents used chemical fertilisers, the reason stated was that while fertilizer application initially increased vegetative growth, the full benefits of this input were not realized because of the lack of soil moisture during the late growth period. Under severe moisture stress conditions, fertilizer application may also 'burn' their crop and result in reduced yields. The potential for this to occur suggests that promotion of moisture conservation techniques are essential to the adoption of chemical fertilizer inputs where periods of moisture stress are part of the growth cycle (Yesuf et al., 2009).

In all the study districts, the Bureau of Agriculture and Rural Development (BoARD) recommend 100 kg ha<sup>-1</sup> each of urea and DAP for tef. This was practised by farmers in all study districts except in Alamata where only 60% of respondents used these fertilizers. A recent manual developed by BoARD and the Agricultural Transformation Agency (ATA) recommends a combined application of 80 and 100 kg ha<sup>-1</sup> of urea and DAP fertilizers, respectively for all clay soils (Ayele et al., 2012). However, as soil fertility status varies across all districts (Table 2.9), localized soil fertility tests that lead to specific fertilizer recommendations should be developed.

Organic fertilizers help improve soil texture and tef yields, especially in less fertile soils (Berhane et al., 2013). These fertilizers are also potentially cost-effective because in mixed-farming systems in particular, they can be prepared from locally available materials. However, except in Ahferom district, composts and animal manure were used by 25% or fewer respondents in tef production. This is because of the high labour demand linked to the preparation and transport of

what amounts to large quantities of material (in this study as much as 3 t/ha was recorded) to the field (Geta et al., 2013). Farmers may also prefer to use weeds and crop residues as sources of animal feed and fuel, rather than sources of organic fertilizer.

The respondents indicated that the use of improved varieties increased steadily in all the study districts, and at the time of the study >88% used at least one of the three common varieties. The variety DZ-Cr-387 (Quncho), which was released in 2006 for its high yield, white seed and relative resistance to lodging (Assefa et al., 2011b), had been widely adopted in Laelay-Maichew, Medebay-Zana and Ahferom. In Alamata, respondents had mainly adopted the variety, DZ-Cr-37, which was released in 1983; this variety is widely grown in areas subject to moisture stress because of its high yield, early maturity and white seed colour (Assefa et al., 2011b). The improved varieties, Gemechis, Simada and Boset, were released in 2007, 2009 and 2012, respectively, for use in such areas (MoA, 2014). Gemechis and Simada have not been widely adopted because of their poor performance compared to DZ-Cr-37; Boset has not yet reached the farmers. Farmers also cultivated landraces, especially in the drier areas of the study districts. Ferede (2011) found that 40% of respondents grew tef landraces in central Ethiopia. These landraces could be a potential source of genes for developing new drought tolerant varieties and should be further evaluated as potential inputs into breeding programs.

#### **2.4.8 Seed system**

Improved seed of high quality and its timely supply is an important determinant of crop productivity. Both formal and informal seed systems operate in Ethiopia. However, the major source of improved seed for tef is the formal seed system (Atilaw and Korbu, 2011) where the Ethiopian Seed Enterprise (ESE) has the mandate to multiply certified seed, and BoARD through its extension services and other non-governmental organizations (NGOs) such as Sasakawa Africa Association (SAA) distribute these seeds to farmers. However, in this study, the majority of the respondents used farmers' cooperatives which have become active distributors of improved seeds. The Integrated Seed Sector Development (ISSD) project (<http://www.issdethiopia.org/index.php/regional-offices/2012-10-29-09-13-27/>) has established a local seed business system for tef in Laelay-Maichew district by supporting the farmers to produce quality seed from improved varieties. These farmers then provide bulked seed to the nearest cooperative. The system is satisfying the local demand for quality seed.

#### 2.4.9 Cropping pattern and cultural management

Numerous grass and broad-leaved weed species competing with tef result in lost tef productivity, especially during establishment (Assefa et al., 2008; Tesfahunegn, 2014) and weeding was required in all districts. Hand weeding was necessary where both grass and broad-leaved weeds were present; in Alamata where the most of the weeds were broad-leaved, chemical control using 2,4-D from local markets was commonly used. As hand-weeding is costly, effective chemical or bio-herbicides to control both grassy and broad-leaved species, are necessary for increasing the profitability of tef production.

In three districts, respondents grew tef as a monoculture; in Laelay-Maichew, a small proportion (15%) inter-cropped tef with safflower (*Carthamus tinctorius L.*) and mustard (*Brassica juncea L.*). Respondents indicated that neither of these oilseed crops affected tef yields. Such inter-cropping, including sesame and sunflower is used elsewhere with tef in Ethiopia (Adamu and Kemelew, 2011). Bayu et al., (2007) reported a yield advantage of 20-77% when tef and sunflower were intercropped compared to a tef monoculture in north-east Ethiopia. As mixed cropping of cereal with N-fixing crops is recommended for improving soil fertility and yield (Malezieux et al., 2009), inter-cropping of tef with the pulse crop, *Vicia faba* (faba bean), may also benefit tef yields (Agegnehu et al., 2006). However, respondents indicated that pulse inter-cropping with tef was not used because the sowing and maturity dates coincide. In contrast, safflower is sown after the tef is well established and matures by taking advantage of the residual moisture after the tef is harvested, in this way contributing to the lack of effect on tef yield.

#### 2.4.10 Planting methods and their application for tef improvement

Tef seed is tiny, so seeding rate is difficult to control during sowing. The crop is also a grass with high susceptibility to lodging which has been estimated to reduce average yields by 17% tef (Ketema, 1993). Transplanting and row sowing are promoted by Ethiopian Agricultural Transformation Agency (ATA) to boost tef productivity by reducing lodging (ATA, 2013); in this study respondents were aware that this was associated with better stack strength, high tillering, greater plant height and spike length, as well as reduced seed cost and moisture stress. Moisture stress usually develops during the grain filling period in Tigray region (Araya and Stroosnijder, 2011). Consequently, early establishment of the crop at the start of the rains maximizes the length of the growing season, and opportunity for the crop to mature before the development of severe moisture stress. However, respondents preferred row sowing to

transplanting as the former resulted in increased yields and it was easier to apply and had a lower labour cost than transplanting.

The cost-benefit analysis showed that transplanting, on average, leads to an additional 62 and 133% yield than row and broadcast sowing, respectively; yield increases of up to 65% have been reported from tef transplanting (Asargew et al., 2014; ATA, 2013; AxARC, 2013; Tesfaye et al., 2015). However, row planting was better in terms of profitability, which added to its first preference by respondents. The potential benefits of transplanting, which could in principle contribute to moving towards food security in Ethiopia, will therefore not be realised until technologies are developed which lower the cost of its implementation.

#### **2.4.11 Constraints to tef production**

The average national tef yield is the lowest ( $1.58 \text{ t ha}^{-1}$ ) of cereal crops cultivated in Ethiopia due to mainly many biotic, abiotic and socio-economic constraints. The effect of these production constraints varies according to the agro-ecological conditions and farming practices. Thus, identifying and prioritizing the constraints will be helpful to devise appropriate improvement strategies.

The priorities of tef production constraints were different for the studied districts, which would indicate the need of different interventions. In Laelay-Maichew, lodging was the major production constraint followed by high cost of fertilizers, weeds, poor soil fertility and pest (shoot fly). In this district, farmers apply fertilizers for tef production and particularly nitrogenous fertilizers lead to crop lodging, which in turn exposes the seed to shattering, insect attack, and poor seed quality. Therefore, it is crucial to improve lodging tolerance of tef in order to make it more responsive to fertilizer application for better productivity and quality. Besides, tackling the lodging problem would help to indirectly reduce the effects of other production problems in tef.

In Medebay-Zana pest (shoot fly), moisture stress and weeds were the most severe tef production constraints. Tef shoot fly is a serious problem in Tigray Region, but not in most other major tef growing parts of Ethiopia (Damte, 2013). In Tigray Region, shoot fly is controlled using chemical spray such as Karate (Cypermethrin).

In Ahferom, weeds, moisture stress, lodging and poor soil fertility were the most severe production problems. In Alamata, moisture stress, weeds, shoot fly and lodging were the most severe constraints to tef production. Kassie (2014) reported that, moisture stress was a major constraint affecting crop production in the central rift valley and the Kobo areas of Ethiopia.

Alamata district borders Kobo area and shares the same agro-ecology, which is also vulnerable to moisture stress.

Seed shattering was also among the major tef production constraints as indicated by more than 50% of the respondents. The majority of early maturing varieties were lodging susceptible along with high shattering problem. Erratic rainfall is the common feature in the region where unexpected rains during maturity and harvest causes seed shattering and heavy yield losses in tef production. Ketema (1997) and Assefa et al. (2011) has also mentioned, shattering as one of the major tef production problems.

The small grain size is another major problem in tef although farmers did not list it as problem because they believed it to be an inherent trait of the crop. Due to the minute seed size of tef, it is not easy to control tef plant population in the field which is a major cause for lodging (Ketema, 1997). Traditionally, farmers sow tef using hand broadcasting with little control on the plant population density and distribution. However, this practice may create problems such as high plant populations and lodging, high cost of seed for planting, impairing efficient use of fertilizers and reduction of yields, among others. Since 2011, the Agricultural Transformation Agency (ATA) of Ethiopia has embarked on demonstration activities such as row planting, transplanting and use of reduced seed rate of tef. These practices have shown promises in alleviating the negative impacts of tef hand broadcasting and boost productivity. Generally, farmers use seed rate of 30 kg ha<sup>-1</sup> or more for broadcast sowing of tef whereas only 5-10 kg ha<sup>-1</sup> seed is needed for row planting (Ayele et al., 2012). Hence, 15-20 kg ha<sup>-1</sup> seed is wasted with broadcast sowing. If tef is space planted it has a high tillering capacity and will develop thicker and strong stems leading to reduced susceptibility to lodging. Space planted tef also produces longer panicles and more spikelet per spike. Space planted tef yielded 29-39%, more than broadcast tef (Asargew et al., 2014). Similarly, about 38% yield increase has been reported from transplanting than broadcasting tef in the central zone of Tigray (AxARC, 2013). Yet, farmers prefer to practice hand broadcasting of tef because of its small seed size for space planting. Transplanting tef is tedious, expensive and labour intensive.

#### **2.4.12 Moisture stress and its effect on growth and productivity of tef**

The impact of moisture stress in affecting crop growth and productivity depends on its severity and the plant growth stage of its occurrence. Information on growth stage of the crop most affected by moisture stress is helpful to devise appropriate breeding strategies and to develop drought tolerant varieties for the targeted production environments. In all the study districts

moisture stress at grain filling stage severely affected grain yield of tef. This stage is critical because grain filling is highly correlated with grain yield (Ayalew et al., 2011). Tef is even more sensitive to moisture stress during its early growth stages than at maturity (Tefera et al., 2000). The interviewees confirmed that predominantly moisture stress occurs in September when tef grain filling occurs in the studied areas. Araya and Stroosnijder (2011) also found that rainfall stopped during September in the southern Tigray Region. Supplemental irrigation during the grain filling stages has reportedly reduced up to 80% yield reduction in this Region. Tef yields were relatively better for Laelay-Maichew under both moisture-stress and non-stress conditions. This is because this district is a more favourable environment for tef production than the other districts in terms of altitude, moisture availability, and soil conditions (Tables 2.1). In Alamata, yields were low during moisture stress condition, but it was better than that in Ahferom when the crop was grown under optimum moisture condition. The effect of moisture stress is more severe and pronounced in Alamata than in other districts.

Over all, above 35% tef yield reductions are recorded due to moisture stress across the study districts. Admass and Belay, 2011 reported that about 25.5% tef grain yield reduction was due to moisture stress. Shiferaw et al., (2012) reported higher grain yield reduction of 51% due to moisture stress for tef genotypes.

#### **2.4.13 Farmers' coping mechanisms against moisture stress in tef production**

The Tigray region of northern Ethiopia, where this study was conducted, is one of the water scarce regions in Ethiopia (Gebrehiwot et al., 2011). Use of early maturing varieties was the highly valued option during moisture stress seasons. Also farmers apply some supplemental irrigation during moisture stress especially in Alamata. In this district, more than half of the respondents adopted a soil and water conservation scheme which is locally referred as "Duga". Water harvested in the scheme is used to supplement their crops during severe moisture stress. In the scheme, farmers harvest rains to conserve moisture using soil structures and divert runoff water into their tef farm during heavy rains. Farmers were asked if they have ever replaced tef with other crops during moisture stressed seasons. They responded that tef is the only hardy cereal crop that survived drought through its early maturity and provided some yield whereas other crop species failed completely due to moisture stress. Likewise, Kassie et al., (2013) reported that farmers in north-eastern Ethiopia allocate their land to tef and other moisture tolerant pulses such as chickpea and haricot bean in the severely moisture stressed seasons.

## 2.5 Conclusions

Tef is an emerging small cereal grain crop presently being world-wide advocated as a health food for people with gluten intolerance. It is one of the most popular and traditional crops in Ethiopia supporting over 50 million people for food security. In the country, tef covers the largest cultivated area of production, but it remains the lowest yielder compared to other cereals. The present yield level of tef needs significant improvement to attain the demand for food security, to buffer the volatile market price of the grain which is most valued to make quality *injera* and consequently to increase profitability of tef growers in the entire value chains of the crop. There is a potential export market of tef in countries such as the USA and those in Europe.

The present study investigated tef production status, input use and tef management practices, identified and prioritized production constraints, variety and trait preferences of tef farmers in moisture stress-prone growing environments. The study was conducted through participatory rural appraisal involving 240 farmers sampled from four major tef growing districts of Tigray Region of northern Ethiopia.

The results showed that tef is the major crop grown by above 95% of the respondent farmers in all the study districts. The major technologies being promoted by the Ethiopian government to boost tef productivity included the use of improved varieties, inorganic fertilizers and planting techniques such as transplanting and row sowing. Constraints on their adoption were the unsuitability of some new varieties, and high labour costs leading farmers to forego transplanting, even though this meant reduced yields.

The grain yield levels in the study areas are lower than the national average of 1.6 t ha<sup>-1</sup>. Low grain yield, moisture stress, weed and shoot fly infestation, lodging and seed shattering were the most important constraints to tef production identified by growers. Moisture stress accounted for 36.7%, 49.0%, 48.2% and 60.3% yield reductions of tef in Laelay-Maichew, Medebay-Zana, Ahferom and Alamata districts, respectively. Changing climates are expected to make moisture stress a more important factor in the growth cycle. If yields are to be maintained and preferably increased, the development of new varieties must therefore focus on traits that result in good adaptation to water stress, the efficient uptake and use of fertilizer inputs, and for the lower rainfall environments, crop maturity in a shorter growing season. This may also require further development of water-conservation practices to help secure productivity gains.

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## Chapter 3: Performance of Tef [*Eragrostis tef* (Zucc.) Trotter] Genotypes for Yield and Yield Components under Drought-Stressed and Non-Stressed Conditions

### Abstract

Tef [*Eragrostis tef* (Zucc.) Trotter] is a staple food crop in Ethiopia, and has become known globally as a health food for its gluten-free flour, which also has a unique nutritional profile. A key to successful variety development through designed breeding is the use of diverse genetic resources, especially when breeding for complex traits such as tolerance to drought stress. The objective of this study was to assess the response of genetically diverse populations of tef genotypes for yield and yield components, with special emphasis on drought-stress tolerance, to select promising parents for breeding. One hundred forty-four tef genotypes were evaluated, in four experiments representing optimum moisture and drought-stressed environments. Data from the non-stressed and drought-stressed environments were subjected to multivariate analysis, including principal component analysis (PCA) and cluster analysis. Days-to-maturity, plant height, panicle length, and panicle seed weight were positively correlated with grain yield in non-stressed conditions, while these traits had negative correlations with grain yield under drought-stressed conditions. The genotypes, DZ-Cr- 387, DZ-01-787, DZ-01-3186, 9432, 9403, 9415, 205917, 205896, 215678, 213237, Jano, Kaye- Agachew, Purpurea, Kaye-Murri, and Dschanger were selected as promising parents with superior grain yields, low levels of lodging, tall plants and long panicles in a non-stressed environment. Conversely, the genotypes DZ-Cr-385, DZ-Cr-37, HO-Cr-136, DZ-01-2053, Dabbi, 207832, Zagure, and Shawa-Gemerra were selected as superior parents for their early maturity and good yield performances under drought stress.

**Keywords:** Cluster analysis, landraces, principal component analysis, quantitative traits, tef

### 3.1 Introduction

Tef [*Eragrostis tef* (Zucc.) Trotter] is the leading cereal crop in Ethiopia, occupying 24% of the crop production area, followed by maize (*Zea mays* L.) and sorghum [*Sorghum bicolor* (L.) Moench] (CSA, 2015). It is valued for the quality of grain and fodder that it provides, both of which have a high market price (Assefa et al., 2011). Furthermore, it is a highly adaptable crop and can be cultivated from sea level up to 3000 m above sea level (m.a.s.l.). However, its best performance occurs between 1800 and 2200 m.a.s.l. (Tefera and Ketema, 2001). It is mainly grown in the regions of Amhara, Oromia, Southern Nation and Nationalities, and Tigray in Ethiopia (Tefera and Ketema, 2001). It is adapted to temperatures ranging from 10 to 27°C and performs best with day lengths of 12 h (Ketema, 1997). The annual rainfall requirement of the crop for optimum yield is in the range of 950 to 1500 mm (FAO, 2011), while moderate yields are still produced under conditions of low rainfall, in the range of 450 to 550 mm (Ketema, 1997).

The mean tef yield is lower (1.58 t ha<sup>-1</sup>) than other cereal crops such as maize (3.4 t ha<sup>-1</sup>), sorghum (2.4 t ha<sup>-1</sup>), wheat (2.5 t ha<sup>-1</sup>), and barley (1.97 t ha<sup>-1</sup>) (CSA, 2015). This is far lower than the potential yield of 6 t ha<sup>-1</sup> estimated by Ketema (1997), primarily because of abiotic stresses, especially drought stress events (Assefa et al., 2011). This constraint increasingly affects tef production, especially in the semi-arid areas of Ethiopia (Teklu, 2014), possibly due to global climate change. Grain yield reductions of between 25.5 and 51.0% have been reported for tef due to drought stress (Admas and Belay, 2011; Shiferaw et al., 2012). Mengistu (2009) also reported reductions of 92.8% for CO<sub>2</sub> assimilation, and 60% for transpiration to have been caused by drought stress in tef. A primary goal of the national tef breeding program in Ethiopia is to develop high yielding, drought tolerant tef varieties (Assefa et al., 2011). This depends on the availability of genetic resources with a range of desirable genes within the national germplasm collection, to develop desirable gene combinations in suitable genetic backgrounds (Rao and Hodgkin, 2002; Aremu, 2011).

To preserve the genetic diversity of the crop, the Ethiopian Biodiversity Institute (EBI) maintains a tef germplasm collection consisting of 5164 landraces and five accessions of wild relatives. Institutions in other countries, including the United States, Japan, Germany, Russia, South Africa, United Kingdom, Yemen, and the Slovak Republic maintained diverse tef collections of 709, 30, 35, 14, 3, 3, 2, and 1 accessions, respectively (Tesema, 2013). Targeted evaluation and utilization of these germplasm collections could contribute to the development of tef

varieties with better drought tolerance, good yield, high biomass, reduced lodging, and other farmers' preferred quality traits.

Understanding the pattern and extent of genetic diversity is essential for systematic genetic resources conservation and gene deployment. Wide genetic diversity for different traits in tef has been reported previously (Assefa et al., 1999, 2001; Ayalew et al., 2011; Plaza-Wuthrich et al., 2013), in assessments conducted under optimal conditions. Screening for genetic diversity under conditions of drought stress, Ayele et al. (2001) and Degu et al. (2008) reported the existence of wide genetic variation for osmotic adjustment, leaf water relations, and root length. They also pointed out that the late maturing and tall tef varieties develop deeper root systems, making them more drought tolerant. However, some tef growing areas of Ethiopia, especially in the northern regions, frequently experience drought stress during the grain-filling periods of tef, that is, in September to November, which leads to significant reductions in tef yields (Tsegay et al., 2012). Their studies only evaluated a limited number of genotypes, and under greenhouse conditions. Screening for drought tolerance is most efficient, if it is applied in the relevant agronomic environment, and uses a broad spectrum of germplasm. The objective of this study was to assess the response of genetically diverse tef varieties and landraces for yield and yield components in drought stressed and non-stressed environments, to select promising parents for subsequent breeding.

## **3.2 Materials and methods**

### **3.2.1 Description of the study environments**

The study was conducted at three sites in the Tigray region of northern Ethiopia, namely, Dura (14°06'76.2" N, 038°39'14.5" E, 2073 m.a.s.l.), Hastebo (14°06'40.2" N, 038°45'45.8" E, 2118 m.a.s.l.) and Dibdibo (14°16'22.1" N, 039°04'15.6" E, 2014 m.a.s.l.), which are the regional research sites of the Axum Agricultural Research Center (AxARC), based in Axum. Dura is situated in a dry environment but has access to irrigation, so it is suitable for offseason experiments. At Hastebo and Dibdibo the absence of irrigation facilities means that field trials have to be timed to coincide with the main rainy season. A scan of 13 year meteorological data from the testing sites shows that their rainfall was variable and erratic over this period [Ethiopian Meteorology Agency, 2014, Annual rainfall and mean temperatures (2002–2014) of Ahferom and Laelay-Maichew districts, Mekelle, Ethiopia, unpublished document]. The rainfall at Hastebo and Dibdibo during 2014 was 937.1 and 992.8 mm year<sup>-1</sup>, respectively, which was ideal for tef

production. The rainfall distribution was high during July and August but considerably lower during the grain-filling periods (September–November) (Appendix 3.1 and 3.2). The mean minimum and maximum temperatures at the Dura and Hastebo sites ranged from 9.9 to 29.4°C, while at Dibdibo it ranged from 7.7 to 36.1°C (Appendix 3.1 and 3.2). The soil types of the sites are clay at Dura, clay loam at Hastebo, and sandy loam at Dibdibo.

### 3.2.2 Plant materials

The test entries included 144 tef genotypes, which are listed in Table 3.1 with their collection sites. Of these 94 accessions were collected from five administrative zones of the Tigray region, namely, the Northwest, Southeast, central, South, East and West zones. Accession refers to the plant materials collected from a particular area. The seed of the accessions was acquired from the Ethiopian Biodiversity Institute (EBI). In addition, 32 improved tef varieties that had been bred and released since 1970 in Ethiopia were included. The remaining test genotypes comprised 18 of the 35 tef landrace cultivars described by Ebba (1975) that originated from various tef growing zones in Ethiopia, including from Shoa (8), Gojam (3), Keffa (2), Welega (3), and Hararge (Table 3.1). A landrace refers to an early cultivated of a crop species, evolved from a wild population. It has historical origin, distinct identity but lacks formal crop improvement. Seed of all the tested genotypes are maintained at the Axum Agricultural Research Center for research purposes.

Table 3.1 List of tef genotypes used in the study.

Population type	Collection Zone/source	No. of entries
Accessions	North-west Tigray	18
	Central Tigray	32
	East Tigray	5
	South-east Tigray	19
	South Tigray	16
	West Tigray	4
Improved varieties	DZARC†	19
	HARC†	3
	MARC†	1
	SARC†	4
	ADARC†	3
	BARC†	1
	ARARC†	1
Landraces	Shoa	8
	Welega	3
	Hararge	2
	Gojam	3
	Keffa	2

†DZARC, Debre-Zeit Agricultural Research Center; HARC, Holleta Agricultural Research Center; MARC, Melkassa Agricultural Research Center; SARC, Sirinka Agricultural Research Center; ADARC, Adet Agricultural Research Center; BARC, Bako Agricultural Research Center; ARARC, Areka Agricultural Research Center.

### 3.2.3 Experimental design and trial management

Each trial was laid out as a 12 by 12 simple lattice design with two replications. Each genotype was planted in four 1 m long rows with an inter-row spacing of 0.25 m. Seeds were drilled in four rows, at a rate of 15 kg ha<sup>-1</sup>. The plot size and design was based on the number of genotypes and the availability of uniform land. Fertilizers in the form of diammonium phosphate (DAP) and urea were each applied at 100 kg ha<sup>-1</sup>, with all of the DAP being applied at planting time, while half the urea was applied at emergence and the other half 4 wk after planting, according to the recommendation made by AxARC (2011). Manual weeding was done twice, at 3 and 5 wk after planting. Plants were harvested from the two central rows of each plot.

Across the three testing sites four trials were conducted, involving two water regimes: Regime I (optimal moisture) at Hastebo (Environment 1) and Dibdibo (Environment 2) (main season rainfall, July–November 2014); and irrigated from planting to physiological maturity (off-season irrigated trial, January–April 2014) at Dura site (Environment 3); and Regime II (drought stressed) at the Dura site (Environment 4), whereby irrigation was withheld after the tef plants reached the stage of 50% heading until maturity, during the off-season (January–April 2014) (Table 3.2).

Table 3.2 Testing sites and the four environment involving two water regimes used for evaluation of 144 tef genotypes during the main and off season of 2014

Site	Environment	Treatment/water regime	Growing season	Total rainfall/irrigated water
Hastebo	E1	Optimal rainfall	Main season (July- November)	937.1 †
Dibdibo	E2	Optimal rainfall	Main season (July- November)	992.8 †
Dura	E3	Irrigated from planting till physiological maturity	Off season (January-April)	739.2 ‡
	E4	Irrigation withheld after 50% days to heading till maturity	Off season (January-April)	494.9‡

† The rainfall amount for the rainy season at Hastebo and Dibdibo is in mm/year

‡ The amount of water applied at Dura for the irrigated experiment is in mm per crop cycle

The crop water requirement was calculated using the following formula:

$$CWR = KC \cdot ET_o,$$

Where CWR is crop water requirement, KC is crop coefficient determined at different growth and developmental stages, and ET<sub>o</sub> is a reference crop evapotranspiration value. The ET<sub>o</sub> was calculated using the CROPWAT 8.0 software developed by the Land and Water Division of FAO (FAO, 1998). Meteorological data for the Dura site, including daily rainfall, maximum and

minimum temperature, relative humidity, sunshine hours and wind speed, were obtained from the Ethiopian Meteorological Agency to calculate the ETo. A total of 739.2 and 494.9 mm irrigated water were applied under the non- and drought-stressed conditions, respectively (Table 3.2).

#### **3.2.4 Data collection**

The following data were collected, on a whole plot basis, considering the two central rows: days to 50% heading; days to 75% maturity; grain-filling period as the difference between days to 75% maturity and days to 50% heading; grain yield (g); biomass yield (g); harvest index; and lodging index. The latter was scored according to the procedure of Caldicott and Nuttall (1979) using a 0 to 5 scale, where plants in an upright position (90°) score 0, and plants lying flat on the ground score 5 (0°). The lodging index was then calculated as the mean of the product sum of the angle of lodging and the corresponding percentage. In addition, individual plant parameters based on 10 randomly selected plants from the two central rows of each plot were recorded at maturity for: plant height (expressed in centimeters); panicle length (cm); number of productive tillers per plant; and panicle seed weight (g).

#### **3.2.5 Data analysis**

Data on the performance of the 144 tef genotypes grown in three non-stressed environments and one stressed environment were subjected to statistical analysis using the simple lattice procedure of SAS 9.3 (SAS Institute, 2011). Combined analysis of variance over the non-stressed environments was performed following Bartlett's homogeneity test of variance. Principal component analysis was conducted using the Statistical Package for Social Sciences (SPSS Inc., 2009). Principal components with eigenvalues greater than unity were considered important in explaining the variability. Cluster analysis was performed using the Euclidean distance-average linkage method, using the GenStat statistical package version 14 (Payne et al., 2011).

### **3.3 Results and discussion**

#### **3.3.1 Analysis of variance**

Genotypes showed highly significant ( $p \leq 0.01$ ) variations for all traits evaluated under the non-stressed conditions (Appendix 3.4). Test environments and genotype, environment interactions

also showed highly significant ( $p \leq 0.01$ ) effects for all traits except for the lodging index. Similarly, in the drought-stressed environment, highly significant ( $p \leq 0.01$ ) differences were observed between genotypes for all traits except biomass yield (Appendix 3.6). In line with the present results, significant genotype differences for the yield and yield components have been reported previously in tef (Assefa et al., 2001; Adnew et al., 2005; Admas and Belay, 2011; Ayalew et al., 2011; Shiferaw et al., 2012; Plaza-Wuthrich et al., 2013). The coefficient of variation (CV%) of the genotypes was higher in the stressed environment than in the non-stressed environments for most of the measured traits. For example, the number of productive tillers per plant had CV% values of 30.4 and 24.7%, in the stressed and non-stressed environments, respectively (Appendix 3.4 and 3.6).

The mean values of the 144 entries under non-stressed and stressed conditions are summarized in Appendix 3.4 and 3.6, respectively. Under the non-stressed conditions, difference in days to maturity of early and late maturing genotypes was 19 d, with a mean value of 96 d. Under stressed conditions the difference was larger (24 d), reflecting the plasticity of the genotypes for maturity. About 44 and 38% of the genotypes were early maturing, in the non-stressed and stressed environments, respectively. Plant height ranged from 67.7 to 103.6 cm in the non-stressed environments, and 44.7 to 80.6 cm in the stressed environment, reflecting the impact of drought stress on plant height. Similarly, Shiferaw et al. (2012) found drought stress to reduce plant height in tef. The difference between the shortest and the tallest plants of the same genotype was 36 cm as a result of the two moisture regimes. Panicle lengths ranged from 24.7 to 41.6 cm under non-stressed and 17.3 to 32.3 cm under stressed conditions. The mean number of productive tillers per plant was 4 in both environments, indicating the limited effect of drought stress on this trait. This could be attributed to drought stress imposed after heading, while plant tillering happens at early stage, before drought stress was imposed. In contrast, Admas and Belay (2011) reported a 16% increase in the number of productive tillers per plant in a non-stressed environment relative to the number of tillers under drought-stressed conditions. The difference in tillering capacity of genotypes in the drought-stressed and non-stressed environments is due to drought stress imposed before heading (i.e., at 45 d after sowing) affecting the tillering stage. The lodging index ranged from 27.3 to 61.4% and 28.0 to 67.3% under non-stressed and stressed conditions, respectively (Appendix 3.4 and 3.6). A higher proportion of the genotypes under drought stress (61%) showed susceptibility to lodging than under non-stressed conditions (55%). The difference in lodging index between tolerant and

susceptible genotypes was 34.2% under non-stressed conditions, and 39.2% under stressed conditions.

Grain yields of genotypes ranged from 0.8 to 2.3 t ha<sup>-1</sup> under non-stressed conditions, and 0.5 to 1.6 t ha<sup>-1</sup> under stressed conditions (Appendix 3.4 and 3.6). The mean yields were 1.4 and 1.0 t ha<sup>-1</sup> under non-stressed and stressed conditions, respectively, a reduction of 29%. Admas and Belay (2011) reported a similar loss in grain yield of 25.5% in tef recombinant inbred lines as a result of drought stress, and Shiferaw et al. (2012) reported a mean grain yield reduction of 51.0% caused by drought stress applied to tef landraces and improved varieties.

A yield difference of 1.5 t ha<sup>-1</sup> was observed between the highest and lowest yielding genotypes in the non-stressed environments, reflecting the range in yield potential of the tested genotypes. Similarly, in the stressed environment a yield difference of 1.1 t ha<sup>-1</sup> was observed between the highest and lowest yielding genotypes. The mean harvest index was 0.24 and 0.21 for non-stressed and stressed plants, respectively (Appendix 3.4 and 3.6). Tef has a low harvest index due to its low grain yield potential, suggesting that future breeding of the crop should aim at improving this valuable trait.

Under the non-stressed conditions, genotypes such as DZ-Cr-387, 9403, 215678, 205896, Dschanger, 9415, 9432, Purpurea, DZ-01-3186 and Jano had grain yields above 2 t ha<sup>-1</sup> (Appendix 3.3). These genotypes were late maturing with tall plants, long panicles, low lodging indexes, and high biomass yields, harvest indexes and panicle seed weights. The reduced lodging indexes of the tall genotypes can be attributed to their thicker stems and longer root systems, relative to the short genotypes (Ayele et al., 2001, Degu et al., 2008 and Van Delden et al., 2010). They also possess longer panicles, which contribute to their higher harvest indexes. Under stressed conditions, genotypes, Dschanger, DZ-Cr-385 and DZ-Cr-37 were the top grain yielders with 1.6, 1.6 and 1.5 t ha<sup>-1</sup>, respectively (Appendix 3.5). Most of the tall genotypes showed late to intermediate maturity, which is a drawback under drought-stressed conditions leading to higher grain yield reductions than early maturing genotypes (Appendix 3.5). To balance the advantage of reduced lodging of the tall plants with their late maturity, selection should be targeted in identifying genotypes with vigorous growth habits, with strong stems and tall plants, combined with early maturity. Alternatively, recombination of the tall but late-maturing genotypes with the early maturing genotypes and selection of segregants for suitable combinations of tall plants, strong stems, long roots and early maturity could reduce the effects of lodging and provide better yield performance under drought-stressed conditions.

Overall, the yield performance of genotypes under stressed and non-stressed conditions were not similar except for the genotype Dschanger, which had high yields in both environments. In addition to good yields in both environments, the genotype, Dschanger, produced tall plants, long panicles and reduced lodging index. However, this genotype has a brown seed color which fetches a lower market price than white seeded tef. Therefore, introgression of genes for white seed color from other genotypes into Dschanger would be worthwhile. Ceccarelli et al. (1992) reported a low to moderate association in yield performance of genotypes under drought stressed and non-stressed conditions, indicating the need for specific breeding to select for drought tolerance in tef.

### 3.3.2 Principal component analysis (PCA)

The PCA is a technique to simplify complex data by transforming the number of associated traits into a smaller number of variables as principal components (PCs). Principal component analyses results of the 11 quantitative traits in non-stressed and stressed environments are presented in Table 3.3. Under non-stressed conditions, three PCs with eigenvalues greater than unity explained 71.39% of the total variation. PC1 explained the most variation (48.24%), and PC2 and PC3 accounted for 12.23 and 10.92% of the total variation, respectively. Days to maturity, plant height, panicle length, panicle seed weight, and grain yield were the largest contributors to PC1. PC2 was correlated mainly with grain-filling period and days to heading, while harvest index accounted for much of PC3.

In the stressed environment, four PCs with eigenvalue  $>1.0$  explained 71.46% of the total variation. PC1, PC2, PC3, and PC4 accounted for 29.51, 17.5, 15.4, and 9.1% of the variation, respectively. Days to heading, days to maturity, plant height, panicle length, biomass yield, and panicle seed weight were the largest contributors to PC1. PC2 was highly correlated with harvest index, grain-filling period, and grain yield. Grain yield was strongly correlated with PC3, and PC4 was highly correlated with the number of tillers per plant and the lodging index (Table 3.3).

Under non-stressed and stressed conditions, the variation within the 11 traits could be explained in three and four dimensions, respectively. Under both stressed and non-stressed conditions, traits such as days to heading, days to maturity, plant height, panicle length, biomass yield, and panicle seed weight showed strong correlation with PC1. Grain yield also showed strong correlation with PC1 under non-stressed conditions whereas there was a poor correlation under stressed conditions (Table 3.3). Accordingly, selection for late maturing genotypes with tall

plants, long panicles, high biomass yields and panicle seed weights would enhance breeding gains for grain yield in non-stressed environments. However, under stressed conditions, breeding for early maturity, short plants, short panicles, and reduced biomass should increase grain yield performance of the progeny. Ayalew et al. (2011) also found that three PCs explaining most of the total variation (70.0%) of 14 agronomic traits of 37 tef genotypes (improved varieties and landraces) tested at Adet in Ethiopia. The authors reported that PC1 was well correlated with grain-filling period, number of productive tillers per plant, grain yield, biomass yield, and the harvest index. Plaza-Wuthrich et al. (2013) found that four PCs explained 80% of the variation of 13 traits for tef landraces grown under greenhouse conditions. In their studies, grain color, days to maturity, the number of panicles and number of internodes per plant, second culm internode diameter, plant height, shoot biomass, grain yield, and harvest index were well correlated with PC1, which explained 40% of the variation. Assefa et al. (2000) reported that five PCs explained 71% of the variation of 17 quantitative traits found in 320 tef lines and 35 landraces, evaluated at two locations in central Ethiopia. These authors found that PC1 was correlated with the number of productive tillers per plant, grain yield and the harvest index. Bedane et al. (2015) found that variation involving 11 traits could be explained by two dimensions (PC1 and PC2), accounting for 73.0% of the total variation. These authors found that their PC1 was mainly due to variations in plant height, peduncle length, panicle length and biomass, and their PC2 was due chiefly to variation in the number of productive tillers per plant and grain yield.

Table 3.3 Eigenvectors and eigenvalues of the first three and four principal components (PC) of 11 quantitative traits of 144 tef genotypes when evaluated under non-stressed and moisture stressed environments, respectively.

Traits	Non-stressed			Stressed			
	PC 1‡	PC 2	PC 3	PC 1	PC 2	PC 3	PC 4
DH†	0.71	-0.58	-0.15	0.63	0.50	-0.35	0.09
DM†	0.83	0.10	-0.37	0.75	-0.26	0.20	-0.18
GFP†	0.20	0.91	-0.31	0.07	-0.73	0.53	-0.24
PLHT†	0.81	-0.01	-0.32	0.81	0.20	0.05	-0.01
PNLG†	0.87	0.09	-0.13	0.79	0.17	-0.12	0.09
NPT†	-0.43	0.05	-0.26	0.09	-0.35	-0.04	0.34
LODG†	-0.61	0.02	0.23	-0.46	-0.26	0.25	0.35
GYLD†	0.83	0.05	0.46	-0.05	0.59	0.76	0.15
BMULD†	0.74	-0.22	0.03	0.60	-0.18	0.35	0.22
HI†	0.50	0.33	0.69	-0.43	0.67	0.48	0.00
PSW†	0.80	0.09	0.16	0.49	-0.06	0.50	-0.02
Eigenvalue	3.07	1.92	1.53	3.25	1.93	1.69	1.0
% of variation	48.24	12.23	10.92	29.51	17.5	15.4	9.1
% of cumulative variation	48.24	60.47	71.39	29.51	47.01	62.41	71.36

† DH, days to 50% heading, DM, days to 75% maturity; GFP, grain filling period; PLHT, plant height; PNLG, panicle length; LODG, lodging index; GYLD, grain yield; BYLD, biomass yield; HI, harvest index; PSW, panicle seed weight; ‡PC, Principal component.

### 3.3.3 Clustering of tef genotypes in the non-stressed environments

The dendrogram generated from data of the non-stressed environments grouped the 144 genotypes into four clusters, which are summarized in Table 3.4. Cluster I included 15 genotypes, three were improved varieties (DZ-Cr-387, DZ-01-3186, and DZ-01-787), while 1, 2, 2, and 2 were accessions acquired from Northwest, central, Southeast, and South zone of Tigray, respectively, and the remaining five were landraces (Jano, Kaye-Agachew, Purpurea, Kaye-Murri, and Dschanger). Cluster II consisted of 8, 6, 3, 1, 1, and 7 genotypes that were improved varieties, accessions from central, East, Southeast, and South Tigray and landraces, in that order. Furthermore, 18 improved varieties and 4, 12, 2, 7, 2, and 2 accessions from Northwest, central, East, Southeast, South, and West Tigray, respectively, were grouped in Cluster III, which was the second largest group. Cluster IV was the largest, consisting of diverse genotypes including improved varieties, landraces and accessions from all collection zones except from eastern Tigray (Table 3.4). The present clustering of genotypes was fewer than the cluster pattern reported by Assefa et al. (1999), who grouped 320 tef accessions into 14 clusters. Similarly, Assefa et al. (2001) grouped 36 tef populations into six classes, each containing 2 to 15 populations. Ayalew et al. (2011) calculated five clusters for 37 tef lines collected from the Amhara Region.

Three improved varieties (DZ-Cr-387, DZ-01-787, DZ-01-3186) and seven accessions (9432, 9403, 9415, 205896, 205917, 215678, 213237) and five landraces (Jano, Kaye-Agachew, Purpurea, Kaye-Murri, and Dschanger) grouped in Cluster I were late maturing with tall plants and long panicles, reduced lodging indexes, and high grain and biomass yields. In Cluster II, the majority of the genotypes were late maturing, with intermediate values for traits such as plant height and panicle length, and high lodging indexes and high grain yield. Cluster III did not show any specific pattern of traits. Most of the improved varieties (about 53%) were grouped within this cluster. Genotypes in Cluster IV showed early maturity, short plants and panicles, high lodging indexes, and intermediate to low grain yields. Only 16% of the improved varieties and 33% of the landraces were grouped within this cluster. About 10 and 22% of the improved varieties were grouped in Clusters I and II, respectively. About 28 and 39% of the landraces were grouped in Clusters I and II, respectively.

Table 3.4 Clustering of 144 tef genotypes of three populations sourced from 18 zones evaluated under optimal moisture conditions.

Population	Collection zone/source†	Cluster and number of genotypes					Total
		I	II	III	IV		
Accessions	North-west Tigray	1	6	4	7	18	
	Central Tigray	2	6	13	11	32	
	East Tigray	-	3	2	-	5	
	South-east Tigray	2	1	7	9	19	
	South Tigray	2	1	2	11	16	
	West Tigray	-	-	2	2	4	
Landraces	Shoa	2	4	-	2	8	
	Gojam	1	1	-	1	3	
	Welega	1	1	-	1	3	
	Keffa	1	-	-	1	2	
	Hararge	-	1	-	1	2	
Improved varieties	DZARC†	2	2	11	4	19	
	HARC†	-	1	1	1	3	
	MARC†	-	-	1	-	1	
	SARC†	-	3	1	-	4	
	ADARC†	1	-	2	-	3	
	BARC†	-	1	-	-	1	
	ARARC†	-	-	1	-	1	
<b>Total</b>		<b>15</b>	<b>31</b>	<b>47</b>	<b>51</b>	<b>144</b>	

†DZARC, Debre-Zeit Agricultural Research Center; HARC, Holleta Agricultural Research Center; MARC, Melkassa Agricultural Research center; SARC, Sirinka Agricultural Research Center; ADARC, Adet Agricultural Research Center; BARC, Bako Agricultural Research Center; ARARC, Areka Agricultural Research Center.

### 3.3.4 Clustering of tef genotypes in the drought-stressed environment

In the drought-stress experiment, the cluster analysis grouped the 144 test tef genotypes into six clusters of 5 to 83 (Table 3.5). Clusters I, II, III, IV, V, and VI consisted 16, 5, 17, 83, 15, and 8 genotypes, respectively. A majority of the clusters were heterogeneous, comprised of accessions, improved varieties and landraces from different zones/sources. Cluster I included seven improved varieties (DZ-Cr-387, DZ-Cr-255, DZ-Cr-44, DZ-Cr-387-RIL127, Acc.205953, DZ-01-1880, and DZ-01-787); three, two, and two accessions from central, Southeast, and South Tigray, respectively. It included two landraces (Jano and Many). Cluster II consisted of one accession each from North western, central, East, and South of Tigray and the landrace Curati. Four improved varieties, one, four, two, and two accessions from Northwest, central, South east, and South zone of Tigray, respectively, and three landraces were included in Cluster III. Cluster IV was the largest group, consisting of 17 improved varieties, 14, 19, 4, 13, 6, and 3 accessions from Northwest, central, East, Southeast, South and West zone of Tigray, respectively. In addition, it included seven landraces. Cluster V was dominated by accessions from all the Tigray zones, except for east Tigray. It included only two of the landraces. There

were, no improved varieties grouped in this cluster. Genotypes grouped in Cluster VI were one accession from central and South Tigray, four improved varieties and two landraces.

A majority of the genotypes in Cluster I, except DZ-Cr-387-RIL127, were late maturing with tall plants and long panicles, relatively lodging resistant, high biomass and intermediate to low grain yields. Cluster II included genotypes of intermediate values for a majority of the traits. Genotypes in Cluster III were late maturing with tall plants, long panicles, intermediate to high lodging, and intermediate biomass, harvest index, panicle seed weight and grain yield, except for the genotype Dschanger, which was the highest grain yielder ( $1.62 \text{ t ha}^{-1}$ ). Other genotypes in this cluster yielded  $0.66$  to  $1.3 \text{ t ha}^{-1}$ . Cluster IV did not show specific trait patterns. Genotypes in Cluster V were characterized by early maturity but low grain yield performance. The cluster consisted of four improved varieties (DZ-Cr-385, DZ-Cr-37, HO-Cr-136, and DZ-01-2053), two accessions (Zagure and 207832) and two landraces (Dabbi and Shawa-Gemerra), which were early maturing with short plants and good grain yields.

The clustering pattern of the genotypes was different in the stressed and non-stressed environments. For instance, the genotypes DZ-Cr-385, DZ-Cr-37, HO-Cr-136, DZ-01-2053, Dabbi, 207832, Zagure, and Shawa-Gemerra were grouped in Cluster V for their early maturity and superior grain yields under stressed conditions. These genotypes were allocated to Cluster IV under non-stressed conditions for their early maturity, with intermediate to low grain yield. Genotypes, DZ-Cr-387, DZ-01-787, 205917, and Jano were grouped in Cluster I for their high grain yield and late maturity in the non-stressed environment, whereas they were found to be low grain yielders in the drought-stressed environment. The landraces were grouped only in three clusters when tested under non-stressed conditions, while they were separated into six clusters when evaluated under stressed conditions. This suggests that the landrace varieties are better distinguished when tested under stressed conditions, an important aspect for selection. This outcome was in agreement with the study of Plaza-Wuthrich et al. (2013) in which 17 landrace varieties were grouped into six clusters under stressed conditions.

Table 3.5 Clustering of 144 tef genotypes of three populations sourced from 18 zones evaluated under moisture stressed environment.

Population	Collection zone/source†	Cluster and number of genotypes						Total
		I	II	III	IV	V	VI	
Accessions	North-west Tigray	-	1	1	14	2	-	18
	Central Tigray	3	1	4	19	4	1	32
	East Tigray	-	1	-	4	-	-	5
	South-east Tigray	2	-	2	13	2	-	19
	South Tigray	2	1	2	6	4	1	16
	West Tigray	-	-	-	3	1	-	4
Landraces	Shoa	1	-	2	3	2	-	8
	Gojam	1	1	-	-	-	1	3
	Welega	-	-	1	2	-	-	3
	Keffa	-	-	1	-	-	1	2
	Hararge	-	-	-	2	-	-	2
Improved varieties	DZARC†	4	-	-	12	-	3	19
	HARC†	1	-	-	1	-	1	3
	MARC†	1	-	-	-	-	-	1
	SARC†	-	-	2	2	-	-	4
	ADARC†	-	-	2	1	-	-	3
	BARC†	1	-	-	-	-	-	1
	ARARC†	-	-	-	1	-	-	1
Total		16	5	17	83	15	8	144

†DZARC, Debre-Zeit Agricultural Research Center; HARC, Holleta Agricultural Research Center; MARC, Melkassa Agricultural Research Center; SARC, Sirinka Agricultural Research Center; ADARC, Adet Agricultural Research Center; BARC, Bako Agricultural Research Center; ARARC, Areka Agricultural Research Center.

### 3.4 Conclusions

The tested tef genotypes showed a wide range of variation for all the traits considered under drought stressed and non-stressed conditions. The PCA analysis revealed that selection in non-stressed environments needs to be for late-maturing genotypes with tall plants, long panicles, high biomass, and reduced levels of lodging, to select for high potential, high yielding tef genotypes. However, in the drought-stressed environment, these traits were correlated with a reduced grain yield. Classification of genotypes for drought-stress tolerance was not consistent when they were evaluated in drought-stressed and non-stressed environments, indicating the diversity of performances of the tested tef genotypes, in response to the two moisture regimes. The genotypes DZ-Cr-387, DZ-01-787, DZ-01-3186, 9432, 9403, 9415, 205917, 205896, 215678, 213237, Jano, Kaye-Agachew, Purpurea, Kaye-Murri and Dschanger were selected as promising parents with good grain yields, reduced lodging, tall plants, and long panicles in a non-stressed environment. On the other hand, genotypes, DZ-Cr-385, DZ-Cr-37, HO-Cr-136, DZ-01-2053, Dabbi, 207832, Zagure, and Shawa-Gemerra, were selected as superior parents for their early maturity and good yields under drought-stressed conditions. This study demonstrated the need to evaluate diverse tef genotypes in both moisture-stressed and non-

stressed environments, to select for varieties with either high yield potential, or yield stability, as a basis for the strategic breeding of tef.

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Appendix 3.1 Monthly and total annual rainfall and mean minimum and maximum temperatures (2002-2014) of Hastebo and Dura sites

Year	Monthly rainfall (mm)												Total rainfall (mm)	Temperature (°C)	
	Jan	Feb	Mar	Apr	May	Jun	Jul	Aug	Sep	Oct	Nov	Dec		Mean max	Mean min
2000	0.0	0.0	2.0	66.5	11.4	13.6	162.8	139.1	100.4	160.5	16.8	0.0	673.1	26.8	10.9
2001	0.0	0.0	1.5	6.4	15.9	74.0	212.1	355.6	25.8	14.5	0.0	0.0	705.8	27.9	11.9
2002	0.0	8.8	10.0	15.3	9.2	31.1	102.3	96.5	50.6	1.3	1.8	27.7	354.6	28.8	12.2
2003	2.5	8.5	2.4	8.3	12.2	126.1	322.7	209.1	89.6	1.4	3.7	0.0	786.5	29.4	11.7
2004	18.2	3.8	3.9	41.0	0.0	132.4	269.9	173.6	16.8	24.4	34.8	0.0	718.8	27.6	12.0
2005	0.0	0.0	129.7	86.0	5.1	85.4	176.6	226.0	67.2	0.0	0.0	0.0	776.0	28.5	11.2
2006	0.0	0.0	0.0	31.1	61.0	86.5	230.7	240.6	123.9	9.5	0.0	30.5	813.8	28.0	10.8
2007	0.0	0.0	7.0	10.9	35.5	112.6	428.1	272.8	154.3	0.0	6.1	0.0	1027.3	28.0	10.6
2008	38.5	0.0	0.0	85.2	41.3	102.3	161.8	174.7	49.9	1.5	6.8	0.0	662.0	26.6	10.2
2009	0.0	0.0	0.9	6.0	8.6	35.6	231.9	288.6	1.8	0.0	2.9	0.0	576.3	27.5	11.3
2010	1.2	0.0	54.2	36.3	17.3	109.4	209.4	223.2	137.5	20.3	0.0	0.0	808.8	27.4	9.9
2011	1.2	0.0	1.9	7.9	58.7	13.5	204.2	151.1	93.7	3.4	11.8	0.0	547.4	26.4	10.0
2012	0.0	0.0	2.7	14.6	4.3	93.9	157.0	289.5	119.5	0.0	3.2	0.0	684.7	27.8	11.2
2013	0.0	0.0	0.0	34.4	20.9	84.8	166.0	248.2	7.5	10.2	0.0	0.0	572.0	28.9	12.7
2014	0.0	0.0	59.4	68.0	55.0	123.2	185.0	217.5	194.0	18.0	13.0	4.0	937.1	27.7	11.9
Mean	4.1	1.4	18.4	34.5	23.8	81.6	214.7	220.4	82.2	17.7	6.7	4.1	709.6	27.8	11.2

Source: Ethiopian Metrology Agency, Mekelle branch (2014)

Appendix 3.2 Monthly and total annual rainfall and mean minimum and maximum temperatures (2002-2014) of Dibdibo site

Year	Monthly rainfall (mm)												Total rainfall (mm)	Temperature (°C)	
	Jan	Feb	Mar	Apr	May	Jun	Jul	Aug	Sep	Oct	Nov	Dec		Mean max	Mean min
2002	0.0	0.3	0.0	0.0	0.0	43.8	85.3	270.7	58.0	1.5	4.0	5.8	469.4	25.1	8.4
2003	0.0	19.9	23.0	36.8	7.2	71.1	237	184.6	60.5	1.9	0.0	0.0	642.0	24.3	11.0
2004	41.7	22.1	24.2	47.9	0.8	0.0	213.2	388.0	23.5	11.3	0.0	0.0	772.7	26.9	7.7
2005	0.0	0.0	71.0	32.2	54.8	98.8	281.4	274.9	0.3	0.0	9.2	0.0	822.6	24.2	10.9
2006	0.0	0.0	18.1	41.3	40.2	103.9	216.6	241.2	95.9	1.0	21.4	27.0	806.6	24.7	5.8
2007	1.0	6.0	17.3	25.2	7.1	99.2	309.8	277.2	103.0	0.0	0.0	0.0	845.8	28.9	16.0
2008	18.9	0.0	0.0	18.5	72.7	87.7	232.6	202.8	56.9	10.5	19.1	0.0	719.7	28.6	10.3
2009	0.0	17.7	20.0	31.0	6.9	3.5	331.6	227.0	1.5	18.5	2.3	0.0	660.0	24.7	16.7
2010	6.3	0.0	0.0	10.5	43.0	44.5	189.3	228.0	86.8	0.0	0.0	0.0	608.4	36.1	8.0
2011	0.0	0.0	15.5	0.0	54.5	0.0	214.1	193.9	22.4	0.0	0.0	0.0	500.4	23.0	15.1
2012	0.0	0.0	0.0	0.5	39.0	98.0	480.2	325.3	25.4	7.7	49.0	0.0	1025.1	23.3	13.2
2013	0.0	0.0	0.0	1.5	26.7	97.3	102.3	199.0	19.2	6.2	5.0	0.0	457.2	35.0	12.4
2014	0.0	0.0	26.0	47.0	29.3	199.5	233.6	230.7	146.0	37.0	21.9	21.8	992.8	27.5	10.9
Mean	5.2	5.1	16.5	22.5	29.4	72.9	240.5	249.5	53.8	7.4	10.1	4.2	717.1	27.1	11.3

Source: Ethiopian Metrology Agency, Mekelle branch (2014)

Appendix 3.3 Mean values for 11 traits of the 144 tef genotypes evaluated under three non-stressed sites

No.	Genotype	Traits										
		DH †	DM †	GFP †	PLHT †	PNLG †	NPT †	LODG †	GYL D †	BYLD D †	HI †	PSW †
1	<b>DZ-Cr-387 ‡</b>	64.8	105.7	40.8	100.6	41.0	3.4	32.9	2.3	8.2	0.28	0.95
2	219873	62.5	102.0	39.5	74.6	33.7	3.0	40.2	1.2	5.3	0.25	0.52
3	acc-13-al	49.2	92.5	43.3	80.8	32.3	4.0	48.7	1.4	5.4	0.26	0.39
4	<b>9403</b>	60.3	103.0	42.7	100.2	39.9	3.2	43.9	2.2	7.4	0.30	0.91
5	9437	53.7	96.2	42.5	90.9	36.2	4.0	44.7	1.4	5.6	0.26	0.72
6	205905	48.8	91.3	42.5	78.7	30.4	5.0	44.2	1.3	6.3	0.22	0.44
7	acc-11-al	51.5	89.5	38.0	76.9	30.1	4.6	43.2	1.4	6.1	0.24	0.57
8	9442	53.0	93.0	40.0	84.0	29.2	5.0	44.9	1.3	6.0	0.23	0.67
9	205925	57.3	103.2	45.8	92.2	35.6	3.8	51.7	0.9	5.6	0.17	0.60
10	DZ-Cr-2675	54.5	102.0	47.5	98.8	39.0	3.9	41.8	1.4	7.2	0.20	0.58
11	222108	59.0	97.2	38.2	91.1	32.2	4.3	42.1	1.1	6.7	0.17	0.61
12	9440	52.3	93.2	40.8	80.1	32.1	4.9	51.7	1.6	7.0	0.23	0.51
13	213231	55.2	97.8	42.7	98.1	37.0	3.9	43.1	0.9	5.0	0.19	0.59
14	acc-18-rm	48.5	89.5	41.0	79.0	33.1	3.6	59.9	1.0	5.5	0.20	0.66
15	205924	49.8	87.7	37.8	83.3	27.4	5.7	47.7	1.0	5.2	0.20	0.45
16	9427	57.3	97.0	39.7	85.4	31.5	6.1	45.2	1.0	5.5	0.18	0.58
17	DZ-Cr-44	54.3	99.2	44.8	102.7	36.0	4.0	53.3	1.4	5.8	0.23	0.69
18	207832	48.5	86.5	38.0	70.6	24.7	6.2	55.5	1.0	5.2	0.19	0.42
19	DZ-01-99	54.2	91.7	37.5	86.0	35.5	5.3	43.7	1.6	5.7	0.28	0.50
20	9443	52.7	96.3	43.7	85.2	31.9	4.2	41.1	1.4	5.6	0.26	0.77
21	207840	55.5	100.3	44.8	95.1	38.5	4.1	37.3	1.4	6.0	0.24	0.49
22	DZ-Cr-385	45.5	86.7	41.2	80.3	30.0	3.7	55.7	1.4	5.0	0.29	0.66
23	Curati	56.7	99.2	42.5	91.3	35.4	4.7	50.6	1.6	6.7	0.24	0.66
24	206442	58.5	98.8	40.3	82.7	36.5	4.8	44.5	1.2	6.0	0.20	0.59

† DH, days to 50% heading, DM, days to 75% maturity; GFP, grain filling period; PLHT, plant height; PNLG, panicle length; NPT, number of productive tillers per plant; LODG, lodging index; GYLD, grain yield; BYLD, biomass yield; HI, harvest index; PSW, panicle seed weight

‡ Bold text denotes the top ten high yielding genotypes under non-stressed condition

## Appendix 3.3 Continued

No.	Genotype	Traits										
		DH	DM	GFP	PLHT	PNLG	NPT	LODG	GYL D	BMYL D	HI	PSW
25	<b>215678</b>	55.2	103.0	47.8	95.2	38.4	4.7	46.5	2.1	6.8	0.32	0.86
26	222077	45.5	86.7	41.2	76.8	30.8	4.8	52.2	1.0	5.3	0.20	0.45
27	9408	51.8	94.3	42.5	86.4	34.6	3.9	45.7	1.3	6.2	0.21	0.41
28	DZ-Cr-37	50.7	86.7	36.0	86.7	33.5	4.0	45.0	1.6	5.9	0.27	0.45
29	222107	53.8	91.3	37.5	86.4	34.1	4.4	56.0	1.6	6.2	0.26	0.65
30	205891	51.5	92.3	40.8	84.8	34.9	4.5	43.4	1.1	4.9	0.23	0.49
31	207831	51.7	95.7	44.0	86.0	31.4	4.0	48.4	1.2	4.9	0.24	0.52
32	DZ-01-146	56.3	98.8	42.5	90.8	35.1	4.2	37.9	1.5	6.3	0.24	0.75
33	DZ-01-1821	59.7	103.5	43.8	84.1	33.5	4.1	50.9	1.2	5.2	0.24	0.75
34	9413	49.8	95.0	45.2	79.8	28.6	5.2	58.8	1.3	5.4	0.25	0.70
35	205944	59.8	101.8	42.0	91.5	33.6	3.5	53.9	1.1	6.0	0.20	0.49
36	DZ-01-409	54.7	100.0	45.3	93.7	34.4	3.9	47.3	1.5	5.5	0.27	0.72
37	9422	51.7	91.7	40.0	81.8	31.7	3.7	51.4	1.2	5.8	0.21	0.62
38	Manya	64.2	98.7	34.5	96.1	33.3	3.5	46.7	1.7	7.0	0.25	0.82
39	205915	54.5	95.5	41.0	87.0	31.6	4.2	58.3	1.0	5.6	0.18	0.43
40	DZ-01-899	57.0	97.2	40.2	90.3	33.8	4.6	49.7	1.2	5.5	0.23	0.70
41	Fesho	60.0	96.2	36.2	81.7	31.7	3.5	57.6	1.3	5.5	0.25	0.51
42	DZ-Cr-387RIL273	59.7	99.0	39.3	96.4	31.4	4.5	50.8	1.4	6.5	0.22	0.55
43	DZ-01-2423	53.3	99.8	46.5	87.6	35.1	4.1	41.5	1.8	6.2	0.29	0.65
44	acc-15-ck	55.3	88.2	32.8	76.6	28.6	3.7	50.6	0.9	4.8	0.20	0.45
45	Ho-Cr-136	50.3	88.2	37.8	82.1	29.5	5.3	38.3	1.1	5.3	0.22	0.41
46	PGRC/E 205396	48.8	95.7	46.8	91.9	35.5	2.8	50.5	1.5	6.4	0.24	0.68
47	205943	53.0	96.3	43.3	93.5	34.7	3.1	49.5	1.4	6.4	0.22	0.54
48	<b>205896</b>	60.7	101.5	40.8	96.4	40.3	2.9	43.8	2.1	8.1	0.27	0.87

## Appendix 3.3 Continued

No.	Genotype	Traits										
		DH	DM	GFP	PLHT	PNLG	NPT	LODG	GYL D	BMYL D	HI	PSW
49	Dabbi	50.7	89.2	38.5	79.6	30.1	4.8	49.2	1.3	4.9	0.26	0.39
50	205899	60.5	97.3	36.8	86.1	34.5	5.7	43.0	1.6	6.8	0.25	0.69
51	acc-01-dib	59.3	98.3	39.0	93.2	32.1	3.5	51.3	1.7	6.6	0.26	0.77
52	9414	52.8	94.2	41.3	93.0	36.3	4.1	51.4	1.3	6.3	0.22	0.70
53	9423	54.7	96.2	41.5	83.9	31.6	3.2	49.9	1.6	6.1	0.26	0.52
54	222078	52.3	93.3	41.0	83.5	32.0	4.0	53.9	1.5	6.2	0.25	0.60
55	207835	47.8	89.8	42.0	78.9	29.1	5.0	54.0	1.1	6.1	0.19	0.55
56	<b>DZ-01-2053</b>	48.5	93.3	44.8	80.9	30.8	4.5	56.0	1.2	5.7	0.23	0.55
57	221704	49.0	90.7	41.7	76.5	29.2	3.9	52.3	1.0	5.6	0.19	0.56
58	205898	53.3	92.5	39.2	87.5	35.4	3.6	50.0	1.5	6.1	0.24	0.58
59	DZ-Cr-358	52.0	90.3	38.3	85.6	33.2	3.1	51.0	1.7	7.0	0.25	0.69
60	219878	61.2	94.5	33.3	76.6	27.8	3.2	47.0	1.4	6.8	0.20	0.41
61	207839	53.2	100.8	47.7	91.8	33.8	3.6	47.7	1.8	6.6	0.28	0.73
62	Kaye-Agachew	59.2	104.0	44.8	92.8	36.7	3.2	44.7	1.8	6.6	0.27	0.62
63	DZ-01-1880	60.5	98.2	37.7	94.3	37.5	4.5	42.3	1.6	6.8	0.23	0.62
64	9396	52.0	93.0	41.0	87.5	31.4	5.1	49.7	1.4	6.0	0.23	0.49
65	Gea-Lamie	54.5	93.7	39.2	82.0	31.3	4.6	46.8	1.3	6.1	0.22	0.58
66	9419	53.3	97.2	43.8	96.2	35.6	3.7	44.4	1.4	6.3	0.22	0.51
67	Rosea	49.8	87.3	37.5	86.3	30.8	3.5	56.4	1.3	5.3	0.24	0.52
68	9438	51.8	90.5	38.7	81.7	32.7	4.0	45.3	1.6	5.1	0.31	0.66
69	DZ-Cr-82	53.5	99.5	46.0	92.7	36.8	3.4	45.8	1.4	5.4	0.26	0.70
70	DZ-01-1281	50.0	93.7	43.7	93.9	33.9	4.0	56.3	1.6	6.6	0.26	0.65
71	219880	48.5	91.3	42.8	84.4	31.0	4.8	52.2	1.2	5.4	0.23	0.66
72	219877	53.7	95.8	42.2	91.9	36.9	3.8	55.9	1.5	6.7	0.23	0.71

## Appendix 3.3 Continued

No.	Genotype	Traits										
		DH	DM	GFP	PLHT	PNLG	NPT	LODG	GYL D	BMYL D	HI	PSW
73	DZ-01-1681	51.3	93.5	42.2	92.7	38.3	4.5	51.0	1.5	6.2	0.24	0.71
74	Shawa-Gemerra	48.7	86.8	38.2	86.0	28.9	5.0	51.1	0.9	4.9	0.21	0.40
75	9433	54.3	93.2	38.8	81.5	33.6	5.1	47.3	1.4	6.2	0.23	0.45
76	Zezew	52.7	97.7	45.0	93.9	33.6	3.9	46.9	1.6	6.3	0.26	0.72
77	Zagure	49.0	89.8	40.8	80.7	31.0	3.7	53.1	1.2	5.2	0.24	0.43
78	9432	59.8	101.2	41.3	100.8	39.9	3.7	43.4	2.1	8.0	0.28	0.76
79	205919	49.3	95.7	46.3	93.5	35.1	3.9	54.4	1.2	5.2	0.24	0.70
80	Rubicunda	60.7	96.3	35.7	92.8	34.4	4.0	45.6	1.4	5.9	0.24	0.61
81	205927	50.2	98.3	48.2	88.9	31.9	3.7	49.5	1.1	5.7	0.22	0.69
82	219849	51.8	94.8	43.0	86.1	32.6	5.3	49.1	1.2	5.9	0.20	0.38
83	206745	61.8	96.7	34.8	91.1	37.3	3.5	58.4	1.3	5.7	0.24	0.48
84	9430	61.5	99.3	37.8	89.4	33.0	3.5	48.2	1.1	6.0	0.22	0.53
85	9435	53.3	97.7	44.3	89.1	33.3	4.1	43.1	1.3	6.0	0.21	0.35
86	DZ-01-196	59.3	99.0	39.7	96.3	36.4	3.7	37.4	1.7	6.7	0.26	0.74
87	207827	50.7	87.8	37.2	80.5	29.0	3.4	51.0	1.5	6.1	0.24	0.53
88	Ada	52.0	93.8	41.8	79.5	27.9	4.1	49.7	1.3	6.0	0.23	0.38
89	Enatite	60.2	98.8	38.7	87.2	35.8	3.9	46.2	1.6	6.6	0.25	0.77
90	205921	62.7	100.5	37.8	103.1	33.5	3.7	55.8	1.3	6.9	0.20	0.73
91	DZ-01-1278	53.7	97.2	43.5	92.8	37.2	3.7	53.4	1.5	6.0	0.24	0.88
92	DZ-Cr-354	53.3	96.8	43.5	98.3	36.2	4.5	47.7	1.5	6.5	0.24	0.84
93	9446	48.8	93.5	44.7	76.9	30.4	4.5	57.1	1.5	6.4	0.24	0.69
94	213237	51.8	98.2	46.3	94.4	40.8	3.6	49.4	1.8	6.4	0.28	0.85
95	207833	52.3	94.3	42.0	82.8	28.8	4.1	54.2	1.3	6.3	0.21	0.59
96	222072	61.8	96.7	34.8	83.6	30.4	5.0	47.8	1.2	6.0	0.20	0.61

## Appendix 3.3 Continued

No.	Genotype	Traits										
		DH	DM	GFP	PLHT	PNLG	NPT	LODG	GYL D	BMYL D	HI	PSW
97	9441	51.7	98.5	46.8	91.1	35.7	4.4	40.8	1.2	5.5	0.25	0.61
98	DZ-01-974	54.2	97.3	43.2	94.4	33.6	3.6	48.4	1.4	7.1	0.20	0.72
99	9424	60.8	97.7	36.8	93.0	37.0	3.7	47.3	1.5	5.7	0.26	0.58
100	DZ-Cr-387-RIL127	53.5	99.2	45.7	97.1	35.5	4.3	43.8	1.3	6.4	0.22	0.83
101	206739	54.2	101.5	47.3	92.1	33.7	3.3	44.3	1.3	5.6	0.24	0.55
102	DZ-01-1868	55.2	98.7	43.5	97.1	38.0	4.4	42.4	1.6	7.5	0.21	0.82
103	Acc.205953	59.7	99.8	40.2	97.6	36.5	3.2	52.5	1.3	6.0	0.21	0.81
104	219858	51.3	95.2	43.8	82.7	30.9	3.7	53.6	1.4	6.0	0.24	0.65
105	Kaye-Murri	56.5	105.0	48.5	94.0	40.3	2.8	34.0	2.0	7.6	0.26	0.95
106	9393	53.5	97.8	44.3	84.3	31.6	4.1	54.3	1.4	6.7	0.21	0.68
107	205923	48.5	93.2	44.7	77.1	30.1	4.6	45.9	1.2	5.2	0.24	0.51
108	DZ-01-787	58.2	101.3	43.2	96.7	35.6	3.1	46.3	1.8	7.4	0.24	0.81
109	acc-16-ck	52.7	90.7	38.0	79.3	27.5	3.6	55.3	1.2	6.9	0.18	0.53
110	205908	55.5	98.8	43.3	93.7	33.7	3.6	33.8	1.1	5.3	0.20	0.59
111	219881	51.2	91.8	40.7	89.8	33.2	4.0	53.3	1.2	6.3	0.20	0.67
112	DZ-01-2054	60.2	97.0	36.8	90.2	33.3	3.4	52.1	1.4	5.8	0.25	0.77
113	219860	57.3	93.7	36.3	91.9	34.5	3.1	54.4	1.5	6.5	0.24	0.84
114	207828	52.7	93.2	40.5	86.2	31.1	3.4	51.6	1.5	6.6	0.24	0.75
115	Denkeye	51.2	94.3	43.2	81.0	29.9	4.6	47.1	1.1	4.7	0.24	0.50
116	9400	60.8	97.0	36.2	97.0	34.2	4.0	47.9	1.3	6.1	0.21	0.73
117	207844	53.2	92.7	39.5	79.1	30.7	4.8	45.6	1.5	6.0	0.25	0.48
118	205931	48.8	90.0	41.2	75.9	33.9	3.0	54.8	1.0	5.0	0.22	0.41
119	<b>Jano</b>	64.5	102.8	38.3	92.2	39.8	3.2	27.3	2.0	6.8	0.30	0.99
120	acc-04-dib	55.3	96.0	40.7	93.0	37.0	3.4	51.4	1.3	5.6	0.24	0.67

## Appendix 3.3 Continued

No.	Genotype	Traits										
		DH	DM	GFP	PLHT	PNLG	NPT	LODG	GYL D	BMYL D	HI	PSW
121	<b>DZ-01-3186</b>	61.2	100.2	39.0	95.8	40.8	3.6	35.8	2.1	7.8	0.27	0.93
122	<b>9415</b>	61.2	97.2	36.0	98.6	40.2	3.1	36.3	2.1	7.7	0.28	0.93
123	acc-17-wj	49.0	89.7	40.7	77.1	28.6	3.8	60.5	1.2	4.6	0.27	0.66
124	205917	61.3	99.8	38.5	103.6	41.6	3.3	33.8	1.9	7.1	0.27	1.01
125	9416	53.0	92.8	39.8	85.5	33.0	3.7	59.9	1.5	5.8	0.25	0.57
126	219865	47.2	86.8	39.7	73.2	24.9	4.8	61.4	1.4	5.7	0.24	0.40
127	219852	51.8	91.8	40.0	85.7	33.4	4.1	51.6	1.4	6.8	0.21	0.38
128	219871	56.3	97.5	41.2	88.6	33.1	4.7	47.3	1.5	6.7	0.22	0.49
129	Gorradie	56.7	99.0	42.3	90.3	32.0	4.5	56.9	1.3	5.3	0.26	0.57
130	206750	54.7	94.7	40.0	89.0	33.8	3.8	52.7	1.2	5.4	0.23	0.88
131	219863	47.8	85.3	37.5	76.0	31.0	2.5	56.7	1.0	4.9	0.22	0.60
132	<b>Purpurea</b>	60.8	105.7	44.8	92.4	39.6	3.8	33.9	2.1	7.1	0.30	1.09
133	<b>Dschanger</b>	60.7	101.8	41.2	93.5	39.4	3.6	30.6	2.1	7.2	0.30	0.94
134	9395	53.3	95.0	41.7	90.6	32.6	3.6	42.1	1.5	6.7	0.23	0.63
135	205913	54.8	95.7	40.8	98.3	36.7	3.1	42.2	1.4	6.3	0.23	0.72
136	205936	49.3	94.3	45.0	75.1	30.6	3.9	57.0	1.3	5.0	0.27	0.47
137	222076	47.7	86.5	38.8	67.7	24.7	4.2	61.3	0.8	3.6	0.24	0.27
138	acc-02-dib	51.8	98.2	46.3	91.0	35.2	4.7	32.7	1.2	5.8	0.21	0.45
139	205922	52.0	99.7	47.7	92.1	36.3	4.2	34.1	1.3	6.0	0.23	0.66
140	207575	53.0	94.8	41.8	81.3	33.4	4.3	54.1	1.6	6.4	0.25	0.59
141	Alba	59.0	98.7	39.7	95.6	28.3	3.3	31.4	1.4	5.1	0.28	0.50
142	206753	51.7	93.8	42.2	73.6	28.3	3.3	56.3	1.1	4.7	0.25	0.57
143	DZ-Cr-255	58.5	100.8	42.3	85.5	34.1	4.4	41.7	1.4	5.4	0.27	0.75
144	DZ-01-1285	54.2	99.2	45.0	89.8	34.5	4.9	45.7	1.4	4.7	0.31	0.71

Appendix 3.4 Mean, minimum, maximum, standard error, mean squares and coefficient of variation for 11 traits of 144 tef genotypes evaluated under three non-stressed sites

No.	Statistics	Traits										
		DH†	DM†	GFP†	PLHT†	PNLG†	NPT†	LODG†	GYLD†	BYLD†	HI†	PSW†
1	Mean	54.4	95.7	41.3	87.9	33.6	4.0	48.0	1.4	6.0	0.24	0.63
2	Minimum	45.5	85.3	32.8	67.7	24.7	2.5	27.3	0.8	3.7	0.17	0.27
3	Maximum	64.8	105.7	48.5	103.6	41.6	6.2	61.4	2.3	8.2	0.32	1.09
4	Standard error	1.0	1.2	1.4	3.3	2.0	0.7	7.2	0.1	0.6	0.03	0.11
5	Genotype MS‡	110.5**	116.2**	59.3**	310.1**	67.8**	2.6**	272.9**	0.5**	3.3**	0.01**	0.2**
6	Environment MS‡	3451.8**	8839.0**	18864.7**	151270.1**	18128.5**	261.5**	122.0Ns#	4.6**	43.3**	0.3**	0.1**
7	Gen. x Env.§ MS‡	26.5**	15.0**	23.6**	66.6**	19.9**	2.3**	117.9Ns#	0.1**	1.7**	0.004**	0.04**
8	CV¶ (%)	2.7	1.8	4.8	5.3	8.6	24.7	21.2	12.8	14.7	16.9	25.4

† DH, days to 50% heading, DM, days to 75% maturity; GFP, grain filling period; PLHT, plant height; PNLG, panicle length; NPT, number of productive tillers per plant; LODG, lodging index; GYLD, grain yield; BYLD, biomass yield; HI, harvest index; PSW, panicle seed weight

‡MS = mean squares

§Gen. x Env., genotype by environment interaction

¶CV, coefficient of variation

#Ns, non-significant

Appendix 3.5 Mean values for 11 traits of the 144 tef genotypes evaluated under moisture stressed condition

No.	Genotype	Traits										
		DH†	DM†	GFP†	PLHT†	PNLG†	NPT†	LODG†	GYLD†	BYLD†	HI†	PSW†
1	DZ-Cr-387	68.5	99.0	30.5	69.7	27.5	2.7	50.5	1.0	7.0	0.15	0.86
2	219873	70.5	93.7	23.2	71.7	27.9	3.4	51.0	1.1	5.1	0.22	0.35
3	acc-13-al	55.0	88.0	33.0	49.8	22.4	3.5	51.0	1.0	5.3	0.18	0.36
4	9403	58.0	97.5	39.5	72.5	28.4	2.9	43.8	1.2	6.2	0.18	0.98
5	9437	55.0	90.5	35.5	63.6	26.0	4.1	54.9	0.8	4.9	0.14	0.51
6	205905	54.5	88.0	33.5	54.1	20.6	4.3	54.6	0.9	3.6	0.25	0.52
7	acc-11-al	55.5	94.0	38.5	58.2	21.6	3.5	55.0	1.1	3.6	0.32	0.33
8	9442	56.5	92.5	36.0	59.9	19.0	3.2	57.9	1.2	4.5	0.27	0.51
9	205925	57.0	95.5	38.5	63.8	29.1	4.2	44.4	0.8	3.1	0.25	0.50
10	<b>DZ-Cr-2675‡</b>	57.5	97.0	39.5	63.5	26.8	3.8	42.2	1.4	5.2	0.26	0.50
11	222108	69.0	95.7	26.7	74.0	32.2	2.9	32.5	0.9	5.4	0.17	0.47
12	9440	57.5	92.5	35.0	63.9	28.5	5.1	41.7	0.8	5.6	0.12	0.30
13	213231	59.0	95.0	36.0	68.3	26.6	3.3	39.8	0.7	4.3	0.18	0.44
14	acc-18-rm	59.5	92.0	32.5	76.0	30.6	4.0	45.2	0.9	3.6	0.24	0.40
15	205924	57.5	90.0	32.5	59.8	23.9	3.4	67.3	1.0	4.1	0.25	0.26
16	9427	59.5	92.5	33.0	59.4	26.1	3.6	52.6	0.9	5.0	0.17	0.52
17	DZ-Cr-44	56.5	91.5	35.0	71.6	27.5	3.2	32.1	1.2	4.8	0.25	0.83
18	207832	55.0	84.5	29.5	48.7	18.7	3.7	56.7	1.2	4.3	0.29	0.23
19	DZ-01-99	59.0	92.5	33.5	58.5	23.9	5.4	55.4	1.3	5.1	0.24	0.42
20	9443	57.0	94.0	37.0	64.9	28.2	3.0	51.0	1.2	5.1	0.25	0.73
21	207840	63.5	92.5	29.0	68.9	28.8	4.0	32.5	0.9	5.2	0.18	0.41
22	<b>DZ-Cr-385</b>	47.5	85.5	38.0	59.3	24.8	3.8	55.4	1.6	5.3	0.30	0.62
23	<b>Curati</b>	68.5	98.0	29.5	80.6	28.9	2.5	32.9	1.3	4.5	0.28	0.45
24	206442	68.0	89.0	21.0	69.5	31.4	3.1	40.4	1.2	5.7	0.21	0.46

† DH, days to 50% heading, DM, days to 75% maturity; GFP, grain filling period; PLHT, plant height; PNLG, panicle length; NPT, number of productive tillers per plant; LODG, lodging index; GYLD, grain yield; BYLD, biomass yield; HI, harvest index; PSW, panicle seed weight.

‡ Bold text denotes the top ten high yielding genotypes under moisture stress condition

## Appendix 3.5 Continued

No.	Genotype	Traits										
		DH	DM	GFP	PLHT	PNLG	NPT	LODG	GYLD	BMULD	HI	PSW
25	<b>215678</b>	58.5	94.0	35.5	68.2	25.5	4.2	51.9	1.3	5.9	0.23	0.88
26	222077	53.5	86.0	32.5	48.8	18.8	2.5	55.7	1.0	4.9	0.20	0.33
27	9408	58.0	92.5	34.5	62.4	26.3	4.5	47.7	0.8	4.9	0.17	0.22
28	<b>DZ-Cr-37</b>	55.0	87.5	32.5	58.4	23.0	2.9	47.9	1.5	5.2	0.30	0.62
29	222107	59.0	95.5	36.5	63.4	26.4	3.5	48.3	0.9	4.9	0.19	0.62
30	205891	58.0	91.0	33.0	58.2	23.3	2.6	51.4	1.1	4.8	0.21	0.32
31	207831	57.0	87.0	30.0	52.2	21.0	4.3	48.5	0.9	3.7	0.25	0.49
32	DZ-01-146	57.0	89.0	32.0	53.4	22.2	3.5	56.3	1.2	4.6	0.25	0.75
33	DZ-01-1821	59.5	95.5	36.0	66.2	26.9	3.8	39.2	0.9	6.1	0.15	0.87
34	9413	56.5	90.0	33.5	49.4	21.2	3.2	52.1	0.9	3.8	0.23	0.59
35	205944	63.0	96.5	33.5	65.3	24.4	3.6	45.1	1.1	3.8	0.29	0.48
36	DZ-01-409	55.5	88.7	33.2	56.4	23.4	3.4	40.0	1.2	4.4	0.25	0.72
37	9422	54.5	93.0	38.5	60.2	23.8	3.8	48.0	0.6	4.1	0.16	0.49
38	Manya	70.5	99.2	28.7	63.1	23.9	4.2	29.3	1.2	5.8	0.20	0.67
39	205915	53.5	96.5	43.0	58.5	22.4	4.2	66.3	0.7	5.2	0.14	0.59
40	DZ-01-899	60.5	95.5	35.0	63.1	25.0	3.1	44.6	0.8	5.2	0.16	0.78
41	Fesho	55.0	87.0	32.0	44.7	20.3	4.6	57.5	0.5	5.0	0.11	0.73
42	DZ-Cr-387RIL273	58.0	94.0	36.0	63.3	21.3	3.6	66.7	1.3	6.3	0.23	0.91
43	DZ-01-2423	57.0	92.5	35.5	60.5	23.4	3.5	36.3	1.2	4.9	0.24	0.66
44	acc-15-ck	71.0	89.0	18.0	59.0	26.6	3.7	31.7	1.0	4.2	0.24	0.57
45	Ho-Cr-136	59.5	84.5	25.0	57.2	22.1	3.0	35.8	1.2	4.0	0.31	0.44
46	PGRC/E 205396	55.5	91.0	35.5	62.3	22.9	2.9	59.2	1.1	4.7	0.24	0.55
47	205943	59.5	90.0	30.5	58.3	26.1	3.7	32.5	0.9	5.1	0.22	0.43
48	205896	58.5	97.5	39.0	61.7	26.5	3.5	35.8	1.2	6.6	0.19	0.67

## Appendix 3.5 Continued

No.	Genotype	Traits										
		DH	DM	GFP	PLHT	PNLG	NPT	LODG	GYLD	BMYLD	HI	PSW
49	<b>Dabbi</b>	58.5	86.5	28.0	56.2	23.9	2.7	58.4	1.3	4.1	0.34	0.33
50	205899	57.0	94.5	37.5	56.3	24.2	3.8	51.7	0.8	4.4	0.21	0.80
51	acc-01-dib	68.0	97.0	29.0	59.9	25.1	3.1	35.4	1.2	4.7	0.25	0.89
52	9414	58.5	93.5	35.0	68.7	23.7	4.0	52.0	1.1	5.2	0.20	0.80
53	<b>9423</b>	60.0	95.0	35.0	64.1	24.0	3.9	50.0	1.4	5.8	0.24	0.60
54	222078	58.5	93.5	35.0	66.6	24.1	3.8	39.3	1.2	5.9	0.19	0.84
55	207835	57.5	89.0	31.5	53.2	21.5	4.0	59.6	1.0	3.9	0.24	0.32
56	<b>DZ-01-2053</b>	55.0	87.0	32.0	55.5	18.3	3.8	47.5	1.4	4.8	0.29	0.34
57	221704	57.0	87.1	30.1	55.3	21.3	2.9	60.0	1.0	5.2	0.21	0.75
58	205898	57.5	92.5	35.0	58.2	25.6	3.0	50.8	1.2	4.6	0.25	0.58
59	DZ-Cr-358	55.0	90.0	35.0	61.7	26.2	2.7	50.8	1.0	5.4	0.20	0.48
60	219878	57.5	95.0	37.5	55.0	24.4	4.0	47.9	0.7	3.8	0.17	0.26
61	207839	58.0	93.5	35.5	66.4	26.7	3.2	39.6	0.9	4.9	0.21	0.50
62	Kaye-Agachew	54.5	99.0	44.5	63.3	21.7	5.1	60.4	1.1	5.7	0.19	0.49
63	DZ-01-1880	55.5	90.5	35.0	66.5	25.4	3.3	33.8	1.2	5.5	0.22	0.51
64	9396	56.5	93.0	36.5	64.1	25.7	3.8	41.3	0.8	5.2	0.14	0.45
65	Gea-Lamie	57.0	95.0	38.0	58.6	20.2	3.6	36.8	1.0	5.0	0.19	0.48
66	9419	59.0	92.5	33.5	68.3	27.9	4.8	52.6	0.9	5.5	0.15	0.51
67	Rosea	54.0	87.5	33.5	60.0	24.3	3.1	51.4	1.3	4.8	0.26	0.29
68	<b>9438</b>	57.5	91.5	34.0	53.1	22.0	4.1	45.2	1.4	4.2	0.32	0.70
69	DZ-Cr-82	54.0	91.5	37.5	71.7	25.3	4.7	38.5	0.7	3.3	0.21	0.82
70	DZ-01-1281	52.0	87.0	35.0	63.3	24.5	2.6	50.4	1.2	4.3	0.27	0.50
71	219880	57.0	90.0	33.0	51.4	21.8	3.2	54.9	1.3	4.4	0.33	0.54
72	219877	59.5	93.0	33.5	63.9	28.1	4.5	44.9	0.9	4.6	0.21	0.58

## Appendix 3.5 Continued

No.	Genotype	Traits										
		DH	DM	GFP	PLHT	PNLG	NPT	LODG	GYLD	BMYPD	HI	PSW
73	DZ-01-1681	57.0	93.0	36.0	65.7	28.4	4.5	33.2	0.8	5.4	0.15	0.55
74	Shawa-Gemerra	54.0	84.5	30.5	57.0	23.2	3.2	30.4	0.9	3.3	0.28	0.33
75	9433	60.0	95.0	35.0	61.2	27.5	2.6	41.1	0.6	3.7	0.17	0.38
76	Zezew	59.5	91.0	31.5	63.8	27.8	3.7	59.2	1.1	4.5	0.26	0.62
77	Zagure	56.5	87.5	31.0	48.3	23.7	4.5	28.0	1.3	4.5	0.29	0.31
78	9432	56.5	96.2	39.7	59.7	25.3	4.1	49.8	1.0	6.4	0.16	0.84
79	205919	56.0	91.0	35.0	65.6	27.1	4.4	50.7	0.8	5.5	0.15	0.61
80	Rubicunda	59.5	92.5	33.0	66.4	26.0	3.5	58.4	0.9	4.5	0.21	0.56
81	205927	60.0	91.0	31.0	67.4	25.1	4.0	62.3	1.0	5.1	0.21	0.43
82	219849	58.0	90.0	32.0	60.4	26.3	6.0	53.5	0.7	4.4	0.16	0.25
83	206745	64.5	96.0	31.5	66.1	26.2	4.1	49.5	1.0	5.4	0.17	0.41
84	9430	60.0	94.0	34.0	65.6	22.4	3.5	38.8	0.8	5.4	0.14	0.39
85	9435	58.0	93.0	35.0	61.4	25.7	4.2	42.9	0.8	5.0	0.15	0.36
86	DZ-01-196	57.0	95.5	38.5	59.2	26.3	2.7	55.6	1.3	5.0	0.30	0.69
87	207827	60.5	91.0	30.5	63.2	23.1	2.1	57.5	0.7	4.4	0.17	0.49
88	Ada	56.0	88.0	32.0	55.1	21.6	3.5	41.2	0.8	5.1	0.16	0.34
89	Enatite	53.5	88.5	35.0	55.8	22.4	2.3	46.4	0.8	3.8	0.21	0.63
90	205921	63.0	97.0	34.0	63.7	29.1	3.4	39.4	0.7	3.3	0.21	0.62
91	DZ-01-1278	57.0	91.5	34.5	68.6	29.8	3.9	61.0	1.0	4.7	0.22	0.83
92	DZ-Cr-354	58.0	91.5	33.5	71.1	29.3	3.5	51.6	0.9	5.3	0.16	0.69
93	9446	58.0	92.0	34.0	51.8	23.7	3.6	44.1	0.8	4.6	0.17	0.50
94	213237	58.5	97.0	38.5	63.9	29.4	2.9	44.8	1.2	5.9	0.19	0.49
95	207833	57.5	89.5	32.0	59.4	22.0	5.0	47.9	1.0	5.3	0.20	0.74
96	222072	60.5	94.0	33.5	71.4	27.0	2.8	37.9	0.8	4.4	0.17	0.42

## Appendix 3.5 Continued

No.	Genotype	Traits										
		DH	DM	GFP	PLHT	PNLG	NPT	LODG	GYLD	BMYLD	HI	PSW
97	9441	55.5	89.0	33.5	53.0	24.0	3.4	48.3	1.0	5.0	0.20	0.46
98	DZ-01-974	58.0	94.2	36.2	72.7	28.3	3.5	46.0	0.7	5.0	0.16	0.53
99	9424	58.0	91.5	33.5	63.9	26.8	3.2	48.5	1.0	4.6	0.21	0.49
100	DZ-Cr-387-RIL127	59.0	88.6	29.6	65.8	28.6	3.3	35.6	1.0	5.1	0.20	0.88
101	206739	60.5	97.0	36.5	67.6	27.9	4.1	52.6	1.1	3.6	0.31	0.52
102	DZ-01-1868	56.0	96.5	40.5	68.4	26.8	3.9	48.0	1.0	5.1	0.21	0.76
103	Acc.205953	62.5	91.5	29.0	72.9	29.4	4.8	48.1	1.2	5.2	0.23	0.66
104	219858	55.5	91.0	35.5	52.2	21.4	3.6	45.8	0.7	5.5	0.13	0.63
105	Kaye-Murri	58.0	99.0	41.0	66.0	25.7	3.3	44.5	1.1	6.5	0.18	0.84
106	9393	58.0	92.5	34.5	57.1	23.5	4.1	42.3	0.7	4.7	0.15	0.45
107	205923	58.0	93.5	35.5	57.9	21.2	3.2	56.7	0.7	4.7	0.16	0.32
108	DZ-01-787	61.5	94.5	33.0	67.3	31.4	4.5	51.4	0.7	6.1	0.12	0.64
109	acc-16-ck	60.0	93.5	33.5	54.7	24.2	3.6	51.4	0.7	5.3	0.14	0.30
110	205908	60.5	96.5	36.0	55.9	22.1	3.2	32.1	0.5	3.1	0.17	0.54
111	219881	55.0	92.5	37.5	53.4	22.5	2.8	45.0	0.7	4.5	0.16	0.55
112	DZ-01-2054	56.5	93.0	36.5	64.1	27.9	4.1	43.8	0.8	4.6	0.17	0.73
113	219860	60.0	90.5	30.5	64.9	24.7	4.6	45.4	0.9	4.0	0.25	0.76
114	207828	57.0	86.0	29.0	52.1	25.3	2.7	46.9	0.9	4.9	0.21	0.62
115	Denkeye	60.0	90.2	30.2	64.5	26.7	4.7	54.9	0.7	4.0	0.19	0.30
116	9400	57.5	93.0	35.5	66.2	27.0	4.2	32.1	1.1	5.0	0.21	0.49
117	207844	57.0	91.5	34.5	56.1	24.7	4.0	51.3	1.1	4.4	0.27	0.34
118	205931	57.0	89.0	32.0	50.5	21.9	3.2	46.4	0.8	4.6	0.18	0.23
119	Jano	69.0	97.0	28.0	67.9	26.1	3.7	35.4	0.9	5.1	0.18	0.90
120	acc-04-dib	59.5	92.0	32.5	71.5	32.3	3.4	38.5	1.3	6.3	0.21	0.60

## Appendix 3.5 Continued

No.	Genotype	Traits										
		DH	DM	GFP	PLHT	PNLG	NPT	LODG	GYLD	BMYLD	HI	PSW
121	DZ-01-3186	58.0	96.0	38.0	62.7	27.1	4.3	40.6	1.0	6.1	0.17	0.93
122	9415	59.0	93.5	34.5	64.1	28.9	3.6	49.8	1.2	5.7	0.22	0.89
123	acc-17-wj	55.0	88.5	33.5	49.4	21.6	3.3	59.5	0.6	4.7	0.14	0.45
124	205917	61.0	89.5	28.5	68.5	27.8	4.0	33.5	1.0	5.9	0.18	0.88
125	9416	56.5	90.5	34.0	59.4	21.8	3.1	47.5	1.0	3.7	0.28	0.52
126	219865	55.0	85.0	30.0	48.6	20.3	3.4	54.8	0.8	4.5	0.19	0.30
127	219852	56.5	93.0	36.5	60.6	26.8	3.1	37.6	0.8	5.2	0.15	0.23
128	219871	59.0	95.5	36.5	57.3	24.7	4.5	53.5	0.6	4.7	0.14	0.48
129	Gorradie	56.0	93.0	37.0	59.3	21.1	2.9	42.8	0.7	4.6	0.19	0.63
130	206750	54.5	90.0	35.5	45.0	20.3	3.5	49.5	0.6	4.7	0.13	0.80
131	219863	57.5	85.5	28.0	63.2	25.1	2.4	42.1	0.9	4.1	0.22	0.48
132	Purpurea	60.0	99.0	39.0	65.1	27.7	4.1	52.9	1.0	6.4	0.17	0.90
133	<b>Dschanger</b>	56.0	97.0	41.0	61.6	23.6	3.2	38.8	1.6	6.0	0.27	0.86
134	9395	58.5	93.5	35.0	62.8	26.7	3.9	44.8	0.7	6.1	0.12	0.62
135	205913	58.5	95.0	36.5	64.3	25.3	3.7	32.1	0.6	4.8	0.12	0.54
136	205936	56.5	93.0	36.5	57.9	22.8	3.6	47.3	0.7	4.6	0.19	0.45
137	222076	54.5	85.0	30.5	45.4	17.9	3.9	61.2	0.8	2.9	0.27	0.36
138	acc-02-dib	58.5	90.0	31.5	67.4	28.6	3.4	44.5	0.7	4.5	0.16	0.41
139	205922	59.0	91.5	32.5	71.8	31.9	3.9	31.6	0.6	6.2	0.10	0.76
140	207575	60.5	95.0	34.5	62.8	27.2	2.4	45.8	0.6	5.0	0.13	0.43
141	Alba	55.5	92.5	37.0	66.6	20.1	3.5	33.3	0.9	4.6	0.19	0.48
142	206753	56.5	87.5	31.0	50.1	21.3	4.0	63.8	0.8	4.6	0.19	0.46
143	DZ-Cr-255	69.5	97.5	28.0	69.3	27.2	3.1	39.3	0.7	5.6	0.13	0.85
144	DZ-01-1285	52.0	94.2	42.2	64.8	22.9	2.3	49.9	0.8	4.5	0.18	0.68

Appendix 3.6 Mean, minimum, maximum, standard error, mean squares and coefficient of variation for 11 traits of 144 tef genotypes evaluated at three non-stressed sites

No.	Statistics	Traits										
		DH†	DM†	GFP†	PLHT†	PNLG†	NPT†	LODG†	GYLD†	BYLD†	HI†	PSW†
1	Mean	58.3	92.2	33.9	61.5	25.0	3.6	46.8	1.0	4.9	0.21	0.57
2	Minimum	47.5	84.5	18.0	44.7	17.9	2.1	28.0	0.5	2.9	0.1	0.22
3	Maximum	71.0	99.2	44.5	80.6	32.3	6.0	67.3	1.6	7.0	0.34	0.98
4	Standard error	1.3	1.8	2.1	3.9	1.8	0.8	6.8	0.1	0.2	0.04	0.1
5	Genotype MS‡	27.7**	23.6**	29.8**	86.1**	17.9**	0.9Ns¶	145.6**	0.1**	1.1Ns¶	0.01**	0.01**
6	CV (%)§	3.2	2.7	8.6	9.0	10.2	30.4	20.5	16.8	19.6	26.1	28.0

† DH, days to 50% heading, DM, days to 75% maturity; GFP, grain filling period; PLHT, plant height; PNLG, panicle length; NPT, number of productive tillers per plant; LODG, lodging index; GYLD, grain yield; BYLD, biomass yield; HI, harvest index; PSW, panicle seed weight

‡MS = mean squares

§CV, coefficient of variation

¶Ns, non-significant

## Chapter 4: **Genetic Variation and Trait Association of Tef [*Eragrostis tef* (Zucc.)Trotter] Evaluated under Optimal and Moisture-Stressed Environments**

### **Abstract**

Tef [*Eragrostis tef* (Zucc.) Trotter] is one of the major cereal crops grown in the Horn of Africa for food and as an export commodity for its unique nutritional qualities. Moisture stress is the leading yield limiting factor of tef production in northern Ethiopia. The objectives of this study were to assess the genetic variability present among 144 tef genotypes of varied population and to identify important agronomic traits with high heritability and correlations for effective breeding. The genotypes were evaluated using four experiments laid down in a 12 x 12 lattice design under moisture stressed and non-stressed conditions in northern Ethiopia. Main shoot panicle seed weight had high genotypic coefficients of variation (GCV) of 22.4% and 25.9% under non-stressed and stressed conditions, respectively. Grain yield had GCV values of 17.6% and 20.0% in the corresponding environments. Heritability was higher under optimum condition than the moisture stressed, which is valuable to conduct effective selection. A path-coefficient analysis indicated that direct selection for high biomass, harvest index and late maturity could increase grain yield under optimal conditions, while under moisture stress conditions early maturity, high biomass and harvest index were important direct selection criteria for tef breeding aiming for drought tolerance.

**Keywords:** Correlation, Ethiopia, genetic advance, heritability, path-analysis

## 4.1 Introduction

Tef Tef [*Eragrostis tef* (Zucc.) Trotter] is the principal food crop supporting more than 50 million people in the Horn of Africa. Further, it is increasingly used in other parts of the world for gluten-free food products (Assefa et al., 2011). In Ethiopia, tef is the most widely grown crop and its production area has expanded from 1.99 million ha in 2004 (CSA, 2004) to 3.02 million ha in 2015 (CSA, 2015), a 51% increase. Correspondingly, total grain production of tef has increased from 1.67 million tons to 47 million tons, translating a productivity of 0.84 to 1.58t ha<sup>-1</sup> (CSA, 2015). In Ethiopia tef yields are significantly low compared to maize, sorghum and wheat yields of 3.43, 2.37 and 2.54 t ha<sup>-1</sup>, respectively (CSA, 2015). Low yields of tef are attributed to its susceptibility to lodging, moisture stress, seed shattering, and poor pre- and post-harvest agronomic systems (Assefa et al., 2011).

Tef varieties with high grain yield, but high tolerance of moisture stress and lodging are the major goals of plant breeders. This could be achieved through exploitation of the genetic variability present in tef germplasm, especially using landraces for their unique traits. The progress of selection in crop improvement programs depends on the extent and magnitude of genetic variation (Aniol, 2001). Previous genetic variability studies (Assefa et al., 1999, 2000, 2001; Admas and Belay, 2011; Ayalew et al., 2011; Shiferaw et al., 2012) reported a wide variability in yield and yield components in tef germplasm in Ethiopia.

The magnitude of heritability and the correlation of traits determine genetic advancement through direct or indirect selection (Falconer and Mackay, 1996; Sleper and Poehlman, 2006). This is particularly important when the heritability of traits is low for effective selection (Singh and Chaudhary, 1977; Dabholkar, 1992). Previous studies (Admas and Belay, 2011; Assefa et al., 2001; Ayalew et al., 2011; Chanyalew, 2010; Debebe et al., 2013) indicated that heritability, genetic advance and correlation of traits of tef genotypes are variable depending upon the test populations and test environments. Therefore, identification of highly heritable and correlated traits in the targeted environment is important for maximum selection response in tef breeding.

Path analysis is one of the powerful statistical tools that permit the measurement of the direct influence of one variable upon another. It helps to identify the most influential predictor variable useful for simultaneous selection (Singh and Chaudhary, 1977; Dabholkar, 1992). Previous path analysis studies in tef reported that harvest index and biomass yield had strong direct effects on grain yield, indicating the importance of these traits for selection for high grain yield under optimal conditions (Ayalew et al., 2011; Ayalneh et al., 2012; Debebe et al., 2013).

A recently released tef variety, namely, DZ-Cr-387 (Quncho), has been promoted nationally for its farmers preferred traits such as high grain yield, white seed color, high biomass yield and good '*injera*' making quality. However, this variety has poor performance and was poorly adopted by tef growers in moisture stressed environments in Ethiopia. Targeted breeding of tef for drought prone environments is needed in Ethiopia. Furthermore, grain yield and quality traits such as white seed colour and good *injera* qualities are important attributes for tef breeding. Therefore, the objectives of this study were to assess the genetic variability present among a diverse tef population and to identify important agronomic traits with high heritability and correlations for effective breeding in moisture stressed environments.

## **4.2 Materials and methods**

### **4.2.1 Description of the study environments**

The experiments were conducted in four environments as described in section 3.2.1

### **4.2.2 Plant materials, experimental design and trial management**

The plant materials, experimental design and managements used for this study were as described in sections 3.2.2 and 3.2.3

### **4.2.3 Data collection**

Data were collected as the procedure described in section 3.2.4

### **4.2.4 Data analysis**

Data collected across the three environments; under two optimal rainfall and a full irrigation regime were regarded as non-stressed environments and the other test involving irrigation being withheld at panicle emergence being considered as moisture stressed environment. Data of the 144 genotypes, grown in the non-stressed and stressed environments, were subjected to statistical analysis using the simple lattice procedure of SAS 9.3 (SAS Institute, 2011). Combined analyses of variance over non-stressed environments were carried out after Bartlett's homogeneity of variance test procedure. The expected mean squares from the analysis of variance were used to estimate the phenotypic coefficient of variance (PCV) and genotypic coefficient of variance (GCV), heritability (H) and genetic advance (GAM). Associations among the 11 traits were assessed, based on means, using the Pearson's correlation procedure of SAS 9.3 (SAS Institute, 2011). Path analysis was conducted according to the procedure by Dewey and Lu (1959).

### 4.3 Results and discussion

#### 4.3.1 Genotypic and phenotypic coefficient of variation, broad sense heritability and genetic advance

Genotypic coefficient of variance (GCV) and phenotypic coefficient of variance (PCV), heritability and genetic advance (GAM) for the 11 quantitative traits of the 144 tef genotypes tested under optimal and moisture stressed environments are presented in Table 4.1. Highest GCV values were observed for the main shoot panicle seed weight, at 22.4% and 25.9%, and for grain yield at 17.6 % and 20.0%, in the non-stressed and stressed environments, respectively. The trends of GCV values of grain yield in the stressed and non-stressed environments observed in the present study are in agreement with the findings of Admas and Belay (2011). The PCV values were 25.9% and 20.3% for main shoot panicle seed weight and grain yield, respectively, in the non-stressed environments. Under moisture stressed conditions PCV values of 37.9%, 30.3%, 28.3% and 26.2% were recorded for main shoot panicle seed weight, harvest index, number of productive tillers per plant and grain yield, respectively. Generally, GCV ranged from 4.3 to 22.4% and 3.2 to 25.9%, while PCV varied from 4.6 to 25.9% and 4.2 to 37.9% in the non-stressed and moisture stressed environments, respectively (Table 4.1). These trends were in agreement with the report of Ayalew et al. (2011) in their evaluation of tef germplasm collected from Amhara region. However, the current range of variability was smaller than that reported by Chanyalew (2010), who found GCV and PCV values ranging from 4.2 to 54.5% and 10.5 to 51.0%, respectively, when testing different sets of tef genotypes at Debre-Zeit and Melkasa in Ethiopia.

The greatest difference between GCV and PCV values were observed in the moisture stressed environment for the majority of the traits. This was especially high for the number of productive tillers per plant, which could be attributed to the high environmental variance in the stressed environments affecting genetic variability (Table 4.1). Similarly, larger differences in GCV and PCV values were reported for the number of productive tillers in stressed than non-stressed environments (Admas and Belay, 2011).

Heritability of 87.1%, 78.5%, 76.0%, 75.5%, 75.0% and 70.7% were observed for days to 75% maturity, plant height, days to panicle emergence, grain yield, main shoot panicle seed weight and panicle length, respectively, under non-stressed conditions. However, heritability of 77.3%, 57.5%, 56.1% and 58.7% were estimated for days to panicle emergence, days to 75% maturity, grain filling period and grain yield respectively in the stressed environment (Table 4.1). Chanyalew (2010) reported intermediate heritability of 50.5% for grain yield of

tef genotypes. Similarly, Ayalew et al. (2011) recorded intermediate heritability values for days to panicle emergence (80.7%), culm length (72.4%), days to maturity (65%), plant height (64.3%), grain filling period (61.6%) and grain yield (57.3%) for tef landraces collected from the Amhara region. Mewa et al. (2013) reported intermediate to high heritability (65.9% to 86.5%) for grain yield of recombinant inbred lines derived from a cross of the tef variety DZ-01-974 with *Eragrostis pilosa*, tested at two locations. Conversely, low heritability values of 25.3% and 25.7% were reported by Assefa et al. (2000) and Assefa et al. (2001), respectively, for grain yield of tef landraces.

Generally, heritability was high for all traits under the non-stressed conditions but comparatively low under moisture stressed conditions (Table 4.1). Decreased heritability of grain yield under the stressed conditions than the non-stressed was also reported by Admas and Belay (2011) and Shiferaw et al. (2012).

Information on the amount of genetic advance that could be achieved under selection is valuable in plant breeding programs. Main shoot panicle seed weight with 40.0% and 36.4% followed by grain yield of 31.5% and 31.6% had higher rates of genetic advance in the non-stressed and stressed environments, respectively (Table 4.1). High genetic advance translates to more progress from plant breeding selection. Assefa et al. (1999) and Ayalew et al. (2011) reported a genetic advance of 24.7% and 24%, respectively, for grain yield of tef landraces. Chanyalew (2010) estimated a genetic advance of 31.3% and 47.9% for grain yield and main shoot panicle seed weight, respectively. In contrast, Assefa et al. (2001) indicated a low genetic advance of 14.5% for grain yield of tef landraces collected from eight regions in Ethiopia.

Table 4.1 Genotypic coefficients of variance (GCV) and phenotypic coefficients of variance (PCV), broad sense heritability (H) and genetic advance (GAM) as percent of mean for 11 agronomic traits of the 144 tef genotypes tested under moisture stressed and non-stressed conditions

Traits	Non-Stressed				Stressed			
	GCV (%)	PCV (%)	H (%)	GAM	GCV (%)	PCV (%)	H (%)	GAM
DPE	6.9	7.9	76.0	12.3	5.6	6.8	77.3	10.8
DM	4.3	4.6	87.1	8.2	3.2	4.2	57.5	5.0
GFP	5.9	7.6	60.2	9.4	9.7	12.9	56.1	14.9
PLHT	7.3	8.2	78.5	13.2	8.1	12.1	44.7	11.1
PNLG	8.4	10.0	70.7	14.6	9.5	14.2	45.1	13.1
NPT	5.6	16.5	11.4	3.9	-	28.3	-	-
LODG	10.6	14.0	56.8	16.4	11.1	23.3	22.5	10.8
GYLD	17.6	20.3	75.5	31.5	20.0	26.2	58.7	31.6
BMYLD	8.5	12.3	47.7	12.1	6.2	20.6	8.9	3.8
HI	5.4	12.0	20.0	4.9	16.2	30.3	28.4	17.7
PSW	22.4	25.9	75.0	40.0	25.9	37.9	46.7	36.4

DPE = days to panicle emergence, DM = days to 75% maturity; GFP = grain filling period; PLHT = plant height; PNLG = panicle length; NPT = number of tillers per plant; LODG = lodging index; GYLD = grain yield; BYLD = biomass yield; HI = harvest index; PSW = main shoot panicle seed weight

#### 4.3.2 Correlations of yield and its components

Genetic correlation coefficients among the 11 traits grown under non-stressed and stressed conditions are presented in Table 4.2. Grain yield showed significant ( $p \leq 0.01$ ) positive correlation with biomass yield ( $r=0.77$ ), harvest index (0.73) and main shoot panicle seed weight (0.66) in the non-stressed environments. Days to panicle emergence (0.46), days to maturity (0.50), plant height (0.50) and spike length (0.63) were also positively associated with grain yield. Under moisture stressed condition, grain yield showed significant association with biomass yield (0.24) and harvest index (0.75) only (Table 4.2). This correlation could be due to linkage or pleiotropic genetic effects causing the traits to change in the same direction (Falconer and Mackay, 1996). Chanyalew (2010) reported positive correlation of grain yield with the majority of the traits tested, while negative correlations were recorded with harvest index and lodging index for tef genotypes tested at two locations. Lule and Mengistu (2014) reported a positive association of grain yield with harvest index but a negative association with biomass yield of tef landraces collected from different zones of Ethiopia.

Plant height and panicle length showed positive associations with grain yield. However, lodging index was negatively correlated with these traits (Table 4.2). Interestingly, this may not translate into a high harvest index and reduced lodging, which have been reported as important traits of semi-dwarf varieties of small cereals such as wheat, barley and rice.

Therefore, development of considerably dwarf tef varieties should not be the goal of tef breeding to enhance grain yields and reduce effect of lodging. In Ethiopia, relatively tall tef varieties are desired by farmers because tef is highly valued for its straw yield as a major source of animal feed (Yami, 2013). Tall tef varieties have relatively thick stems. Tef has a shallow root system and is sensitive to lodging (Van Delden et al., 2010). Late maturing and tall tef varieties possess deeper root systems than early maturing genotypes that have shorter plant heights (Ayele et al., 2001). Therefore, breeding tef varieties with a good stem thickness and improved root depth could offer high adoption rate of tef varieties by farmers than breeding dwarf varieties to reduce lodging.

Number of productive tillers per plant revealed a significant ( $p \leq 0.01$ ) negative association ( $r = -0.30$ ) with grain yield in the non-stressed environment and a non-significant negative association ( $r=-0.11$ ) in the stressed environment (Table 4.2). Unlike these findings, Ayalew et al. (2011) reported a positive correlation between the number of productive tillers per plant with grain yield. Days to 75% maturity was positively associated with grain yield ( $r=0.50$ ) in the non-stressed environment, while there was a non-significant negative association ( $r=-0.06$ ) in the stressed environment, supporting the importance of early maturity in moisture stressed environments (Table 4.2). Similarly, Tefera et al. (2003) and Plaza-Wuthrich et al. (2013) found negative correlations between grain yield and days to maturity in tef recombinant inbred lines, and landraces, respectively.

Table 4.2 Genotypic correlations coefficients for 11 quantitative traits of 144 tef genotypes tested in moisture stressed and non-stressed environments

Traits	Env.	Traits										
		DPE	DM	GFP	PLHT	SPLG	NPT	LODG	GYLD	BMYLE	HI	PSW
DPE	N	1.00										
	S	1.00										
DM	N	0.73**	1.00									
	S	0.44**	1.00									
GFP	N	-0.33**	0.41**	1.00								
	S	-0.58**	0.47**	1.00								
PLHT	N	0.56**	0.70**	0.21*	1.00							
	S	0.46**	0.52**	0.02ns	1.00							
SPLG	N	0.53**	0.69**	0.25**	0.78**	1.00						
	S	0.46**	0.40**	-0.09ns	0.76**	1.00						
NPT	N	-0.28**	-0.28**	-0.02ns	-0.31**	-0.36**	1.00					
	S	-0.03ns	0.07ns	0.09ns	0.03ns	0.08ns	1.00					
LODG	N	-0.45**	-0.52**	-0.12ns	-0.45**	-0.52**	0.06ns	1.00				
	S	-0.33**	-0.20*	0.14ns	-0.32**	-0.34**	0.08ns	1.00				
GYLD	N	0.46**	0.50**	0.08ns	0.50**	0.63**	-0.30**	-0.43**	1.00			
	S	0.01ns	-0.06ns	-0.06ns	0.09ns	-0.02ns	-0.11ns	0.05ns	1.00			
BMYLE	N	0.50**	0.49**	0.01ns	0.56**	0.57**	-0.25**	-0.37**	0.77**	1.00		
	S	0.19*	0.37**	0.15ns	0.32**	0.32**	0.08ns	-0.09ns	0.24**	1.00		
HI	N	0.17*	0.26**	0.14ns	0.18*	0.37**	-0.24**	-0.26**	0.73**	0.14ns	1.00	
	S	-0.10ns	-0.30**	-0.17*	-0.13ns	-0.22**	-0.15ns	0.11ns	0.75**	-0.41**	1.00	
PSW	N	0.47**	0.57**	0.17*	0.60**	0.66**	-0.35**	-0.34**	0.66**	0.54**	0.45**	1.00
	S	0.12ns	0.35**	0.20*	0.29**	0.21**	0.00ns	-0.11ns	0.19ns	0.43**	-0.08**	1.00

DPE = days to panicle emergence, DM = days to 75% maturity; GFP = grain filling period; PLHT = plant height; PNLG = panicle length; NPT = number of tillers per plant; LODG = lodging index; GYLD = grain yield; BYLD = biomass yield; HI = harvest index; PSW = main shoot panicle seed weight;

ns, \* and \*\* indicate non-significant, significant at  $p \leq 0.05$  and  $p \leq 0.01$ , respectively

N and S denote Non-stressed and Stressed environments, respectively; Env. = environments

### 4.3.3 Path coefficient analysis

Table 4.3 presents the direct and indirect effect of the 11 quantitative traits on grain yield under non-stressed and moisture stressed conditions. Biomass yield and harvest index showed strong positive effects of 0.66, 0.68 and 0.62, 1.03 on grain yield in both the non-stressed and stressed environments, respectively (Table 4.3). Therefore, selection for these characters would give good responses to yield improvement. Previously, Ayalew et al. (2011) reported biomass yield, number of productive tiller per plant and harvest index for their highest direct effect and their correlation with grain yield of tef landraces. The authors suggested that selecting for these traits indirectly selects for grain yield. Similarly, Ayalneh et al. (2012) and Debebe et al. (2013) reported that harvest index and biomass yield had a strong direct effect and positive correlation with grain yield in tef landraces.

Days to panicle emergence and grain filling period showed strong negative direct effects of -2.42 and -1.84 under the non-stressed conditions and strong positive direct effects of 2.31 and 2.38 under stressed conditions. Conversely, days to maturity had a strong positive direct effect in the non-stressed environment (2.50) and a strong negative effect in the stressed environment (-2.16). This indicates that late maturity tends to decrease grain yield performance during drought stress. Similarly, Admass and Belay (2011) found better grain yield performance of early maturing recombinant inbred lines than late maturing types in a moisture stressed environment. In addition to its direct effect, days to maturity showed relatively strong negative indirect effects via grain filling period, plant height, panicle length, number of productive tillers per plant, biomass yield and main shoot panicle seed weight in the stressed environment, and positive indirect effects via all traits except lodging index and number of productive tillers per plant in the non-stressed environments (Table 4.3).

Plant height and panicle length exerted weak direct effects in both environments. A significant positive association of these traits with grain yield under the non-stressed conditions was due to positive indirect effects via biomass yield and days to maturity. Similarly lodging index, number of productive tillers per plant and main shoot panicle seed weight had minimal direct effects under both environments (Table 4.3). Overall, the path analysis indicated selection for high biomass yield, harvest index and long maturity could provide increased grain yield in optimal environments. While in moisture stressed environments, yield improvement could be achieved through selection for reduced days to maturity, high biomass yield and harvest index.

Table 4.3 Direct (diagonal and bold faced scripts) and indirect (off diagonal) path coefficients of 11 quantitative traits of 144 tef genotypes tested under moisture stressed and non-stressed conditions

Trait	ENV	Traits										
		DPE	DM	GFP	PLHT	SPLG	NPT	LODG	BMYLD	HI	SWPP	rgGYLD
DPE	N	<b>-2.42</b>	1.81	0.61	0.00	0.01	0.00	0.01	0.33	0.11	0.01	0.46**
	S	<b>2.31</b>	-0.96	-1.38	0.03	-0.02	0.00	0.00	0.13	-0.11	0.00	0.01ns
DM	N	-1.75	<b>2.50</b>	-0.75	-0.01	0.02	0.00	0.01	0.32	0.17	0.01	0.50**
	S	1.02	<b>-2.16</b>	1.13	0.03	-0.02	0.00	0.00	0.25	-0.30	-0.01	-0.06ns
GFP	N	0.80	1.02	<b>-1.84</b>	0.00	0.01	0.00	0.00	0.00	0.09	0.00	0.08ns
	S	-1.34	-1.03	<b>2.38</b>	0.00	0.00	0.00	0.00	0.10	-0.17	-0.01	-0.06ns
PLHT	N	-1.36	1.74	-0.39	<b>-0.01</b>	0.02	-0.01	0.01	0.37	0.11	0.01	0.50**
	S	1.06	-1.12	0.05	<b>0.06</b>	-0.03	0.00	0.00	0.22	-0.14	-0.01	0.09ns
SPLG	N	-1.27	1.72	-0.45	-0.01	<b>0.02</b>	-0.01	0.01	0.38	0.23	0.01	0.63**
	S	1.07	-0.87	-0.22	0.05	<b>-0.04</b>	0.00	0.00	0.22	-0.23	-0.01	-0.02ns
NPT	N	0.67	-0.70	0.04	0.00	-0.01	<b>0.02</b>	0.00	-0.16	-0.15	0.00	-0.30**
	S	-0.06	-0.14	0.20	0.00	0.00	<b>0.00</b>	0.00	0.05	-0.16	0.00	-0.11ns
LODG	N	1.09	-1.30	0.21	0.00	-0.01	0.00	<b>-0.02</b>	-0.24	-0.17	0.00	-0.43**
	S	-0.77	0.44	0.34	-0.02	0.01	0.00	<b>-0.01</b>	-0.06	0.11	0.00	0.05ns
BMYLD	N	-1.21	1.22	-0.01	0.00	0.01	0.00	0.01	<b>0.66</b>	0.09	0.01	0.77**
	S	0.44	-0.80	0.35	0.02	-0.01	0.00	0.00	<b>0.68</b>	-0.43	-0.01	0.24**
HI	N	-0.41	0.66	-0.26	0.00	0.01	0.00	0.01	0.09	<b>0.62</b>	0.01	0.73**
	S	-0.24	0.64	-0.40	-0.01	0.01	0.00	0.00	-0.28	<b>1.03</b>	0.00	0.75**
SWPP	N	-1.12	1.43	-0.31	0.00	0.02	-0.01	0.01	0.36	0.28	<b>0.01</b>	0.66**
	S	0.29	-0.76	0.46	0.02	-0.01	0.00	0.00	0.30	-0.08	<b>-0.03</b>	0.19ns

DPE = days to panicle emergence, DM = days to 75% maturity; GFP = grain filling period; PLHT = plant height; PNLG = panicle length; NPT = number of tillers per plant; LODG = lodging index; GYLD = grain yield; BYLD = biomass yield; HI = harvest index; PSW = main shoot panicle seed weight; rgGYLD = genotypic correlation of grain yield; N, and S denote for Non-stressed and Stressed environments, respectively; ENV= environment

#### 4.4 Conclusions

The current study indicated that the existence of substantial genetic variability within tef genotypes screened in optimal and moisture stressed environments was useful for tef improvement. Heritability values of traits were higher in the optimal environments than they were in the moisture stressed environment due to the relatively high environmental variability. The relatively high genetic advance of grain yield and main shoot panicle seed weight under both optimum and moisture stressed conditions would be desirable for genetic gains through selection. Direct selection for high biomass, harvest index and late maturity could increase grain yield in optimal environments, while under moisture stress conditions early maturity, high biomass and harvest index were important direct selection criteria to use when breeding tef for drought tolerance. Due to the limited number of stressed environments used in the present study, further evaluation of the genotypes across more representative and water stressed agro-ecologies would be useful.

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## Chapter 5: Assessment of the Genetic Relationship of tef (*Eragrostis tef*) Genotypes using SSR Markers

### Abstract

Knowledge of the genetic relationships of plant genetic resources is fundamental for effective selection and conservation. Tef [*Eragrostis tef* (Zucc.) Trotter] is a gluten-free small cereal crop that exhibits considerable genetic variation. The objective of this study was to determine the genetic relationships among 60 diverse tef genotypes and to select unique and genetically unrelated lines by using 10 selected diagnostic and polymorphic simple sequence repeat (SSR) DNA markers. The results indicated that the number of alleles per locus varied from 10 to 23 with a mean of 16. The polymorphic information content (PIC) ranged from 0.64 for marker CNLTS11 to 0.94 (CNLTS136A/B) with a mean of 0.84, suggesting sufficient discrimination power of the markers to discriminate the tested genotypes. The analysis of molecular variance showed that 63% and 35% of the total variability could be attributed to differences within and among tef genotypes, respectively. The high level of genetic dissimilarity within the tested tef genotypes provides an opportunity for systematic selection and conservation. Overall, the SSR analysis identified distinct genotypes such as 'DZ-Cr-385', '222076' and '213237', which are known for their early maturity and good yields under moisture stress. The analysis identified the genotypes, 'DZ-Cr-387', '205896', '205917' and 'Dschanger', which are consistent with their unique agronomic attributes such as late maturity, high grain yields, relatively good plant heights and long panicles under optimum rainfall conditions. The identified and agronomical complementary tef genotypes are valuable genetic resources for further breeding.

**Keywords:** Genotype, genetic relatedness, landrace, simple sequence repeat markers, tef

## 5.1 Introduction

Systematic genetic characterization and well defined genetic relationships among plant genetic resources are fundamental for effective selection and conservation. This would allow for the identification of genetically unrelated and agronomical complementary genotypes for designed crosses and improved selection for important traits. Molecular markers are more efficient in germplasm characterization than phenotypic or biochemical markers (Aremu, 2011; Jonah et al., 2011; Ranade and Yadav, 2014; Jingura and Kamusoko, 2015).

Various molecular marker systems were used effectively to assess the genetic relationships and patterns of association among plant genetic resources. These included restricted fragment length polymorphism (RFLP), random amplified polymorphic DNA (RAPD), amplified fragment length polymorphism (AFLP), sequence characterized amplification regions (SCARS), simple sequence repeat (SSR) or microsatellites and single nucleotide polymorphism (SNP) (Datta et al., 2011; Govindaraj et al., 2015). SSR markers have been extensively used in genetic diversity analysis of different cereal crops including rice (Ishak et al., 2015; Sarma et al., 2015), wheat (Bafghi et al., 2014; Hamdalla, 2014; Drikvand et al., 2015), sorghum (Beyene et al., 2014) and barley (Hua et al., 2015). These markers are known for their reproducibility, multi-allelic nature, co-dominant inheritance, relative abundance and good genome coverage (Gyulai et al., 2006, 2012; Senan et al., 2014; Ali et al., 2014, 2015).

Tef [*Eragrostis tef* (Zucc.) Trotter] is an allotetraploid ( $2n=4x=40$ , AABB) autogamous small cereal crop widely cultivated in the horn of Africa (Eritrea and Ethiopia) for over 2,000 years (Ponti, 1978). In the region, tef supports some 60 million people providing food security and rural livelihoods. Tef products such as breakfast cereal, porridge, bread, pancake, waffles and juices are becoming popular worldwide for their gluten-free status which is helpful to people with gluten intolerance. Tef straw is also a valuable source of livestock feed. In South Africa, India, Pakistan, Uganda, Kenya and Mozambique tef is mainly grown as a forage or pasture crop (Assefa et al., 2011).

Ethiopia is believed to be the centre of origin and diversity of tef (Vavilov, 1951). In the country tef remains the number one crop in terms of area coverage with an estimated annual acreage of more than 3 million ha (CSA, 2014). Tef shows considerable phenotypic variation, with wide adaptation across a range of agro-ecologies (Assefa et al., 2015). However, tef yields are low, with a mean national yield of  $1.47 \text{ t ha}^{-1}$  (CSA, 2014). The low yield of tef is attributed to its susceptibility to lodging, frequent moisture stress and poor agronomic management with few

inputs (Ketema, 1997). Tef yield could be enhanced through selective breeding using locally adapted, farmer-preferred, genetically complementary lines. This requires a genetic diversity analysis using effective and diagnostic molecular markers, and evaluation of economic agronomic traits.

Several studies have been conducted to examine the diversity in tef germplasm using morphological and agronomic traits (Assefa et al., 2000, 2001a,b, 2002a,b, 2003b; Adnew et al., 2005; Admas and Belay, 2011; Ayalew et al., 2011; Shiferaw et al., 2012; Plaza-Wuthrich et al., 2013). However, few attempts have been undertaken to study the diversity using DNA markers. Ayele et al. (1999) by using AFLP markers, and Bai et al. (2000) by using RAPD markers reported genetic similarity coefficients of 85-90% and 84-96%, which indicated a high level of genetic similarity of the tested genotypes. Previous research (Assefa et al., 2003, Zeid et al., 2012) has shown the usefulness of SSR markers in genetic diversity analysis and in establishing genetic relationships among tef germplasm. Therefore, the objectives of this study were to determine the genetic relationship present among 60 diverse tef genotypes, and to select unique and genetically unrelated lines using 10 known diagnostic and polymorphic SSR markers.

## **5.2 Materials and methods**

### **5.2.1 Plant materials**

Sixty diverse tef genotypes were used for this study. Table 5.1 presents the names and origins of the genotypes. Thirty three genotypes were improved varieties that were released by six agricultural research centres (MoA, 2014), 18 were landraces identified by Ebba (1975), and nine were new accessions collected from the Tigray region of northern Ethiopia. A landrace refers to an early cultivated of a crop species, evolved from a wild population. It has historical origin, distinct identity but lacks formal crop improvement. Accession refers to the plant materials collected from a particular area. The lines identified by Ebba (1975) were originally collected from diverse tef growing zones in Ethiopia including Shoa (8), Gojam (3), Keffa (2), Welega (3) and Hararge (2) (Table 5.1). Photos of the popular landrace varieties '*Dschanger*' and '*Kaye-Murri*' are shown in Figure 5.1.



Figure 5.1 Pictures of the two landrace varieties evaluated in the study: ‘Dschanger’ (left) and ‘Kaye-Murri’ (right)

Table 5.1 List of the 60 tef [*Eragrostis tef* (Zucc.) Trotter] genotypes used in the present study

Populations	Collection zones or sources	Name of genotypes	
Landraces	Shoa	‘Ada’, ‘Rosea’, ‘Fesho’, ‘Kaye-Agachew’, ‘Purpurea’, ‘Manya’, ‘Enatite’, ‘Rubicunda’	
	Welega	‘Gea-Lamie’, ‘Alba’, ‘Kaye-Murri’,	
	Hararge	‘Gorradie’, ‘Denkeye’	
	Gojam	‘Dabbi’, ‘Curati’, ‘Jano’	
New accessions	Keffa	‘Shawa-Gemerra’, ‘Dschanger’	
	West Tigray	‘9446’	
	South Tigray	‘213237’	
	South east Tigray	‘205921’, ‘205896’, ‘205917’	
Improved varieties	Central Tigray	‘222076’, ‘9415’, ‘Zezew’, ‘Zagure’	
	DZARC	‘DZ-Cr-387’, ‘DZ-01-2675’, ‘DZ-Cr-44’, ‘DZ-01-99’, ‘DZ-Cr-37’, ‘DZ-Cr-409’, ‘DZ-01-899’, ‘Ho-Cr-136’, ‘DZ-01-1681’, ‘DZ-Cr-358’, ‘DZ-Cr-82’, ‘DZ-01-1281’, ‘DZ-01-196’, ‘DZ-Cr-354’, ‘DZ-01-974’, ‘DZ-01-787’, ‘DZ-Cr-385’, ‘DZ-Cr-255’, ‘DZ-01-1285’	
		HARC	‘DZ-01-2053’, ‘DZ-01-1278’, ‘Acc.205953’
		MARC	‘DZ-Cr-387-RIL127’
		SARC	‘DZ-01-2054’, ‘DZ-01-146’, ‘DZ-01-1821’, ‘DZ-Cr-387RIL273’
		ADARC	‘DZ-01-1868’, ‘DZ-01-2423’, ‘DZ-01-3186’
		BARC	‘DZ-01-1880’, ‘23-Tafi-Adi-72’
		ARARC	‘PGRC/E 205396’

DZARC= Debre-Zeit Agricultural Research Centre, HARC = Holleta Agricultural Research Centre, MARC= Melkassa Agricultural Research Centre, SARC= Sirinka Agricultural Research Centre, ADARC = Adet Agricultural Research Centre, BARC = Bako Agricultural Research Centre, ARARC = Areka Agricultural Research Centre

### 5.2.2 DNA sampling, SSR markers and PCR amplification

The 60 tef genotypes were grown in the glasshouse at the University of KwaZulu-Natal. Leaf samples were taken from three week old seedlings. All samples were used in bulked amplification using DNA from 10 individual leaf samples. For each, 2 µL of each of bulked sample was used in the polymerase chain reaction (PCR).

The SSR analysis was done at the INCOTEC-PROTEIOS laboratory in South Africa (Incotec, SAPty. Ltd. South Africa). Ten polymorphic SSR markers were used in this study (Table 5.2). The primer sequences used for polymerase chain reaction (PCR) amplification were selected from published SSR-based map of *tef* (Zeid et al., 2011). PCR was done for all of the 10 primers. PCR products were fluorescently labelled and separated by capillary electrophoresis using an ABI 3130 automatic sequencer (Applied Bio systems, Johannesburg, South Africa).

Table 5.2 Ten selected microsatellites primers used to study 60 *tef* [*Eragrostis tef* (Zucc.) Trotter] genotypes

Primers	Forward primer	Reverse primer
CNLTS11	GTTTCATGTGCCTGCGCGTGT	TCCACGGGGAGAGCGACAGA
CNLTS33	TTTGACCTAGTCTCCCATG	ACGATCGGATGTTTTGCTTT
CNLTS60	AGGGTGATAGCTGCCAGAC	CCCGAGTAATTGGTCGCTAA
CNLTS133	GGGGAGACTGCATTGGACTA	CAAGAGGGACTGCACAGTGA
CNLTS136A/B	TGAGAAGGTAATAACTGGTGAAGC	CAAGGTTTACACACCGTGACTT
CNLTS455A/B	ACTCCGGAAGAACCACAACA	ACATGGAAAGAGGTGGCAAG
CNLTS42	ATGCATGGATGGATGGCTA	TTACCCAATTGCCCTAGCTG
CNLTS380	ACTGCAACGACAACGCTATG	GGGTACATTCGCGAAAAGAG
CNLTS538	CCATCTTAGCTTTGGCGAGA	ACAAGAGGCAACAAGCCAGA
CNLTS 255	TCTCAGCATCGTCTTTGTGTG	TTTTGTGCACGTATTTTTGGA

Source: Zeid et al., 2011

### 5.2.3 Data analysis

Genotypic data were subjected to various measures of the genetic relationships within and among the *tef* genotypes using GenAlex version 6.5 (Peakall and Smouse, 2007). Genetic parameters such as total number of alleles per locus ( $N_a$ ), number of effective alleles per locus ( $N_e$ ), Shannon's Information Index ( $I$ ), and gene diversity were determined according to the protocol described by Nei and Li (1979). The F-statistics such as genetic differentiation ( $F_{ST}$ ), fixation index or inbreeding coefficient ( $F_{IS}$ ) and overall fixation index ( $F_{IT}$ ) were calculated according to Wright's original derivation (Wright, 1951). Based on Euclidian distances, analysis of molecular variance (AMOVA) was conducted to partition the total genetic variation within and among genotypes. Among other genetic parameters gene flow ( $N_m$ ) was calculated using the following:  $N_m = \frac{1}{4} * [(1 - F_{ST}) / F_{ST}]$  where,  $F_{ST}$  is genetic differentiation (Slatkin and Barton, 1989). Polymorphic information content (PIC) was estimated using the formula:  $PIC = 1 - \sum P_i^2$  where  $P_i$  is the frequency of the  $i^{th}$  allele.

Cluster analysis was carried out by using neighbour-joining (NJ) algorithm using the un-weighted pair group method (UWPGM) in DARwin 5.0 software (Perrier and Jacquemoud-

Collet, 2006). A dendrogram was then generated using the dissimilarity matrix. Bootstrap analysis was performed for node construction using 1000 bootstrap value.

### 5.3 Results and discussion

#### 5.3.1 Genetic polymorphism of SSR markers

Table 5.3 Summarizes the genetic diversity parameters of the 10 SSR markers. The difference between the longest and shortest amplified fragment size ranged from 158 to 334 bp. The highest variation in fragment size was observed for primer CNLTS455A/B (220-299 bp) and the lowest was for primer CNLTS33 (235-257 bp). All the 10 SSR primer pairs were polymorphic and a total of 164 alleles were detected. The number of alleles per locus ranged from 10 for SSR markers CNLTS11 and CNLTS380 to 23 for markers CNLTS136A/B and CNLTS42, with a mean of 16.4 alleles per locus. This was in agreement with an average 8 to 23 alleles per locus reported by Zeid et al. (2012). The number of effective alleles ( $N_e$ ) ranged from 3.21 for marker CNLTS11 to 10.95 for marker CNLTS136A/B, with a mean of 7.14 (Table 5.3).

The observed heterozygosity ( $H_o$ ) varied from 0.38 for marker CNLTS538 to 1.00 for CNLTS136A/B, with a mean  $H_o$  of 0.67. Gene diversity ( $H_e$ ) of the markers ranged from 0.67 (CNLTS11) to 0.94 (CNLTS136A/B), with a mean of 0.86. Similarly Zeid et al. (2012) reported  $H_e$  ranged from 0.75-0.91 when using these markers. The high heterozygosity and low genetic fixation in the current study signals high genetic variability within the presently sampled tef genotypes. Some loci were more monomorphic (CNLTS255, CNLTS11 and CNLTS380) with less gene flow than other loci such as CNLTS455A/B and CNLTS133 (Table 5.3).

The polymorphic information content (PIC) of loci ranged from 0.64 (CNLTS11) to 0.94 (CNLTS136A/B), with a mean of 0.84 (Table 5.3). Four microsatellites (CNLTS133, CNLTS136A/B, CNLTS455A/B and CNLTS42), each with more than 20 alleles per loci had PIC values higher than 0.9. This indicates the high discrimination ability of the microsatellites for the studied tef genotypes. These are useful markers for further genetic analysis and marker assisted selection in tef. Likewise, the markers may be used to determine relationships and genetic diversity in tef germplasm for breeding and germplasm conservation. Likewise, PIC values ranges of 0.05 to 0.86 were reported by Zeid et al. (2012) using 39 microsatellites. The 10 microsatellites used in this study were selected from these 39 markers for their high PIC values that ranged from 0.68 to 0.91, which is similar to the current findings.

Table 5.3 Genetic parameters estimated using 10 selected SSR markers and 60 tef [*Eragrostis tef* (Zucc.) Trotter] genotypes

Locus	Fragment size range	Genetic parameter							
		Na	Ne	I	Ho	He	F	Nm	PIC
CNLTS11	205-242	10	3.21	1.42	0.60	0.67	0.07	3.71	0.64
CNLTS33	235-257	12	5.76	1.86	0.76	0.85	0.07	4.15	0.83
CNLTS60	303-334	13	7.46	2.12	0.61	0.91	0.29	5.56	0.90
CNLTS133	260-302	21	9.52	2.35	0.93	0.92	0.00	7.53	0.92
CNLTS136A/B	243-282	23	10.95	2.52	1.00	0.93	0.00	6.22	0.94
CNLTS455A/B	220-299	20	10.3	2.45	1.00	0.94	0.00	8.63	0.93
CNLTS42	176-220	23	9.65	2.40	0.65	0.93	0.28	7.15	0.92
CNLTS380	220-290	10	4.14	1.60	0.41	0.78	0.44	3.99	0.79
CNLTS538	192-250	12	4.26	1.54	0.38	0.78	0.52	4.87	0.69
CNLTS255	158-199	20	6.13	1.94	0.62	0.85	0.24	2.02	0.86
Mean		16.4	7.14	2.02	0.70	0.86	0.16	5.38	0.84
SE		5.44	0.60	0.09	0.05	0.02	0.05	0.64	0.10

Na = Number of alleles; Ne = Number of effective alleles; I = Shannon's information index; Ho = Observed heterozygosity; He = Unbiased expected heterozygosity (gene diversity); F = Fixation index; Nm = Gene flow; PIC = Polymorphic information content; SE = standard error.

### 5.3.2 Patterns of genetic relationship within and among tef genotypes

Table 5.4 presents the genetic diversity parameters among the studied tef genotypes. Knowledge of frequency and distribution of alleles is important to identify genetically distinct parents. The numbers of observed and effective alleles were higher for the landraces and improved varieties than for the new accessions collected from the Tigray region. Similarly, the highest Shannon diversity index of 2.16 was recorded for the landraces and the lowest (1.90) was for the new accessions from Tigray. The mean value was 2.02 and this was higher than the mean Shannon diversity index of 0.73 reported by Assefa et al. (2003). Gene diversity was 0.67, 0.65 and 0.77 for the improved varieties, landraces and new accessions, respectively (Table 5.4). The high genetic variation present among the landraces will assist in future tef breeding programs. Furthermore, the high variability noted within the tested improved varieties indicated that the tef breeding program in Ethiopia has been utilizing diverse sources of genes for developing new varieties. The present results are in agreement with the high genetic variability reported by Assefa et al. (2003a) and Zeid et al. (2012). Contrary to these findings, Ayele et al. (1999) and Bai et al. (2000) reported low genetic variability in selected tef genotypes when using AFLP markers and RAPD markers, respectively. This suggests that the SSR markers are more valuable and informative for genetic diversity analysis than AFLP and RAPD markers. The mean observed heterozygosity ( $H_o$ ) of 0.67 was less than the average expected heterozygosity ( $H_e$ ) of 0.87 (Table 5.4), suggesting the existence of high level of inbreeding in the population

owing the autogamous nature of tef. The fixation index was lowest for the tef new accessions (0.05) collected from Tigray region, indicating higher genetic variability in the accessions than in the improved varieties and landraces. Overall, high PIC values (>0.7) were observed for all the populations but the landraces displayed the highest mean PIC value of 0.85.

Table 5.4 Mean of genetic parameters for three groups of the 60 tef [*Eragrostis tef* (Zucc.) Trotter] genotypes studied using 10 selected SSR markers

Populations	Genetic parameters							
	N	Na	Ne	I	Ho	He	F	PIC
Landraces	18	11.20	7.79	2.16	0.67	0.88	0.22	0.85
Improved varieties	33	12.20	7.06	2.00	0.65	0.81	0.21	0.76
New accessions	9	8.10	6.56	1.90	0.77	0.87	0.05	0.78
Mean	20	10.47	7.14	2.02	0.70	0.86	0.16	0.80
SE	-	0.81	0.61	0.09	0.05	0.02	0.05	0.05

N = Number of genotypes tested; Na = Number of alleles; Ne = Number of effective allele; I = Shannon's information index; Ho = Observed heterozygosity; He = Unbiased expected heterozygosity (gene diversity); F = Fixation index; Nm = Gene flow; PIC = Polymorphic information content; SE = standard error.

### 5.3.3 Analysis of molecular variance (AMOVA)

Analysis of molecular variance (AMOVA) showed significant differences among the populations. A low level of variations was observed among populations (2%), while high level of variation was revealed among and within genotypes at 35% and 63%, respectively (Table 5.5). The magnitudes between and within population differentiation were quantified using the F-statistics of Wright (1951). Genetic differentiation among the populations was low ( $F_{ST} = 0.02$ ), with a moderate effect of non-random mating within the populations ( $F_{IS} = 0.36$ ). The low genetic differentiation among populations was due to the high variability within the populations, which could be due to gene flow (Gepts and Papa, 2003). The exchange of genes through hybridization of the landraces in developing improved tef varieties could be one of the causes of the current low level of genetic differentiation among populations. The wide agro-ecological adaptation of tef (Ketema, 1997), farmer's diverse selection practices such as selecting for diverse market segments, grain and straw yields, seed colors, flat bread (*injera*) quality and maturity all of which contribute to the considerable genetic diversity of tef genotypes (Tefera et al., 2005; Belay et al., 2008; Assefa et al., 2015).

Table 5.5 Sum of squares, mean squares, estimated variance, proportion of explained variance and F-statistics from the analysis of molecular variance (AMOVA) of 60 tef [*Eragrostis tef* (Zucc.) Trotter] genotypes based on populations using 10 selected SSR markers

Sources of variation	df	SS	MS	Estimated Variance	Proportion of variance (%)	F-Statistics
Among populations	2	17.63	8.81	0.09	2%	$F_{ST} = 0.02$
Among genotypes	57	332.25	5.83	1.54	35%	$F_{IS} = 0.36$
Within genotypes	60	165.00	2.75	2.75	63%	$F_{IT} = 0.37$
Total	119	514.88		4.37	100%	

df = degree of freedom; SS = sum of square; MS = mean square

#### 5.3.4 Genetic interrelationships among tef genotypes

The dendrogram of cluster analysis based on neighbor-joining algorithm using the un-weighted pair group method (UWPGM) categorized the 60 tef genotypes into three major clusters consisting of 28, 57 and 15% of the genotypes in cluster I, II and III, respectively (Figure 5.2). 83% of the 18 landraces grouped in Cluster II. This indicated that majority of the landraces were genetically similar showing low differentiation. The landraces sub-clustered into three sub-groups, which indicated their distinct morphological performance. This was also reported by Plaza-Wuthrich et al. (2013) that 18 landraces could be group into six clusters based on their morphological performance. In the present study the landraces '*Alba*' and '*Gea-Lamie*' were grouped in the same cluster but in different sub-groups. Similarly, Ayele et al. (1999) grouped these landraces, in different clusters based on their distinct morphological traits. In addition, two landraces, '*Fesho*' and '*Kaye-Murri*', because of their contrasting morphological characteristics were formerly used to develop mapping populations (Bai et al., 1999). These grouped in same cluster but in different sub-groups in this study. However, Assefa et al., (2003a) based on ISSR markers put these two landraces into separate genetic groups.

Out of the nine new tef accessions collected from Tigray region, 1, 5 and 3 grouped into clusters I, II and III, respectively (Figure 5.2), indicating their distinctness. The present grouping pattern of the new accessions does not correlate with their site of collection, showing the same trend as reported by Assefa et al. (2003a). Similarly, out of the 33 improved varieties, 45%, 43% and 12% were grouped into Cluster I, II and III, respectively (Figure 5.2). Assefa et al. (2003a) grouped 10 improved varieties released until 1995 in one cluster indicating their genetic similarity, whereas the present study distributes these varieties into three genetic clusters. The two varieties, '*DZ-Cr-37*' recently released for its high grain yield and late maturity and variety,

'*DZ-Cr-387-RIL127*', known for its early maturity, were all derived from crosses of '*DZ-01-974*'x '*DZ-01-196*'. These varieties were grouped in the same cluster in the present study indicating their genetic relatedness, in agreement with the report of Zeid et al. (2012).

The genotypes, '*DZ-Cr-385*' and '*213237*' were grouped into Cluster I and '*222076*' in cluster III are early maturing types and display good grain yield performances in moisture stressed environments in Ethiopia. However, the genotypes, '*DZ-Cr-387*', '*205896*', and '*Dschanger*' grouped into Cluster II and '*205917*' in Cluster III, are late maturing entries with high grain yields, tall plant height and long panicles in optimum rainfall environments. The unique agronomic attributes of the genotypes were consistent with the present genetic analysis using SSR markers, and these genotypes would be useful for breeding for drought tolerance and better yield performance.

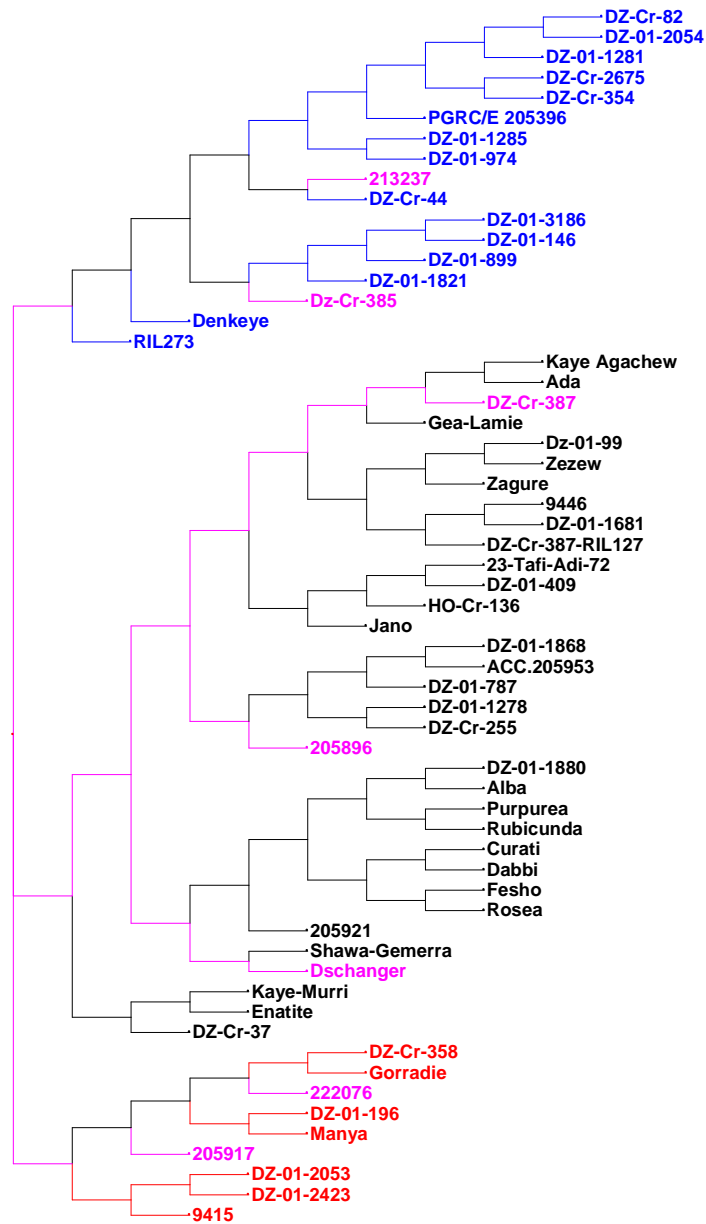


Figure 5.2 Un-weighted pair group method (UWPGM) dendrogram showing genetic relationship of the 60 tef genotypes determined using 10 selected SSR markers. The genotypes marked with purple are selected for future tef breeding program.

## 5.4 Conclusions

In the current study the SSR markers were used to determine the genetic relationships within 60 tef genotypes thereby, indicating their usefulness in genetic studies and tef breeding programs. The tested SSR markers could be helpful in conserving and managing tef germplasm. Marker based identification and selection of the distinct genotypes could be helpful for the development of improved tef varieties. The improved tef varieties ('DZ-Cr-385' and 'DZ-Cr-387'), new accessions that were collected from the Tigray region ('222076', '213237', '205896' and '205917') and landraces originating from Kaffa ('Dschanger') were identified with distinct genomic attributes useful in future tef breeding programs.

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## Chapter 6: **Gene Action and Early Generation Selection of Yield and Related Traits among Tef Populations under Moisture-Stressed and Non-Stressed Environments**

### **Abstract**

The nature of combining ability of chosen parents and types of gene action involved in their families are fundamental for breeding. The aim of this study was to determine gene action, genetic variation and inheritance of grain yield, and component traits of newly developed tef populations under moisture-stressed and non-stressed conditions for drought tolerance breeding. Eight selected parents were crossed using a half-diallel mating design. Seventeen successful crosses along with the parents were evaluated in the F<sub>2</sub> generation under moisture-stressed and non-stressed conditions in northern Ethiopia during 2015 and 2016. Additive gene action predominantly controlled the inheritance of grain yield, biomass yield, harvest index, days-to-panicle emergence, days-to-maturity, panicle length, plant height, main shoot panicle seed weight, number of productive tillers per plant and relative water content under both the moisture-stressed and non-stressed conditions. Non-significant general combining ability (GCA) and specific combining ability (SCA) effects were detected for biomass yield, relative water content and peduncle length under the non-stressed condition. Under both test conditions, genotypes, DZ-Cr-387 and 9415, were the best general combiners for increased grain yield and yield components except for days-to-maturity. Conversely, Genotype, 222076 was the best general combiner for reduced maturity period only. The selected parents are novel genetic materials for tef breeding programs to improve grain yield and yield components with reduced days-to-maturity for drought tolerance breeding. The families of DZ-Cr-387 x 207832 and DZ-Cr-387 x 222076 were high grain yielders with early maturity under both test conditions. There were marked genotypic and phenotypic variation among the crosses in the F<sub>2</sub> generation for plant height, panicle length, peduncle length, number of productive tillers per plant, main shoot panicle seed weight, biomass yield and grain yield under both test conditions, important for successful selection and genetic advancement.

**Keywords:** combining ability, drought tolerance, genetic variation, half-diallel, tef

## 6.1 Introduction

Tef is the leading cereal crop in Ethiopia and preferred for food, feed and as cash crop. Tef products are gluten-free and hence they are used by people with gluten intolerance worldwide (Gebremariam et al., 2012; Bultes and Umeta, 2013). The crop has wide adaptability and is relatively tolerant to biotic and abiotic stresses (Assefa et al., 2011). The annual rainfall requirement of the crop for optimum yield varies from 950 to 1500 mm (FAO, 2011), while a reasonable yield can be harvested under low rainfall conditions of 450 to 550 mm (Ketema, 1997).

In Ethiopia, tef yield is generally low with the national average being 1.6 t ha<sup>-1</sup> (CSA, 2015). Moisture-stress remains the main constraint affecting tef production and productivity (Assefa et al., 2011). About 25 to 51% grain yield reduction in tef has been reported due to moisture-stress (Admas and Belay, 2011; Shiferaw et al., 2012; Abraha et al., 2016). Knowledge of the genetic variability and gene action present in a genetic pool is a pre-requisite for identify the best parents and families for drought tolerance breeding programmes (Mohammadi et al., 2010; Nouri et al., 2011).

Combining ability analysis has been widely used to derive information on the breeding potential of parents via progeny tests and to compare the performance of genotypes in hybrid combinations (Griffing, 1956). It also helps to identify the best hybrid combinations and supplies data on the type of gene action, which controls different agronomic traits. Furthermore, knowledge on gene action within the breeding population is a key to selecting an appropriate breeding procedure in any crop breeding program. The general combining ability (GCA) and specific combining ability (SCA) effects are effective genetic parameters in choosing the best parents and crosses. General combining ability is the average performance of a parent genotype across several cross combinations and is associated with additive gene effect, while SCA reveals the relative performance of a cross, and is typically associated with non-additive gene effects (Chahal and Gosal, 2002; Sleper and Poehlman, 2006). The diallel mating design is the most commonly used method to estimate GCA and SCA effects (Falconer and Mackay, 1996) in several crops such as wheat (Rad et al., 2013; Golparvar, 2014; Zare-Kohan and Heidari, 2014), rice (Adilakshmi and Upendra, 2014; Mandal et al., 2015), sorghum (Mengistu et al., 2010) and maize (Zare et al., 2011; Moradi et al., 2014). However, its application in tef breeding is scarce due to the difficulty in making successful crosses because of its extremely small flower size, its flowering time, the delicate nature of the flower and its mating system.

Early generation selections among cross combinations reduce the number of poorly performing families forwarded for future selection. This helps in efficient utilization of land, time and human resources and allows handling of sufficient segregating populations derived from a few promising crosses. This is carried out through careful examination of the extent of genetic variation present among the segregating populations at early generations. Heritability is an important genetic parameter that measures the phenotypic variance attributable to genetic causes. Heritability estimates have predicative value of the potential genetic advances of a selection effort. High genetic gains coupled with high heritability estimates, offer the most effective situation for selection (Falconer and Mackay, 1996; Sleper and Poehlman, 2006). The extent of genetic variation for yield and yield components and heritability of traits in tef genotypes have been reported in previous studies (Assefa et al., 1999, 2000, 2001; Chanyalew, 2010; Admas and Belay, 2011; Ayalew et al., 2011; Shiferaw et al., 2012; Debebe et al., 2013). Heritability and genetic parameter estimates are specific to the test populations and environments (Falconer and Mackay, 1996).

Over the last five decades the Ethiopian tef breeding program has released some 35 improved varieties (MoA, 2014). Of these, varieties, Quncho (DZ-Cr-387) and Tsedey (Dz-Cr-37) are the most popular and widely grown under optimum and moisture-stressed areas, respectively. Wide adoption of improved tef varieties and production inputs such as fertilizers and pesticides have contributed to increased tef productivity in the country (Minten et al., 2013). Recently, a tef variety referred to as 'Kora', has been released for its better yield and lodging tolerance under optimum rainfall areas. This variety has shown an 18% yield advantage over DZ-Cr-387 under high rainfall conditions (Hailekiros, 2015; AxARC, 2016) in central Tigray. However, it has delayed maturity, leading to low yield levels under low rainfall seasons. Therefore, this variety is not suitable for moisture-stressed areas such as the northern parts of Ethiopia and similar agro-ecologies. In these agro-ecologies rainfall is generally low and erratic; especially during the grain filling stage (September) (Tsegay et al., 2012). Therefore, there is a need to develop improved tef varieties that combine early maturity as a moisture-stress escape mechanism, and a better yield performance, through designed crosses and selection. Therefore, the aim of this study was to determine gene action, genetic variation and inheritance of grain yield and its components among newly developed tef populations under moisture-stressed and non-stressed conditions for drought tolerance breeding.

## 6.2 Materials and methods

### 6.2.1 Description of the study environments

The study was conducted at three sites in the Tigray region of northern Ethiopia. These were Dura (14°06'76.2"N, 038°39'14.5"E, 2073 meter above sea level [m.a.s.l.]) and Hastebo (14°06'40.2"N, 038°45'45.8"E, 2118 m.a.s.l.) which are on-farm research sites of Axum Agricultural Research Center (AxARC); and Adi-gidad (14°04'88"N, 38°21'71"E, 1916 m.a.s.l.) of the May-stebri Agricultural Research Centre (MARC). Dura has an established irrigation system, which makes it suitable to conduct offseason experiments. At Hastebo and Adigdad tef is grown as a rain-fed crop. Eleven years of rainfall data of the testing sites showed the variable and erratic trend of rainfall across the years (Ethiopian Meteorology Agency, 2016). The annual rainfall figure during 2015 was 615.8 mm at Hastebo and Dura, and 988.5 mm at Adi-gidad (Appendix 1 and 2). The test populations were grown in the middle of the main growing season, i.e., August to December. Hastebo and Adi-gidad sites received 318.5 and 524.5 mm rainfall respectively, during the experimental period (Table 6.3), which were low compared to the ideal rainfall requirement of 950 to 1500 for optimum yield of tef (FAO, 2011). The mean minimum and maximum temperatures at Dura and Hastebo sites varied from 9.9 to 28.5°C, while that of Adi-gidad ranged from 13.86 to 29.07 °C (Appendix 6.1 and 6.2). The soil types were predominantly clay at all the three testing sites.

### 6.2.2 Plant materials

Eight parents were selected for crossing, consisting of five accessions (9415, 205896, 222076, acc-15-ck and 207832), a landrace (Dschanger) and two released varieties (DZ-Cr-387 and DZ-Cr-385). Parents, 9415, 205896, DZ-Cr-387 and Dschanger were selected for their high grain yield, tall plants and long panicles, and reduced lodging susceptibility under optimum-moisture conditions. These varieties produced reduced grain yield under moisture-stressed conditions (Abraha et al., 2016). Descriptions of the parents are summarised in Table 6.1 and Figure 6.1. The remaining four parents namely, 222076, Acc-ck-15, 207832 and DZ-Cr-385, were selected for their early maturity and better yield potential under moisture-stressed conditions. The selected parents show varied lemma and seed color, serving as useful markers in identifying true crosses.

Table 6.1 Name and qualitative trait description of tef parents used in the study.

Genotype name	Lemma colour	Panicle form	Seed colour
DZ-Cr-387	Yellow green with red tips	Semi-loose	White
9415	Yellow green with red tips	Semi-loose	White
205896	Yellow green with purple tips	Semi-loose	Pale white
Dschanger	Gray	Semi-loose	Dark brown
acc-ck-15	Greyish purple	semi-loose	Dark brown
DZ-Cr-385	Yellow green	Semi-loose	White
207832	Variegate	Semi-loose	Light brown
222076	Yellow green	Very-loose	Pale white



Figure 6.1 Panicle forms and lemma colours of selected tef parents used in the study: A=DZ-Cr-387, B=9415, C=205896, D=Dschanger, E=acc-ck-15, F=222076, G=DZ-Cr-385 and H=207832

### 6.2.3 Crosses and mating design

Tef is a highly self-pollinated crop with bisexual flowers (florets) that are very small in size, making artificial hybridization a difficult task. Hand emasculatation and pollination were done under a stereo-microscope (x15 magnification) according to the technique developed by Berhe (1975). The crossing procedures are pictorially summarised in Figures 6.2A and B. Briefly, parents were grown in pots in a glasshouse. At flowering, pots were individually transported to a microscopy laboratory and one to three flowers of the female parents were carefully emasculated at 14h00hr to 16h00hr. Emasculated flowers were bagged for controlled pollination on the following day. The selected male parents as pollen donors were kept in a dark room overnight. In the following morning (7h00hr) plants were exposed to full sunlight to open their flowers. Pollen grains were collected from the male parent and pollinated onto the emasculated female flowers under a stereo-microscope. Only two to three crosses were possible per day because of the tedious process of flower preparation and making crosses. Crosses involving the eight parents were performed using a half-diallel mating design (Dabholkar, 1992), according to the scheme shown in Table 6.2. This provided 28 cross combinations of which 17 crosses were successful and available for individual selection



Figure 6.2 Photos showing crossing of tef under a stereomicroscope

Table 6.2 An 8 x 8 half-diallel mating scheme in tef showing overall crosses

Parents	205896	9415	Dschanger	DZ-Cr-385	acc-ck-15	207832	222076
DZ-Cr-387	(1) DZ-Cr-387 X 205896 <b>X</b>	(2) DZ-Cr-387 X 9415 <b>X</b>	(3) DZ-Cr-387 X Dschanger <b>X</b>	DZ-Cr-387 X DZ-Cr-385 205896 X DZ-Cr-385 9415 X DZ-Cr-385 Dschanger X DZ-Cr-385 <b>X</b>	(4) DZ-Cr-387 acc-ck-15 205896 X acc-ck-15 9415 X Acc-ck-15 Dschanger X acc-ck-15 (14) DZ-Cr-385 X acc-ck-15 <b>X</b>	(5) DZ-Cr-387 X 207832 205896 X 207832 9415 X 207832 Dschanger X 207832 DZ-Cr-385 X 207832 acc-ck-15X 207832 <b>X</b>	(6) DZ-Cr-387 X 222076 205896 X 222076 9415 X 222076 Dschanger X 222076 (15) DZ-Cr-385 X 222076 acc-ck-15X 22076 X 222076 <b>X</b>
205896		(7) 205896 X 9415 <b>X</b>	(8) 205896 Dschanger (11) 9415 X Dschanger <b>X</b>	X DZ-Cr-385 9415 X DZ-Cr-385 Dschanger X DZ-Cr-385 <b>X</b>	X acc-ck-15 9415 X Acc-ck-15 Dschanger X acc-ck-15 (14) DZ-Cr-385 X acc-ck-15 <b>X</b>	X 207832 9415 X 207832 Dschanger X 207832 DZ-Cr-385 X 207832 acc-ck-15X 207832 <b>X</b>	X 222076 X 222076 (12) 9415 X 222076 (13) Dschanger X 222076 (15) DZ-Cr-385 X 222076 acc-ck-15X 22076 X 222076 <b>X</b>
9415		<b>X</b>	(11) 9415 X Dschanger <b>X</b>	X DZ-Cr-385 9415 X DZ-Cr-385 Dschanger X DZ-Cr-385 <b>X</b>	9415 X Acc-ck-15 Dschanger X acc-ck-15 (14) DZ-Cr-385 X acc-ck-15 <b>X</b>	X 207832 9415 X 207832 Dschanger X 207832 DZ-Cr-385 X 207832 acc-ck-15X 207832 <b>X</b>	X 222076 X 222076 (12) 9415 X 222076 (13) Dschanger X 222076 (15) DZ-Cr-385 X 222076 acc-ck-15X 22076 X 222076 <b>X</b>
Dschanger			<b>X</b>	X DZ-Cr-385 9415 X DZ-Cr-385 Dschanger X DZ-Cr-385 <b>X</b>	X acc-ck-15 9415 X Acc-ck-15 Dschanger X acc-ck-15 (14) DZ-Cr-385 X acc-ck-15 <b>X</b>	X 207832 9415 X 207832 Dschanger X 207832 DZ-Cr-385 X 207832 acc-ck-15X 207832 <b>X</b>	X 222076 X 222076 (12) 9415 X 222076 (13) Dschanger X 222076 (15) DZ-Cr-385 X 222076 acc-ck-15X 22076 X 222076 <b>X</b>
DZ-Cr-385				X DZ-Cr-385 9415 X DZ-Cr-385 Dschanger X DZ-Cr-385 <b>X</b>	X acc-ck-15 9415 X Acc-ck-15 Dschanger X acc-ck-15 (14) DZ-Cr-385 X acc-ck-15 <b>X</b>	X 207832 9415 X 207832 Dschanger X 207832 DZ-Cr-385 X 207832 acc-ck-15X 207832 <b>X</b>	X 222076 X 222076 (12) 9415 X 222076 (13) Dschanger X 222076 (15) DZ-Cr-385 X 222076 acc-ck-15X 22076 X 222076 <b>X</b>
acc-ck-15				X DZ-Cr-385 9415 X DZ-Cr-385 Dschanger X DZ-Cr-385 <b>X</b>	X acc-ck-15 9415 X Acc-ck-15 Dschanger X acc-ck-15 (14) DZ-Cr-385 X acc-ck-15 <b>X</b>	X 207832 9415 X 207832 Dschanger X 207832 DZ-Cr-385 X 207832 acc-ck-15X 207832 <b>X</b>	X 222076 X 222076 (12) 9415 X 222076 (13) Dschanger X 222076 (15) DZ-Cr-385 X 222076 acc-ck-15X 22076 X 222076 <b>X</b>
207832				X DZ-Cr-385 9415 X DZ-Cr-385 Dschanger X DZ-Cr-385 <b>X</b>	X acc-ck-15 9415 X Acc-ck-15 Dschanger X acc-ck-15 (14) DZ-Cr-385 X acc-ck-15 <b>X</b>	X 207832 9415 X 207832 Dschanger X 207832 DZ-Cr-385 X 207832 acc-ck-15X 207832 <b>X</b>	X 222076 X 222076 (12) 9415 X 222076 (13) Dschanger X 222076 (15) DZ-Cr-385 X 222076 acc-ck-15X 22076 X 222076 <b>X</b>
222076				X DZ-Cr-385 9415 X DZ-Cr-385 Dschanger X DZ-Cr-385 <b>X</b>	X acc-ck-15 9415 X Acc-ck-15 Dschanger X acc-ck-15 (14) DZ-Cr-385 X acc-ck-15 <b>X</b>	X 207832 9415 X 207832 Dschanger X 207832 DZ-Cr-385 X 207832 acc-ck-15X 207832 <b>X</b>	X 222076 X 222076 (12) 9415 X 222076 (13) Dschanger X 222076 (15) DZ-Cr-385 X 222076 acc-ck-15X 22076 X 222076 <b>X</b>

Note:

- Numbers (1) to (17) denote successful crosses for individual plant selection at the F<sub>2</sub>.
- Sufficient seeds of the following 10 crosses were available for genetic studies at the F<sub>2</sub>: 1, 2, 3, 6, 7, 8, 10, 11, 12 and 13
- The following parents were included for genetic analysis: DZ-Cr-387, 9415, 205896, Dschanger and 222076

Crosses were performed during February - March 2015. The F<sub>1</sub> generations were developed immediately after harvesting the F<sub>1</sub> seeds. The F<sub>1</sub> seeds were first germinated in a Petri-dish and later transferred into a plastic pot for successful progeny development. Vigorous and health F<sub>1</sub> plants were established to harvest enough seeds for field evaluation and selection at the F<sub>2</sub>. The F<sub>1</sub> plants were checked for true hybridity using lemma and seed colors. In tef, purple, red and gray lemma colors are dominant over yellowish green lemma color; purple is dominant over red and gray; and red is dominant over gray (Berhe, 1981).

#### 6.2.4 Experimental design and trial management

The 17 crosses and eight parents were laid out in a 5 x 5 simple lattice design with two replications. The F<sub>2</sub> seeds were sown in plastic pots and field transplanted after 30 days. Field transplanting was done to reduce seed loss and to facilitate space planting. Typically tef is direct planted through hand broadcasting or seed drilling and it is usually not possible to control spacing between plants due to tef's tiny seeds. Segregating plants were planted in two 1.5 m long rows with an inter-row spacing of 0.25 m, and a row of a released tef variety Kora (DZ-Cr-438, RIL No. 133B), was planted as a border on both sides of each plot. Each plot measured 1.5 m<sup>2</sup>. During the main cropping season (July to December), planting was done in mid-August 2015, i.e., that was about three weeks after the regular planting timeline. This was done to expose the segregating populations to terminal moisture-stress. During the offseason

(December to April, 2016) planting was done in mid-December, 2015. Fertilizers, diammonium phosphate (DAP) and urea were each applied at  $100 \text{ kg ha}^{-1}$  with the DAP applied entirely at planting time, while urea was split applied (half during emergence and the remaining half, four weeks after planting (WAP) according to the recommendation made by AxARC (2011). Weeding was manually done twice, at three weeks and five to six WAP. Harvesting for data collection was done from the two central rows of each plot.

Across the three testing sites four experiments were conducted (Table 6.3). These were two water regimes: Regime I (moisture-stressed) under rain-fed conditions during the main season (August to December, 2015) at Hastebo (Environment 1) and Adi-gidad (Environment 2) and at the Dura site (Environment 3) with irrigation withheld after days to 50% panicle emergence till maturity during the off-season (December 2015 - April 2016). Regime II (optimal-moisture), irrigated plots from planting till physiological maturity (off-season, December to April 2016) at Dura site (Environment 4) (Table 6.3).

Crop water requirement was calculated using the following formula:  $CWR = KC \times ETo$ , where CWR is crop water requirement, KC is the crop coefficient determined at different growth and developmental stages and ETo is the reference crop evapotranspiration. ETo was calculated using CROPWAT 8.0 software developed by the Land and Water Division of FAO (FAO, 1998). Climate data including daily rainfall, maximum and minimum temperature, relative humidity, sunshine hours and wind speed of the Dura site were obtained from the Ethiopian Meteorological Agency of Mekelle Branch to calculate ETo. A total of 714.4 mm and 514.4 mm irrigated water were applied for the non-stressed and moisture-stressed experiments, respectively (Table 6.3). The required amount of water needed to be applied for a plot was measured using partial flow.

Table 6.3 Testing sites and the four environments involving two water regimes used for evaluation of 17 F<sub>2</sub> tef population and 8 parents during the main growing season of 2015 and off season of 2016

Site	Environment	Treatment/water regime	Growing season	Total rainfall/irrigated water per cropping (mm)
Hastebo Adi-gidad	E1	Moisture-stressed (rain fed)	Main-season (August-December)	318.5
	E2	Moisture-stressed (rain fed)	Main season (August-December)	524.5
Dura	E3	Irrigation withheld after 50% days to panicle emergence till maturity	Off-season (December-April)	514.4
	E4	Irrigated from planting till physiological maturity	Off-season (December-April)	714.4

### 6.2.5 Data collection

Days to 50% panicle emergence, days to 75% maturity, plant height (cm), panicle length (cm), peduncle length (cm), number of effective tillers per plant, biomass yield (g plant<sup>-1</sup>), grain yield (g plant<sup>-1</sup>) and main shoot panicle seed weight (g panicle<sup>-1</sup>) were recorded based on all individual plants in the two rows. Harvest index was estimated as a ratio of mean grain yield (g plant<sup>-1</sup>) to mean biomass yield (g plant<sup>-1</sup>). Lodging was scored according to the procedure of Caldicott and Nuttall (1979) for each plot using a 0-5 scale, where 0 indicates, plants in an upright position and 5 for plants lying flat on the ground. The lodging index was then calculated as the mean of the product sum of the angle of lodging and the corresponding percentage. To measure relative water content, one top fully expanded leaf per plant was taken from all the plants in a plot. Leaves were immediately weighed to obtain the fresh weight. The leaves were kept in small sealed plastic bags and kept in an ice box. The leaves were then soaked in water for 4 hr and weighed to obtain the turgid weight. The turgid leaves were oven-dried at 80°C for 24 hr and weighed to get their dry leaf weight. The relative water content was calculated according to the following formula proposed by Slavick (1979):

$$\text{RWC (\%)} = [(\text{FW}-\text{DW})/(\text{TW}-\text{DW})] \times 100$$

Where, FW is fresh weight, TW is turgid weight and DW is dry weight.

### 6.2.6 Data analysis

It was only one to three F<sub>1</sub> seeds harvested, because of the very difficult crossing of tef. Due to this limitation it was not possible to collect sufficient data from F<sub>1</sub> generation and hence to undertake genetic analysis. Therefore, genetic analysis was conducted in the F<sub>2</sub> involving sufficient number of individuals. Hallauer et al. (2010) indicated that in situations difficult to get

enough F<sub>1</sub> seed, it is possible to conduct diallel analysis using F<sub>2</sub> generation. Data collected from five parents and ten successful crosses with a sufficient number of F<sub>2</sub> plants (see footnote in Table 6.2) were used for genetic analyses under moisture-stressed and non-stressed environments. Data were used to compute the GCA and SCA effects using the SAS diallel procedure of Griffings Model I Method II (parents, F<sub>1</sub>'s) according to Griffings (1956) as follows:

$$Y_{ij} = \mu + g_i + g_j + s_{ij} + \epsilon_{ij}$$

Where, Y<sub>ij</sub> = Mean value of the progeny derived from the cross of i<sup>th</sup> female parent with j<sup>th</sup> male parent,  $\mu$  = Overall mean, g<sub>i</sub> = the GCA effects of the i<sup>th</sup> female parent, g<sub>j</sub> = the GCA effects of the j<sup>th</sup> male parent, s<sub>ij</sub> = the SCA effects for the cross between the i<sup>th</sup> female parent and the j<sup>th</sup> male parent and  $\epsilon_{ij}$  = experimental error with ij<sup>th</sup> genotype in the k<sup>th</sup> environment.

The relative importance of GCA and SCA effects were estimated using the general predicted ratio (GPR) for the traits observed (Baker, 1978). Ratios close to one indicate that additive effects in the inheritance of the trait are important, while ratios close to zero indicate the dominance effects are important in the inheritance. The GCA/SCA ratio formula is as:

$$\text{GCA/SCA} = (2\text{MS}_{\text{GCA}})/(\text{MS}_{\text{GCA}} + \text{MS}_{\text{SCA}}) \text{ (Baker, 1978).}$$

Data of the 17 crosses and 8 parents grown in the non-stressed and moisture-stressed environments were subjected to statistical analysis using a simple lattice procedure of SAS 9.3 (SAS Institute, 2011). Variances for each cross were analyzed using the Statistical Package for Social Sciences (SPSS Inc., 2009) using the data collected from individual plants of the parents and F<sub>2</sub> populations. Genotypic coefficient variance (GCV) and phenotypic coefficient variances (PCV), heritability (H) and genetic advance as percent of the mean (GAM) were computed according to the following formulas below:

$$\text{PCV} = \frac{\sigma^2_p}{\bar{X}} \times 100$$

Where  $\sigma^2_p$  is variance of F<sub>2</sub> population for a specific trait and  $\bar{X}$  is mean of the F<sub>2</sub> population.

$$\sigma^2_p = \frac{\sum x_i - \bar{X}}{n-1}, \text{ where } x_i \text{ is the individual value, } \bar{X} \text{ is mean and } n \text{ is the number of } F_2 \text{ progenies}$$

$$\text{GCV} = \frac{\sigma^2_g}{\bar{X}} \times 100$$

Where  $\sigma^2_g$  is genotypic variance calculated as  $\sigma^2_g = \sigma^2_P - \sigma^2_e$ ; where  $\sigma^2_e$  is the environmental variance calculated as  $\sigma^2_e = \sqrt{\sigma^2_{P1} * \sigma^2_{P2}}$  (Tefera et al., 2008)

Where  $\sigma^2_{P1}$  and  $\sigma^2_{P2}$  are variance of parent 1 and parent 2 of a specific  $F_2$  population

$$\text{Heritability (\%)} = \frac{\sigma^2_g}{\sigma^2_p} * 100$$

$$\text{GAM} = ih^2\sigma_p$$

Where  $i$  is coefficient of selection at 10% selection intensity,  $h^2$  is heritability, and  $\sigma_p$  is phenotypic standard deviation.

### 6.3 Results and discussion

#### 6.3.1 Analysis of variance and significant tests

To investigate the effect of different moisture condition, variance analyses for the moisture-stressed and non-stressed conditions were conducted. Under non-stressed condition entries showed significant differences ( $p \leq 0.01$ ) for harvest index, days-to-panicle emergence, days-to-maturity, panicle length, plant height, main shoot panicle seed weight and lodging index (Tables 6.4 and 6.5). The moisture-stressed environments were significantly ( $p \leq 0.01$ ) different for all the traits except peduncle length, lodging index and relative water content. Entries were significantly different ( $p \leq 0.01$ ) for all the traits except harvest index and lodging index, while entries by environment interaction was significant ( $p \leq 0.01$ ) for biomass yield, days-to-maturity, panicle length and plant height only (Tables 6.4 and 6.5).

The mean squares of GCA and SCA under moisture-stressed and non-stressed conditions are presented in Tables 6.4 and 6.5, respectively. Under the moisture-stressed conditions, GCA and SCA effects were significant for days-to-panicle emergence, days-to-maturity, panicle length, plant height, main shoot panicle seed weight, number of productive tillers per plant and relative water content (Table 6.4). This suggests the importance of both additive and non-additive gene actions in determining the inheritance of these characters. In the non-stressed environment, the GCA and SCA effects were significant for days-to-panicle emergence, days-to-maturity, panicle length, plant height and main shoot panicle seed weight. Determining the relative importance of additive and non-additive gene action is essential for development of an efficient hybridization program, and for selection (Falconer and Mackay, 1996; Chahal and Gosal, 2002; Sleper and Poehlman, 2006; Acquah, 2007). Under both moisture conditions, the GCA and SCA ratio

analysis showed that the additive gene action was more important in controlling inheritance of the major yield and yield component traits (Tables 6.4 and 6.5). In line with the current results, Tefera et al. (1997) reported higher additive genetic variance than dominance variance in controlling inheritance of plant height, panicle length, days-to-panicle emergence and days-to-maturity in tef. Yao et al. (2011) found that plant height was predominantly controlled by additive gene action in wheat genotypes tested using a half diallel mating design. Farshadfar and Amiri (2015) noted the importance of additive genes in controlling relative water content in bread wheat. Conversely, Farshadfar et al. (2013) found that the GCA and SCA effects were significant for plant height and peduncle length, while SCA was only significant for relative water content, indicating the predominance of non-additive gene action in controlling the inheritance of this trait. Rad et al. (2013) reported significant GCA and SCA effects for relative water content in bread wheat genotypes evaluated under moisture-stressed conditions.

In the moisture-stressed environments, GCA was significant for grain yield, harvest index and biomass yield, indicating the greater importance of additive gene action in controlling the inheritance of these characters. Likewise in the non-stressed environments, GCA was significant for grain yield, harvest index, number of productive tillers per plant and lodging index (Tables 6.4 and 6.5). Contrary to the current findings, Tefera et al. (1997) reported that dominance variance was larger than the additive variance in controlling the inheritance of grain yield in tef. In wheat Golparvar (2014) reported that both additive and non-additive gene actions controlled inheritance of harvest index, while additive gene action was responsible for biomass and grain yield. Zare-Kohanand Heidari (2014) reported the primary importance of additive genes in controlling inheritance of yield and its components in wheat. If additive gene action is predominant in self-pollinated species, then the breeder can effectively select desirable traits at various levels of inbreeding. This may result in substantial improvement of the trait of interest, because additive effects are fixable and readily transmissible from one generation to another. On the other hand, sufficient non-additive gene action may warrant the production of hybrids as it maximizes heterosis, if effective crosses can be performed (Falconer and Mackay, 1996; Chahal and Gosal, 2002; Sleper and Poehlman, 2006; Acquaah, 2007).

SCA showed significant interaction with the environment for panicle length and plant height only (Table 6.4). This is an indication of the consistency of the combining abilities of the tested populations across the environments for majority of the traits and therefore parents selected for their combining abilities can be used in any of the environments.

Table 6.4 Mean squares and significant tests from combined analysis of variance and general combining ability (GCA) and specific combining ability (SCA) and their ratios for the 12 quantitative traits evaluated under three moisture-stressed conditions

Traits	Mean squares									GCA SCA ratio
	ENV (2)	REP (ENV) (3)	Entries (14)	ENV x Entries (28)	GCA (4)	SCA (10)	GCA*ENV (8)	SCA*E NV (20)	Error (42)	
DPE	529.39**	27.43*	261.03**	13.10ns	811.82**	40.71**	11.76ns	8.25	4.13	0.98
DM	806.00**	11.31ns	331.65**	19.01*	1104.56**	22.48*	3.39ns	9.33	4.67	0.99
PNLG	425.00**	5.61ns	92.46**	20.48**	216.07**	43.01**	6.89ns	4.6	2.3	0.91
PLHT	5133.79**	33.25ns	490.68**	75.57**	1395.18**	128.87**	16.47ns	19.41	9.71	0.96
PDL	2.40ns	6.98*	3.84*	1.23ns	3.90ns	3.82ns	0.79ns	1.89	0.95	0.67
TILL	287.49**	34.10ns	108.26**	54.90ns	151.81*	90.84*	45.71ns	35.87	17.94	0.77
PNW	0.39**	0.01ns	0.05**	0.02ns	0.07**	0.04**	0.00ns	0.01	0.01	0.78
BM	505.50**	1.03ns	22.94**	10.00*	60.12**	8.07ns	0.66ns	5	2.5	0.94
YLD	21.46**	0.20ns	1.76*	1.36ns	3.49***	1.11ns	1.77ns	0.89	0.45	0.86
HI	0.08**	0.002ns	0.005ns	0.003ns	0.012**	0.001ns	0.002ns	0.003	0.003	0.96
LODG	121.14ns	279.12ns	172.89ns	105.50ns	214.08ns	179.34ns	17.28ns	203.72	203.72	0.70
RWC	16.29ns	5.65ns	62.36**	3.42ns	84.59**	50.56**	1.06ns	16.81	16.81	0.77

DPE = days-to-panicle emergence; DM = days-to-maturity; PLNG = panicle length (cm); PLHT = plant height (cm); PDL = peduncle length (cm); TILL = number of productive tillers per plant; PNW = main shoot panicle seed weight of the main shoot (g panicle<sup>-1</sup>); YLD= grain yield (g plant<sup>-1</sup>); BM = biomass yield (g plant<sup>-1</sup>); HI = harvest index; LODG = lodging index (%); RWC= relative water content (%)

ns, and \* and \*\* denote non-significant and significant at  $p \leq 0.05$  and  $p \leq 0.01$ , respectively

ENV x Entries = environment by entries interaction

Numbers in parenthesis show degrees of freedom

Table 6.5 Mean squares and significant tests from combined analysis of variance; and general combining ability (GCA) and specific combining ability (SCA) and their ratios for the 12 quantitative traits evaluated under optimum-moisture condition

Traits	Mean squares					GCA/SCA ratio
	REP (1)	Entries (14)	GCA (4)	SCA (10)	Error (14)	
DPE	13.56*	59.41**	151.84**	22.44**	3.73	0.93
DM	12.51*	107.66**	260.45**	46.54**	1.85	0.92
PNLG	3.90ns	63.75**	100.33**	49.12**	4.28	0.8
PLHT	10.15ns	373.98**	783.09**	210.34**	14.89	0.88
PDL	4.96ns	2.53ns	1.15ns	3.08ns	1.9	0.43
TILL	0.07ns	95.35ns	201.97*	52.70ns	40.75	0.88
PNW	0.06*	0.08**	0.15**	0.05**	0.01	0.86
BM	2.60ns	26.19ns	48.61ns	17.22ns	17.55	0.85
YLD	1.00ns	3.88ns	8.84*	2.44ns	2.8	0.88
HI	0.001ns	0.01*	0.01**	0.004ns	0.002	0.83
LODG	6.53ns	238.87**	575.71**	74.23ns	223.38	0.94
RWC	77.35ns	16.76ns	7.33ns	18.76ns	27.19	0.44

DPE= days-to-panicle emergence; DM= days-to-maturity; PLNG = panicle length; PLHT = plant height; PDL= peduncle length; TILL= number of productive tillers per plant; PNW = main shoot panicle seed weight of the main shoot; YLD= grain yield; BM= biomass yield; HI = harvest index; LODG= lodging index; RWC= relative water content  
ns, and \* and \*\* denote non-significant and significant at  $p \leq 0.05$  and  $p \leq 0.01$ , respectively  
Numbers in parenthesis show degrees of freedom

### 6.3.2 General combining ability effects

The GCA effects of the five parents under moisture-stressed and non-stressed conditions are presented on Table 6.6. Combining ability of parents gives useful information on the choice of parents in terms of expected performance of their progenies. Parents with high GCA effect can be utilized in a hybridization programme for selection of recombinants in segregating progenies. Therefore, knowledge of combining ability with mean performance of parents has great value in selecting suitable parents for any hybridization programme (Chahal and Gosal, 2002; Sleper and Poehlman, 2006). Under the moisture-stressed conditions, Genotype, DZ-Cr-387 showed significant and the highest positive effect for grain yield, panicle length and plant height, followed by genotype 9415, which had a significant positive GCA effect for panicle length, plant height and main shoot panicle seed weight. Under the non-stressed conditions, significant positive GCA values were noted for DZ-Cr-387 for panicle length and plant height; and for genotype 9415, for plant height and main shoot panicle seed weight. The genotype, Dschanger, also showed significant positive GCA values for biomass yield and panicle length and significant

negative GCAs for plant height and harvest index under the moisture-stressed conditions. In the non-stressed environments this genotype showed significant positive GCA effects for panicle length and plant height and significant negative GCAs for grain yield, harvest index, lodging index and main shoot panicle seed weight (Table 6.6).

Under the moisture stressed conditions, Genotype 222076, showed significant negative GCA values for panicle length, plant height, peduncle length, panicle seed weight, biomass yield, grain yield and relative water content and significant positive effects for number of productive tillers per plant and harvest index. In the non-stressed environments, this genotype had significant negative GCA values only for panicle length, plant height, main shoot panicle seed weight and biomass yield (Table 6.6). Regarding days-to-panicle emergence and days-to-maturity, negative GCA effects are important and in a desirable direction. Therefore, genotype, 222076, was better for its significantly negative GCA values, indicating its suitability in breeding for early maturity (Table 6.6).

Generally, the results from both the moisture-stressed and non-stressed conditions were similar; indicating that the genotypes, DZ-Cr-387 and 9415, were best combiners for increased grain yield and yield components and genotype 222076, was the best combiner for reduced days-to-maturity. Development of high yielding varieties for moisture-stressed areas is one of the major objectives of the tef improvement program in Ethiopia. The results indicated that the selected parents used in the current study would be useful for increasing the productivity of tef in moisture-stressed areas for their complementary characteristics, including high grain yield, and reduced days-to-maturity as a mechanism to escape late moisture-stress.

Table 6.6 General combining ability effects for 12 quantitative traits of five parental genotypes of tef evaluated under moisture-stressed and non-stressed conditions

Traits	Non-stressed					Moisture-stressed				
	DZ-Cr-387	205896	9415	Dschanger	222076	DZ-Cr-387	205896	9415	Dschanger	222076
DPE	3.18**	-0.41ns	2.06**	0.50ns	-5.34**	3.21**	0.88*	3.70**	-0.57ns	-7.22**
DM	-0.47ns	-0.37ns	3.67**	3.85**	-13.38**	2.99**	1.63**	2.86**	1.62**	-9.10**
PNLG	2.51**	-0.48ns	1.06ns	1.31*	-4.39**	2.12**	-0.23ns	1.03**	0.87*	-3.77**
PLHT	4.30**	-1.12ns	6.87**	2.26*	-12.31**	5.82**	1.27*	4.10**	-2.61**	-8.59**
PDL	-0.40ns	0.14ns	-0.10ns	0.37ns	-0.03ns	0.04ns	0.03ns	0.05ns	0.37ns	-0.48*
TILL	-1.13ns	-2.48ns	-3.14ns	0.40ns	6.35ns	0.89ns	-1.05ns	-1.55ns	-1.23ns	2.94**
PNW	0.01ns	0.02ns	0.15**	-0.06*	-0.12**	0.01ns	-0.03*	0.06**	0.01ns	-0.05**
BM	1.87ns	-1.07ns	1.63ns	0.12ns	-2.56*	0.43ns	0.31ns	0.20ns	1.11**	-2.04**
YLD	0.64ns	0.2ns	0.71ns	-1.36**	-1.32ns	0.40**	0.02ns	-0.12ns	0.07ns	-0.38*
HI	0.005ns	0.03*	0.01ns	-0.06**	0.07ns	0.02**	-0.01ns	-0.01ns	-0.02**	0.017*
LODG	1.28ns	-0.02ns	-3.12ns	-8.32**	8.4ns	-2.49ns	1.51ns	-2.72ns	2.11ns	0.87ns
RWC	0.27ns	-0.53ns	0.22ns	1.01ns	2.03ns	1.96**	0.32ns	-1.1*	0.66ns	-2.04**

DPE = days-to-panicle emergence; DM = days-to-maturity; PNLG = panicle length (cm); PLHT = plant height (cm); PDL = peduncle length (cm); TILL = number of productive tillers per plant; PNW = main shoot panicle seed weight of the main shoot (g panicle<sup>-1</sup>); YLD= grain yield (g plant<sup>-1</sup>); BM = biomass yield (g plant<sup>-1</sup>); HI = harvest index; LODG = lodging index (%); RWC= relative water content (%)  
ns, and \* and \*\* denote non-significant and significant at  $p \leq 0.05$  and  $p \leq 0.01$ , respectively

### 6.3.3 Specific combining ability effects

Estimates of SCA effects of the ten successful crosses under moisture-stressed and non-stressed conditions are presented in Table 6.7. The SCA effects provide important information about the non-additive gene effects that are the result of inter-allelic (epistasis) and intra-allelic (dominance) interactions (Falconer and Mackay, 1996). The extent of better performance of a specific cross depends on the extent of the favourable genes for the particular trait from the two parents complementing each other (Sleper and Poehlman, 2006). The cross, 205896 x Dschanger, had significant negative SCA for biomass yield under the moisture-stressed conditions, and it also had negative SCA for grain yield. The cross, Dschanger x 222076, had the highest significant negative SCA under the non-stressed conditions for harvest index, which could be because of its lower grain yield performance than its parents. For main shoot panicle seed weight, the cross, DZ-Cr-387 x 205896 and Dschanger x 222076 under the moisture-stressed conditions, and 9415 x 222076 in the non-stressed environments revealed significant high positive SCA values (Table 6.7).

Breeding for lodging tolerance is one of the main goals of tef improvement. However, there is limited useful natural variability of the crop for this trait (Assefa et al., 2011; Assefa et al., 2015). Negative SCA effects are in a desirable direction for lodging tolerance, i.e., towards a reduced lodging index. The cross, Dschanger x 222076, showed significant negative SCA under the non-stressed conditions (Table 6.7). However, this cross had low grain yield potential, limiting its potential to be selected for lodging tolerance.

In tef, relatively tall plants and long panicles are desired for their positive association with grain yield (Chanyalew, 2010; Mewa et al., 2013; Lule and Mengistu, 2014; Abraha et al., 2016). This is contrary to other cereal crops such as wheat and rice, in which short stature plants are desired for their better harvest index and reduced lodging (Khush, 2005). Therefore crosses with positive effects on plant height and panicle length would be preferred for their increased grain yield and lodging tolerance. Accordingly, crosses, 9415 x 222076 and 205896 x 222076 showed significant and high positive SCA values for plant height and panicle length under both the moisture-stressed and non-stressed conditions (Table 6.7). These crosses had better combinations of tall plants and long panicles than the mean performances of their respective parents and would be useful for transgressive breeding. Crosses from unrelated parents can produce better gene combinations than crosses from related parents (Sleper and Poehlman, 2006), which is noted from the cross 9415 x 222076. The female parent, 9415, had positive GCA effects while the male parent 222076 had negative GCA effects under both moisture stressed and non-stressed conditions (6.7).

Crosses, 205896 x Dschanger and 9415 x Dschanger, under the non-stressed conditions and DZ-Cr-387 x 222076 under both moisture-stressed and non-stressed conditions showed highly significant negative SCA effects for days-to-maturity in a desirable direction (Table 6.7). Genotype, 222076, was a good general combiner for early maturity but was a good specific combiner only in one cross (Table 6.7). The Tigray region is known for its erratic rainfall leading to moisture-stress especially during the grain filling period of the crops (Hadgu et al., 2013). Moisture-stress escape mechanisms such as early maturity are desired traits particularly if accompanied with good grain yield performance (Araus et al., 2002). Therefore the cross, DZ-Cr-387 x 222076, would have potential for selecting promising segregants with early maturity and high grain yield.

Relative water content is another drought tolerance mechanism. It is believed that high relative water content is the result of higher osmotic regulation of tissue with lower elasticity. It is one of

the indirect moisture-stress tolerance parameters, and it is relatively easy to measure (Roberts, 1987). Under moisture-stressed conditions, crosses DZ-Cr-387 x 205896, 9415 x 222076 and DZ-Cr-387 x Dschanger, had significant positive SCA values. The first two crosses had high SCA effects for grain yield in the moisture-stressed environments, in which improved relative water content could be one possible positive contributor. On the contrary, crosses, 205896 x 9415 and 205896 x Dschanger, showed significant negative SCA values (Table 6.7).

Table 6.7 Specific combing ability effects for yield and yield components of the ten F<sub>2</sub> tef populations evaluated under moisture-stressed and non-stressed conditions

Cross	Moisture stressed											
	DPE	DM	PNLG	PLHT	PDL	TILL	PNW	BM	YLD	HI	LODG	RWC
DZ-Cr-387 x 205896	-0.69ns	0.37ns	-0.31ns	-3.45*	0.85ns	0.67ns	0.13**	1.64*	0.66ns	0.013ns	-1.61ns	2.97**
DZ-Cr-387 x 9415	-4.04*	-0.44ns	1.68*	-2.81ns	0.62ns	0.42ns	0.03ns	0.05ns	0.23ns	-0.003ns	-4.55ns	-1.19ns
DZ-Cr-387 x Dschanger	1.20ns	2.15ns	0.83ns	2.76ns	-0.27ns	-1.44ns	-0.06ns	0.13ns	-0.25ns	-0.02ns	6.29ns	3.24**
205896 x 9415	-0.37ns	1.88ns	-1.97*	-3.16ns	-0.24ns	0.21ns	-0.09*	-0.81ns	-0.52ns	-0.002ns	-3.38ns	-3.09**
205896 x Dschanger	-2.73*	-2.05ns	-2.50**	-3.15ns	0.98ns	0.48ns	-0.06ns	-1.95*	-0.18ns	0.02ns	1.95ns	-2.27*
9415 x Dschanger	1.75ns	2.24ns	-0.76ns	-1.09ns	-0.12ns	1.36ns	-0.09*	0.59ns	-0.07ns	-0.01ns	0.52ns	1.67ns
DZ-Cr-387 x 222076	-0.04ns	-3.36**	-2.37**	-2.56ns	-0.53ns	0.26ns	-0.05ns	-0.40ns	-0.06ns	0.02ns	8.1ns	3.52ns
205896 x 222076	3.50*	0.74ns	4.99**	7.17**	-1.48**	-3.66ns	-0.02ns	0.99ns	-0.04ns	-0.02ns	10.9ns	0.31ns
9415 x 222076	1.98ns	-1.24ns	2.15*	5.68**	-0.54ns	-3.24ns	0.07*	0.15ns	0.26ns	0.01ns	-5.97ns	5.05**
Dschanger x 222076	-0.51ns	-0.89ns	1.54ns	1.92ns	-0.14ns	-2.57ns	0.12**	0.88ns	0.41ns	0.01ns	0.87ns	-0.50ns
Non-stressed												
DZ-Cr-387 x 205896	-1.06ns	2.64**	1.59ns	5.40*	1.97*	-2.70ns	0.11ns	-0.75ns	0.16ns	0.01ns	-0.58ns	-0.36ns
DZ-Cr-387 x 9415	-2.61ns	1.57ns	-0.53ns	-3.75ns	0.34ns	-3.08ns	-0.11ns	-0.42ns	-0.11ns	-0.01ns	1.02ns	1.79ns
DZ-Cr-387 x Dschanger	0.27ns	4.03**	3.39*	7.67*	0.47ns	0.52ns	0.04ns	5.63ns	0.15ns	-0.04ns	-6.78ns	-1.69ns
205896 x 9415	2.41ns	1.24ns	-0.82ns	-6.38*	0.26ns	3.84ns	0.03ns	-1.46ns	-0.37ns	0.01ns	0.32ns	-5.66ns
205896 x Dschanger	-0.63ns	-2.95**	-3.74*	-6.77*	0.81ns	2.25ns	-0.08ns	-1.78ns	0.26ns	0.03ns	8.52ns	3.82ns
9415 x Dschanger	1.14ns	-3.15**	-2.24ns	-3.55ns	0.40ns	4.96ns	-0.25**	1.59ns	-0.99 ns	-0.10ns	-1.88ns	-0.01ns
DZ-Cr-387 x 222076	1.51ns	-9.14**	-4.93**	-7.11*	-1.47ns	4.61ns	-0.03ns	-2.19ns	1.93 ns	0.1ns	-4.9ns	2.8ns
205896 x 222076	2.91*	6.25**	6.02**	12.34**	-2.70*	-5.37ns	-0.07ns	3.82ns	1.3ns	0.03ns	9.3ns	0.44ns
9415 x 222076	0.53ns	5.76**	5.41**	14.15**	-0.24ns	-6.60ns	0.33**	1.76ns	1.22ns	-0.007ns	3.7ns	1.62ns
Dschanger x 222076	0.57ns	10.52**	1.63ns	4.22ns	-0.59ns	-5.75ns	0.18*	-3.19ns	-3.13ns	-0.16**	-16.5*	-6.89ns

DPE = days-to-panicle emergence; DM = days-to-maturity; PLNG = panicle length (cm); PLHT = plant height (cm); PDL = peduncle length (cm); TILL = number of productive tillers per plant; PNW = main shoot panicle seed weight of the main shoot (g panicle<sup>-1</sup>); YLD= grain yield (g plant<sup>-1</sup>); BM = biomass yield (g plant<sup>-1</sup>); HI = harvest index; LODG = lodging index (%); RWC= relative water content (%)  
 ns, and \* and \*\* denote non-significant and significant at p ≤ 0.05 and p ≤ 0.0

### 6.3.4 Mean yield and yield components performance

The mean squares of the 12 quantitative traits of the 17 F<sub>2</sub> families and their eight parents tested in the moisture-stressed and non-stressed environments are presented in Table 6.8. Under the moisture-stressed conditions, test environments had a significant effect ( $p \leq 0.01$ ) on all the traits except lodging index and relative water content. Entries were also significantly different ( $p \leq 0.01$ ) for all the traits except peduncle length and lodging index. The environment by entry interaction effects was also significant ( $p \leq 0.01$ ) on all traits except peduncle length, lodging index and relative water content. In the non-stressed environments, the entries were significantly different ( $p \leq 0.01$ ) for all traits except peduncle length, relative water content, harvest index, biomass yield and grain yield. The coefficient of variation (CV) was relatively the higher for the majority of the traits in the moisture-stressed environments (Table 6.8), mainly was due to the high genotypic and environmental variance.

Table 6.8 Mean squares and coefficient of variation (CV) for 12 quantitative traits of 17 F<sub>2</sub> tef populations and 8 parents when evaluated under non-stressed and moisture-stressed conditions

Traits	Mean squares							
	Moisture-stressed					Non-stressed		
	Env. (df=2)	Entries (df=24)	Env x Entry (df=48)	Error (df=64)	CV (%)	Entries (df=24)	Error (df=16)	CV (%)
DPE	745.94**	274.39**	20.32**	7.71	16.01	88.86**	4.83	18.8
DM	1408.21**	306.48**	19.35**	9.65	4.81	104.46**	7.64	3.5
PLHT	6955.04**	465.86**	69.14**	21.46	2.68	349.21**	10.06	2.2
PNLG	654.49**	91.93**	15.45**	5.00	9.07	57.52**	2.84	28.4
PDL	6.99*	1.93ns	1.18ns	1.30	27.43	1.35ns	2.05	4.1
TILL	1092.09**	138.87**	72.58*	36.87	8.52	136.08*	45.22	5.5
LODG	109.81ns	205.40ns	133.20ns	174.44	30.73	362.07**	76.05	20.5
RWC	30.56ns	58.96**	2.65ns	14.36	27.13	17.28ns	17.01	4.6
PNW	0.43**	0.04**	0.01*	0.01	22.66	0.08**	0.01	24
HI	0.09**	0.00n*	0.00**	0.00	23.1	0.003ns	0.00	16.2
BM	654.91**	26.06**	8.67**	4.07	5.02	30.09ns	18.90	22.2
YLD	38.66**	2.77**	1.20*	0.70	16.45	3.94ns	2.73	16

DPE = days-to-panicle emergence; DM = days-to-maturity; PLNG = panicle length (cm); PLHT = plant height (cm); PDL = peduncle length (cm); TILL = number of productive tillers per plant; PNW = main shoot panicle seed weight of the main shoot (g panicle<sup>-1</sup>); YLD= grain yield (g plant<sup>-1</sup>); BM = biomass yield (g plant<sup>-1</sup>); HI = harvest index; LODG = lodging index (%); RWC= relative water content (%)  
Df= degrees of freedom

ns, \* and \*\* indicate non-significant, significant at  $p \leq 0.05$  and  $p \leq 0.01$ , respectively  
Env x entry = environment by entry interaction; CV= coefficient of variation in percent

Under the moisture-stressed conditions, crosses, DZ-Cr-387 x acc-ck-15, DZ-Cr-387 x 207832, DZ-Cr-387 x 205896, DZ-Cr-387 x 9415 and DZ-Cr-387 x 222076, showed higher mean grain yields of 4.42, 5.46, 4.88, 4.31 and 4.35 g plant<sup>-1</sup>, respectively. These crosses also showed greater grain yields than their respective parents. Under the non-stressed conditions, crosses, 9415 x 222076, DZ-Cr-387 x 207832, DZ-Cr-387 x 205896 and DZ-Cr-387 x 222076, had the best grain yield performances of 8.77, 9.04, 8.55, 8.54 g plant<sup>-1</sup>, respectively (Tables 6.9 and 6.10). The overall mean grain yield performance was 3.7 and 6.87 g plant<sup>-1</sup> in the moisture-stressed and non-stressed environments, respectively. About 47% of grain yield reduction could be attributed to moisture-stress. The three crosses: DZ-Cr-387 x 207832, DZ-Cr-387 x 205896 and DZ-Cr-387 x 222076, had superior grain yield performances under both the moisture-stressed and non-stressed conditions (Tables 6.9 and 6.10) and resulted from DZ-Cr-387, the female parent. This suggests that DZ-Cr-387 was the best parent for increasing grain yield under both moisture conditions. Genotypes with superior performance under diverse environments are desired to reduce the yield reduction due to environmental variability and at the same time to attain higher yields under optimum environments (Annicchiarico, 2002; Belay et al., 2008).

Crosses DZ-Cr-387 x 207832 and 9415 x 222076, had a high harvest index of 0.36, the former in the moisture-stressed environments and the latter in the non-stressed environments. The mean harvest index was 0.30 in both environments (Tables 6.9 and 6.10), is relatively low. Harvest indexes of 0.33-0.36 have been reported by Adnew et al. (2005), and these were higher than the harvest indexes of 0.21 reported by Chanyalew et al. (2010) and 0.11 to 0.17 by Shiferaw et al. (2012).

Crosses DZ-Cr-387 x 207832, 9415 x Dschanger and DZ-Cr-387 x 205896 in the moisture-stressed environments and DZ-Cr-387 x acc-ck-15, 9415 x Dschanger and DZ-Cr-387 x 9415 in the non-stressed environments produced superior biomass yields of 15.31, 15.05, 15.53, 27.73, 28.06 and 27.19 g plant<sup>-1</sup>, respectively (Tables 6.9 and 6.10). Tef straw is highly desired as livestock feed (Yami, 2013) and as mud-plastering/ cementing material of local houses. Both the seed and straw of tef fetch higher market prices than any other cereal crop in Ethiopia. Therefore a variety which is good for its superior grain yield and straw yield would have a high level of adoption. From the current study, crosses such as DZ-Cr-387 x 207832 and DZ-Cr-387 x 205896 are worth considering for their high grain yield and biomass yield.

The following crosses were selected for early maturity: 207832 x 222076 (105 days-to-maturity), DZ-Cr-385 x 222076 (103 days), acc-ck-15 x 222076 (106 days) and DZ-Cr-385 x acc-ck-15 (107 days) in the moisture-stressed environments and 207832 x 222076 (120 days), DZ-Cr-385 x 222076 (120 days) and DZ-Cr-387 x 222076 (115 days) under the non-stressed conditions (Tables 6.9 and 6.10). The female parent (DZ-Cr-387) in the cross DZ-Cr-387x 222076 is late maturing with high grain yield potential, while the male parent is early maturing. This allowed for selection of promising segregants with early maturity and better grain yield performance in both environments. Variety Tsedey (DZ-Cr-37) was released in 1983 for moisture-stressed growing areas (MoA, 2014). This variety has been widely grown for its high yield and relatively early maturity. Other improved varieties such as DZ-Cr-387 (RIL-127) (Gemechis) and DZ-Cr-385 (RIL 295) (Simada) were released in 2007 and 2009 for moisture-stressed areas. However, they were not widely adopted because of their poor grain yield performance despite their early maturity. Varieties Boset (DZ-Cr-409, RIL-50d) and Worekiyu (214746A) were released in 2012 and 2014, respectively (MoA, 2014) and are still under promotion for wide-area production. Cross DZ-Cr-387 x 222076, was identified in the current study as a valuable genetic resource for breeding tef for high grain yield and early maturity.

The tallest plants (64.81 cm) and the longest panicles of 30.39 cm under the moisture-stressed and 98.85 cm and 41.34 cm under the non-stressed conditions were recorded for the cross DZ-Cr-387 x Dschanger. The following crosses: DZ-Cr-387 x 205896 and DZ-Cr-387 x 9415 had tall plants and long panicles under the moisture-stressed and non-stressed conditions (Tables 6.9 and 6.10). These crosses resulted from parents selected for their tall plant heights and long panicles. However, the new families were late maturing and can be recommended for selection only for non-stressed environments.

Lodging index values ranged from 46.17 to 78.17% in the moisture-stressed environments and 32.9 to 84.4% in the non-stressed environments with a mean of 57.17% and 53.74%, respectively. The lodging indices of cross, DZ-Cr-387 x 9415 were 46.17% and 41% in the moisture-stressed and non-stressed environments, respectively. Crosses 9415 x Dschanger had a lodging index of 38.1%; DZ-Cr-387 x Dschanger a value of 42.0%; and Dschanger x 222076 that of 36.3% in the non-stressed environments (Tables 6.9 and 6.10). These are the best lodging indices in the study. These crosses exhibited intermediate to tall plant heights. There is a negative association between lodging index and plant height (Mewa et al., 2013; Lule and Mengistu, 2014; Abraha et al., 2016). The taller plants show reduced lodging for their thicker stems and longer roots (Ayele et al. 2001; Assefa et al., 2011).

The highest number of productive tillers per plant were noted for crosses 207832 x 222076, DZ-Cr-387 x 207832 and DZ-Cr-387 x 222076 (29, 25 and 25 in the moisture-stressed environments) and 207832 x 222076 and acc-ck-15 x 222076 with 50 and 43 in the non-stressed environments (Tables 6.9 and 6.10). The parents, 207832 and 222076, contributed for their high numbers of productive tillers per plant, albeit with a high lodging index.

Table 6.9 Mean values for 12 quantitative traits of the 17 F<sub>2</sub> tef populations and eight parents evaluated at three moisture-stressed sites

Entry	DPE	DMT	PHT	PLG	PDL	TILL	LOD	RWC	PN		BM	YLD
									W	HI		
Parents												
P1	70.95	125.66	76.53	30.97	3.66	23.18	46.83	76.03	0.30	0.33	12.58	4.00
P2	63.02	123.31	63.98	25.89	4.21	21.51	55.00	78.07	0.30	0.27	13.88	3.91
P3	69.05	122.96	68.43	27.53	4.63	19.46	57.17	73.04	0.51	0.28	13.56	3.66
P4	60.10	122.78	53.18	29.20	4.55	21.02	55.33	76.65	0.44	0.26	15.73	4.02
P5	41.16	102.63	40.43	19.67	3.84	19.98	55.00	73.96	0.18	0.34	5.81	1.97
P6	60.46	118.62	45.09	22.44	2.32	41.40	48.67	78.82	0.23	0.29	11.89	3.47
P7	52.62	110.62	44.46	19.14	3.45	21.20	65.33	74.52	0.21	0.29	13.26	3.98
P8	46.45	102.09	40.35	15.74	4.17	34.70	51.33	69.79	0.23	0.33	7.79	2.60
Crosses												
P3 x p8	58.74	114.64	58.65	27.07	3.00	18.19	55.50	77.28	0.32	0.32	11.44	3.44
P1 x p6	62.36	119.20	62.26	29.61	3.27	21.06	61.17	77.65	0.23	0.30	14.23	4.24
P2 x p7	54.29	115.74	48.99	21.41	5.23	23.17	52.67	68.46	0.21	0.27	11.05	2.98
P7 x p8	48.44	105.06	44.03	19.16	4.23	26.80	52.33	75.43	0.25	0.27	11.67	3.21
P2 x p3	65.17	126.05	61.05	25.41	4.10	18.91	51.33	72.56	0.26	0.28	12.85	3.17
P1 x p7	62.50	117.77	56.26	24.42	5.47	24.68	54.17	77.21	0.29	0.36	15.31	5.46
P3 x p4	65.85	126.41	59.25	27.71	4.55	19.88	55.83	77.65	0.30	0.26	15.05	3.67
P1 x p4	64.81	126.45	64.81	30.39	4.40	19.53	61.83	82.25	0.29	0.28	14.83	4.01
P1 x p2	64.37	124.68	62.48	28.15	5.17	21.81	53.33	81.64	0.44	0.32	15.53	4.88
P2 x p8	57.83	112.59	56.10	27.78	2.43	17.22	66.00	76.18	0.19	0.28	12.29	3.31
P4 x p8	52.38	111.47	50.01	24.32	4.44	18.27	55.67	73.60	0.31	0.30	12.76	3.79
P1 x p3	63.83	125.10	65.95	31.39	4.96	21.07	46.17	76.10	0.42	0.31	13.83	4.31
P5 x p8	45.61	102.89	42.99	20.00	4.07	14.69	78.17	71.67	0.25	0.26	9.70	2.57
P2 x p4	58.54	119.58	54.35	24.71	5.63	19.50	61.50	75.11	0.24	0.29	12.63	3.69
P1 x p8	53.34	111.55	47.46	22.39	3.96	25.31	59.00	75.72	0.29	0.34	12.57	4.35
P6 x p8	50.07	106.07	45.59	20.21	4.14	24.43	73.83	69.92	0.17	0.32	12.98	4.12
P5 x p6	50.25	106.72	46.81	21.60	3.99	22.62	56.17	78.68	0.24	0.31	11.76	3.66
Mean	57.69	116.03	54.38	24.65	4.15	22.38	57.17	75.52	0.28	0.30	12.60	3.70
LSD (0.05)	3.20	3.58	5.34	2.58	1.31	7.00	15.23	4.37	0.10	0.06	2.33	0.97

p1= DZ-Cr-387, p2= 205896, p3= 9415, p4 = Dschanger, p5= DZ-Cr-385, p6= acc-ck-15, p7= 207832, p8 = 222076

DPE = days-to-panicle emergence; DM = days-to-maturity; PLNG = panicle length (cm); PLHT = plant height (cm); PDL = peduncle length (cm) ; TILL = number of productive tillers per plant; PNW = main shoot panicle seed weight of the main shoot (g panicle<sup>-1</sup>); YLD= grain yield (g plant<sup>-1</sup>); BM = biomass yield (g plant<sup>-1</sup>); HI = harvest index; LODG = lodging index (%); RWC= relative water content (%)

Table 6.10 Mean values for 12 quantitative traits of the 17 F<sub>2</sub> tef populations and eight parents evaluated at non-stressed environment

Entry	DPE	DMT	PHT	PLG	PDL	TILL	LOD	RWC	PN W	HI	BM	YLD
Parents												
P1	76.01	129.3	90.14	38.53	3.31	24.46	56.7	85.05	0.52	0.28	26.77	7.68
P2	61.88	126.3	76.86	30.18	5.2	22.84	43.3	87.58	0.64	0.3	22.5	6.61
P3	70.39	133.6	95.98	33.45	4.22	17.87	32.9	88.09	0.82	0.33	25.5	8.47
P4	66.27	132.8	86.9	37.09	4.85	28.56	42.1	92.3	0.51	0.27	23.14	6.27
P5	47.21	111.6	44.63	17.55	5.41	29.94	70.2	88.56	0.13	0.29	15.52	4.6
P6	61.89	133.7	80.56	29.45	5.12	50.33	46	80.27	0.3	0.27	20.78	5.53
P7	54.18	110.6	73.72	29.33	3.76	17.97	15.7	90.33	0.88	0.26	25.53	6.4
P8	49.77	111.7	41.32	18.4	5.43	47.28	76.9	83.48	0.12	0.33	14.5	4.77
Crosses												
P3 x p8	65.09	130.4	89.61	36.26	5.39	23.9	59.6	89.73	0.85	0.36	23.74	8.77
P1 x p6	62.17	130.2	86.91	34.54	5.14	29.68	70.8	94.92	0.62	0.3	27.73	8.05
P2 x p7	63.05	127.7	71.95	27.88	4.33	27.41	53.6	89.82	0.43	0.32	21.27	6.69
P7 x p8	57.24	120.4	57.87	21.46	4.25	50.35	84.4	93.68	0.2	0.31	18.03	5.81
P2 x p3	70.56	134.2	80.61	32.26	4.97	31.49	50.5	83.66	0.72	0.34	23.66	7.76
P1 x p7	69.08	124.5	81.19	30.56	5.49	34.4	67.7	90.49	0.47	0.33	26.83	9.04
P3 x p4	71.33	134.9	86.63	32.15	5.57	31.8	38.1	88.7	0.32	0.2	28.06	5.92
P1 x p4	72.1	135.9	98.85	41.34	6.12	26.86	42	91.21	0.53	0.23	32.78	7.44
P1 x p2	68.5	129.8	92.29	37.48	6.82	21.78	50.3	87.6	0.72	0.33	25.74	8.55
P2 x p8	68.27	126.2	85.69	37.1	3.2	27.17	59.8	88.96	0.32	0.33	23.97	7.96
P4 x p8	63.67	132	78.64	31.21	5.78	29.95	36.3	84.19	0.41	0.19	20.16	3.79
P1 x p3	69.39	131.9	90.08	37.03	4.71	17.96	41	91.26	0.65	0.3	27.19	8.26
P5 x p8	50.31	120.3	55.96	22.61	4.57	35.09	63	89.61	0.21	0.3	13.83	4.21
P2 x p4	65.96	129.1	77.82	31.63	6.5	24.34	51.5	92.38	0.51	0.29	22.28	6.41
P1 x p8	64.92	115.3	70.6	26.42	4.9	31.32	62.4	86.47	0.41	0.36	24.01	8.54
P6 x p8	52.04	127.7	69.59	27.03	5.76	42.48	67.5	87.93	0.33	0.3	20.33	6.02
P5 x p6	53.5	121.3	71.39	29.38	5.16	31.27	61.2	92.61	0.41	0.33	24.92	8.18
Mean	63.95	127.7	79.16	31.55	5.22	30.43	56.45	89.6	0.48	0.30	23.80	7.14
LSD (0.05)	5.51	6.9	7.96	4.23	3.6	16.87	21.87	10.34	0.25	0.12	10.9	4.14

p1= DZ-Cr-387, p2= 205896, p3= 9415, p4 = Dschanger, p5= DZ-Cr-385, p6= acc-ck-15, p7= 207832, p8 = 222076

DPE = days-to-panicle emergence; DMT = days-to-maturity; PNG = panicle length (cm); PHT = plant height (cm); PDL = peduncle length (cm) ; TILL = number of productive tillers per plant; PNW = main shoot panicle seed weight of the main shoot (g panicle<sup>-1</sup>); YLD= grain yield (g plant<sup>-1</sup>); BM = biomass yield (g plant<sup>-1</sup>); HI = harvest index; LOD = lodging index (%); RWC= relative water content

### 6.3.5 Genotypic and phenotypic variance

The crosses showed a wide range of GCV and PCV under the moisture-stressed and non-stressed conditions (Tables 6.11 and 6.12). Relatively high GCV ranges of 20 to 68%, 16 to 49%, 11 to 63%, 17 to 51%, 9 to 79% in the moisture-stressed and 19 to 65%, 14 to 67%, 18 to 55%, 18 to 40% and 27 to 62% in the non-stressed environments were noted for grain yield, biomass yield, number of productive tillers per plant, main shoot panicle seed weight and peduncle length, respectively. In line with the current findings Tefera et al. (2008) reported higher GCV values for number of productive tillers per plant (14 to 43%), main shoot panicle seed weight (16% to 26%) and grain yield (23 to 91%) in F<sub>2</sub> tef populations evaluated under

optimum conditions. Shiferaw et al (2012) reported high GCV values of 41% and 74% for seed weight in moisture-stressed and non-stressed environments, respectively. Similarly Admass and Belay (2011) reported high GCV values of 41% and 36% for grain yield in non-stressed and moisture-stressed environments, respectively.

The highest GCV and PCV values for grain yield were recorded for crosses DZ-Cr-385 x 222076 (48% and 71%) and DZ-Cr-385 x acc-ck-15 (68% and 83%) in the moisture-stressed environments; and DZ-Cr-387 x acc-ck-15 (64% and 77%), DZ-Cr-387 x 207832 (61% and 72%), DZ-Cr-387 x 222076 (63% and 71%) and DZ-Cr-385 x acc-ck-15 (65% and 75%) in the non-stressed environment (Tables 6.11 and 6.12). Relatively high GCV and PCV values were noticed for days-to-maturity from the cross DZ-Cr-387 x 222076 at 9% and 10% followed by 205896 x 222076 (9% and 9%) and 205896 x 207832 (9% and 9%) in the moisture-stressed environment and for DZ-Cr-385 x 222076 (7% and 6%) and DZ-Cr-385 x acc-ck-15 (5% and 5%) in the non-stressed environment, in that order. The high GCV and PCV values offer greater opportunity for selection of early maturing genotypes combined with high grain yield potential, which is one of the approaches to improve tef productivity in moisture-stressed environments. Overall, the results from this study indicated that crosses derived from parents with complementary traits provided high variability (Tables 6.11 and 6.12).

Table 6.11 Genotypic (GCV) and phenotypic (PCV) coefficients of variance for 12 quantitative traits of the 17 F<sub>2</sub> segregating tef populations tested under moisture-stressed conditions

Cross	CV	DPE	DMT	PHT	PLG	PDL	TILL	PNW	BM	YLD
9415 x 222076	GCV	13.70	7.04	19.30	18.85	-	-	54.93	34.60	31.54
	PCV	15.88	8.86	24.91	25.06	94.05	43.78	63.86	53.81	55.98
DZ-Cr-387 x acc-ck-15	GCV	11.23	4.87	17.37	17.50	62.63	32.55	45.51	-	47.99
	PCV	13.27	7.31	21.89	23.20	106.66	49.25	72.74	30.24	64.17
205896 x 207832	GCV	13.37	9.07	21.44	16.16	25.38	62.82	-	-	-
	PCV	16.71	9.60	29.18	26.30	57.03	71.30	51.63	42.87	62.75
207832 x 222076	GCV	15.70	5.28	17.35	7.38	53.80	37.98	-	21.36	19.74
	PCV	18.52	6.90	26.49	25.41	80.99	45.64	35.49	43.50	61.37
205896 x 9415	GCV	6.35	2.96	14.75	20.40	41.29	55.28	44.64	28.82	29.24
	PCV	10.49	4.81	22.71	26.52	77.74	62.96	83.42	61.67	74.44
DZ-Cr-387 x 207832	GCV	11.28	6.09	22.03	18.38	52.46	29.91	39.03	25.98	27.92
	PCV	13.69	6.97	26.78	24.86	69.06	40.99	57.67	44.90	43.40
9415 x Dschanger	GCV	-	-	-	7.26	58.05	21.00	-	31.52	-
	PCV	7.83	5.18	22.45	18.19	82.33	46.19	41.19	57.19	47.26
DZ-Cr-387 x Dschanger	GCV	7.28	4.08	23.70	24.95	78.72	-	51.97	46.10	41.35
	PCV	10.24	6.04	30.03	27.87	101.88	38.92	70.92	61.67	56.48
DZ-Cr-387 x 205896	GCV	7.48	4.14	22.02	19.62	58.88	37.21	24.63	28.34	33.85
	PCV	10.61	5.08	26.27	23.20	77.77	49.09	51.76	53.61	49.83
205896 x 222076	GCV	11.54	8.45	23.92	20.99	62.61	10.46	51.02	27.42	-
	PCV	14.36	9.26	28.39	25.04	133.33	42.78	71.58	50.72	52.50
Dschanger x 222076	GCV	14.26	6.23	17.80	21.67	56.64	-	30.41	-	-
	PCV	16.83	8.48	28.65	27.14	84.82	46.00	44.10	35.98	43.90
DZ-Cr-387 x 9415	GCV	6.56	0.35	18.15	19.25	56.57	32.36	24.08	15.74	46.28
	PCV	9.94	4.30	23.31	23.37	83.22	48.67	51.83	54.52	61.85
DZ-Cr-385 x 222076	GCV	14.09	4.75	15.12	16.19	9.01	18.39	-	48.64	47.82
	PCV	17.79	6.51	23.81	24.13	72.86	64.89	36.71	63.38	70.99
205896 x Dschanger	GCV	12.29	7.02	16.28	17.68	58.00	36.24	-	-	36.45
	PCV	15.41	8.18	28.57	23.77	75.48	53.37	49.80	51.14	67.23
DZ-Cr-387 x 222076	GCV	18.52	9.12	18.86	21.95	64.09	45.14	27.09	42.39	42.25
	PCV	20.17	10.08	25.47	27.35	92.99	54.78	51.94	58.01	55.57
acc-ck-15 x222076	GCV	14.81	1.62	21.87	6.89	58.98	55.91	16.47	42.24	43.86
	PCV	17.12	7.39	28.38	25.36	87.34	67.90	52.39	57.82	62.50
DZ-Cr-385 x acc-ck-15	GCV	12.73	5.62	19.37	11.54	30.06	37.90	-	23.66	67.98
	PCV	16.17	8.27	25.32	22.71	71.80	55.80	47.11	55.97	82.56

CV=Coefficient of variation; DPE = days-to-panicle emergence; DMT = days-to-maturity; PNG = panicle length (cm); PHT = plant height (cm); PDL = peduncle length (cm) ; TILL = number of productive tillers per plant; PNW = main shoot panicle seed weight of the main shoot (g panicle<sup>-1</sup>); YLD= grain yield (g plant<sup>-1</sup>); BM = biomass yield (g plant<sup>-1</sup>); HI = harvest index; LOD = lodging index (%); RWC= relative water content (%)

Dash line (-) indicates absence of genetic variance

Table 6.12 Genotypic (GCV) and phenotypic (PCV) coefficients of variance for 12 quantitative traits of the 17 F<sub>2</sub> segregating tef populations tested under non-stressed condition

Cross	CV	DPE	DMT	PHT	PLG	PDL	TILL	PNW	BM	YLD
	GCV	3.04	3.19	7.34	11.62	47.35	17.62	31.41	42.77	36.66
9415 x 222076	PCV	4.41	4.27	9.52	13.22	73.11	47.14	36.34	55.17	52.65
	GCV	7.46	3.48	4.33	10.09	51.01	34.32	26.74	62.06	63.65
DZ-Cr-387 x acc-ck-15	PCV	8.44	4.07	8.73	11.77	75.75	47.72	38.53	72.75	76.62
	GCV	5.11	4.67	7.05	10.92	27.06	17.68	-	19.97	18.90
205896 x 207832	PCV	6.17	5.51	12.20	14.51	54.02	40.08	37.60	54.65	54.91
	GCV	9.07	2.87	-	11.43	-	33.36	-	-	-
207832 x 222076	PCV	10.18	4.36	12.20	16.59	65.28	40.65	46.18	44.33	53.40
	GCV	5.65	2.73	9.30	11.59	40.26	11.68	24.82	22.79	33.46
205896 x 9415	PCV	6.19	3.77	11.01	13.69	61.61	38.60	35.96	51.02	58.68
	GCV	3.75	4.12	-	13.09	55.21	32.53	24.71	15.82	61.47
DZ-Cr-387 x 207832	PCV	5.48	4.89	9.30	14.67	68.44	48.47	53.66	50.06	71.87
	GCV	5.08	1.51	7.00	14.15	48.44	46.72	21.92	14.44	21.45
9415 x Dschanger	PCV	6.09	3.00	10.06	16.23	68.83	54.95	53.51	46.42	66.17
	GCV	4.31	2.64	-	8.79	49.94	-	28.01	36.10	38.73
DZ-Cr-387 x Dschanger	PCV	5.86	3.45	8.67	10.50	66.92	34.87	43.12	52.43	60.25
	GCV	4.76	2.42	3.70	10.18	41.27	-	36.03	-	49.46
DZ-Cr-387 x 205896	PCV	5.68	3.36	8.26	11.72	54.79	35.68	43.13	42.73	62.91
	GCV	5.43	2.68	10.99	13.46	50.92	-	38.99	40.48	33.91
205896 x 222076	PCV	6.18	3.95	13.41	15.28	107.42	33.20	56.29	53.44	53.61
	GCV	5.29	4.08	-	11.48	39.66	40.24	18.28	52.89	48.40
Dschanger x 222076	PCV	6.71	4.90	10.24	14.88	62.73	51.50	39.76	69.25	88.36
	GCV	5.76	2.69	11.19	15.88	62.44	-	38.83	28.54	38.69
DZ-Cr-387 x 9415	PCV	6.60	3.54	12.83	16.69	83.90	40.00	50.47	51.21	55.15
	GCV	4.61	5.60	4.57	14.48	-	44.94	20.83	40.82	41.69
DZ-Cr-385 x 222076	PCV	6.47	6.24	14.31	17.31	68.73	51.91	47.30	68.66	73.00
	GCV	5.73	2.06	8.18	2.18	27.30	27.49	27.92	29.33	37.57
205896 x Dschanger	PCV	6.66	3.37	12.64	10.12	44.88	43.70	49.96	59.00	65.20
	GCV	6.97	4.03	13.14	22.98	32.61	51.06	40.38	67.03	62.59
DZ-Cr-387 x 222076	PCV	7.98	4.93	17.24	24.37	73.69	58.65	54.85	77.29	71.44
	GCV	6.23	3.81	9.40	17.57	37.67	54.55	27.72	49.75	35.48
acc-ck-15 x222076	PCV	7.65	4.62	13.16	20.14	67.92	60.52	56.86	64.33	63.45
	GCV	6.57	4.73	6.27	13.56	-	36.19	18.31	42.32	64.79
DZ-Cr-385 x acc-ck-15	PCV	7.81	5.29	11.30	15.60	69.71	44.42	36.96	58.64	74.54

CV=coefficient of variation; DPE = days-to-panicle emergence; DMT = days-to-maturity; PNG = panicle length (cm); PHT = plant height (cm); PDL = peduncle length (cm) ; TILL = number of productive tillers per plant; PNW = main shoot panicle seed weight of the main shoot (g panicle<sup>-1</sup>); YLD= grain yield (g plant<sup>-1</sup>); BM = biomass yield (g plant<sup>-1</sup>); HI = harvest index; LOD = lodging index (%); RWC= relative water content (%)

Dash line (-) indicates absence genetic variance

### 6.3.6 Heritability and genetic advance

Heritability and genetic advance of yield and yield components for each cross under moisture-stressed and non-stressed conditions are presented in Tables 6.13 and 6.14, respectively. Relative to others, days-to-panicle emergence and days-to-maturity, showed intermediate to high heritability and low genetic advances in both environments. Under the moisture-stressed conditions, the cross, 205896 x 207832, showed a high heritability of 89.18% and a genetic advance of 15.02% for days-to-maturity, followed by 205896 x 222076 (83.32% and 13.54%) and DZ-Cr-387 x 222076 (81.85% and 14.48%) in that order. Under the non-stressed conditions, crosses DZ-Cr-385 x 222076 (80.53% and 8.82%) and DZ-Cr-385 x acc-ck-15

(80.14% and 7.44%) depicted high heritability and genetic advance values (Tables 6.13 and 6.14). The current results agree with those of Admas and Belay (2011) who in tef recombinant inbred lines reported high heritability values of 82.6% and 93.1% for days-to-panicle emergence and 97.3% and 84.5% for days-to-maturity under non-stressed and moisture-stressed conditions, respectively. The authors indicated that under non-stressed and moisture-stressed conditions, genetic advances were higher for days-to-panicle emergence with 20.9% and 22.4% and lower for days-to-maturity (8.9% and 6.7%). Assefa et al. (2000) and Assefa et al. (2001) reported high heritability values for days-to-panicle emergence of 68.96% and 72.52%, but lower values for days-to-maturity at 32.50% and 28.96%, respectively. The authors also reported a low genetic advance of 11.65% and 11.07% for days-to-panicle emergence and 1.83% and 1.88% for days-to-maturity. Shiferw et al. (2012) reported high heritability values of 84% and 94% for days-to-maturity under non-stressed and moisture-stressed conditions respectively. Ayalew et al. (2011) determined heritability of 80.73% and 65.03% for days-to-panicle emergence and days-to-maturity, respectively. The low genetic advance and high heritability for days-to-panicle emergence and days-to-maturity found in this and previous studies suggest a low level of the heritable components (additive genes), implying reduced responses to selection for these traits.

High heritability values of 68% and 76% were measured for grain yield from the cross DZ-Cr-385 x acc-ck-15, followed by DZ-Cr-387 x 222076 (58% and 77%) and DZ-Cr-387 x acc-ck-15 (56% and 69%) in the moisture-stressed and non-stressed environments, respectively (Tables 6.13 and 6.14). Genetic advance estimates for grain yield were also the highest for these crosses with 98%, 56% and 63% under the moisture-stressed conditions and 99%, 96% and 93% under the non-stressed conditions. These results suggest that selection could be effective in these segregating populations. Heritability and genetic advance of the above crosses were higher under the non-stressed than in the moisture-stressed environments because of the lower environmental effects on the genotypes. Decreased heritability values of grain yield under the moisture-stressed conditions than under the non-stressed conditions have been reported in a previous study (Shiferaw et al., 2012). The ranges of heritability for grain yield were from 10.34% to 67.80% in the moisture-stressed environments and 10.51% to 76.75% in the non-stressed environments. These were in line with the report of Tefera et al. (2008) who found heritability ranging from 16.79% to 80.18% in the F<sub>2</sub> tef populations. Admas and Belay (2011) reported higher heritability and genetic advance for grain yield in that order of 93.4% and 80.9% under non-stressed conditions and 85.7% and 68.6% in moisture-stressed environments.

Conversely, lower heritability values of 22.57% to 25.68% and genetic advances of 0.45% to 15.82% were reported for grain yield by Assefa et al. (2000, 2001) and Tefera et al. (2003). Ayalew et al. (2011) reported an intermediate heritability value of 57.32% for grain yield.

Furthermore, the other traits such as plant height, panicle length, peduncle length, number of productive tillers per plant, main shoot panicle seed weight and biomass yield showed a wide range of heritability and genetic advance values (Tables 6.13 and 6.14). Tefera et al. (2008) reported variable heritability values for  $F_2$  tef populations evaluated under optimum conditions including for plant height (5.97% to 45.93%), panicle length (0.81% to 36.76%), number of productive tillers per plant (10.17% to 53.41%) and main shoot panicle seed weight (20 to 74%). In moisture-stressed and non-stressed environments, Shiferaw et al. (2012) reported high heritability values of 89% and 73% for biomass yield, 80% and 93% for plant height and 96% and 86% for seed weight, respectively. For tef landraces, Ayalew et al. (2011) obtained heritability values of 64.33%, 55.55%, 51.90%, 20.38%, 54.58% and 57.58% and genetic advance values of 12.79%, 6.46%, 3.54%, 0.07%, 0.24% and 0.24% for plant height, panicle length, peduncle length, number of productive tillers per plant, biomass yield and grain yield, respectively.

Table 6.13 Broad sense heritability (H) and genetic advance as percent of mean (GAM) for 12 agronomic traits of the 17 F<sub>2</sub> tef populations tested under moisture-stressed conditions

Cross	Parameter	DPE	DMT	PHT	PLG	PDL	TILL	PNW	BM	YLD
9415 x 222076	H	74.43	63.00	60.07	56.54	-	-	74.00	41.35	31.74
	GAM	20.75	9.80	26.26	24.87	-	-	82.93	39.05	31.19
DZ-Cr-387 x acc-ck-15	H	71.58	44.36	62.92	56.92	34.49	43.67	39.15	-	55.93
	GAM	16.67	5.69	24.18	23.18	64.55	37.75	49.98	-	62.99
205896 x 207832	H	64.02	89.18	53.98	37.77	19.81	77.65	-	-	-
	GAM	18.77	15.02	27.64	17.43	19.83	97.16	-	-	-
207832 x 222076	H	71.87	58.59	42.87	8.43	44.13	69.25	-	24.12	10.34
	GAM	23.36	7.09	19.93	3.76	62.73	55.47	-	18.41	11.14
205896 x 9415	H	36.69	37.89	42.20	59.15	28.21	77.09	28.64	21.83	15.43
	GAM	6.75	3.20	16.81	27.53	38.48	85.18	41.93	23.63	20.15
DZ-Cr-387 x 207832	H	67.94	76.26	67.69	54.68	57.69	53.24	45.79	33.47	41.38
	GAM	16.32	9.33	31.81	23.86	69.93	38.30	46.34	26.37	31.52
9415 x Dschanger	H	-	-	-	15.94	49.71	20.68	-	30.38	-
	GAM	-	-	-	5.09	71.83	16.76	-	30.49	-
DZ-Cr-387 x Dschanger	H	50.52	45.66	62.27	80.16	59.71	-	53.70	55.89	53.59
	GAM	9.08	4.84	32.82	39.20	106.76	-	66.84	60.49	53.12
DZ-Cr-387 x 205896	H	49.65	66.34	70.25	71.56	57.32	57.48	22.64	27.94	46.13
	GAM	9.25	5.91	32.40	29.13	78.24	49.52	20.56	26.29	40.34
205896 x 222076	H	64.51	83.32	70.99	70.31	22.05	5.98	50.79	29.21	-
	GAM	16.26	13.54	35.37	30.89	51.60	4.49	63.81	26.01	-
Dschanger x 222076	H	71.74	53.89	38.60	63.75	44.59	-	47.55	-	-
	GAM	21.19	8.02	19.41	30.36	66.38	-	36.80	-	-
DZ-Cr-387 x 9415	H	43.57	0.68	60.61	67.81	46.20	44.22	21.59	8.33	56.00
	GAM	7.60	0.05	24.79	27.82	67.47	37.77	19.64	7.97	60.78
DZ-Cr-385 x 222076	H	62.67	53.11	40.31	45.04	1.53	8.03	-	58.90	45.38
	GAM	19.57	6.07	16.84	19.07	1.96	9.15	-	65.52	56.54
205896 x Dschanger	H	63.56	73.71	32.46	55.30	59.06	46.12	-	-	29.40
	GAM	17.19	10.58	16.27	23.07	78.23	43.20	-	-	34.69
DZ-Cr-387 x 222076	H	84.31	81.85	54.88	64.38	47.51	67.89	27.20	53.40	57.79
	GAM	29.84	14.48	24.53	30.91	77.53	65.27	24.79	54.36	56.37
acc-ck-15 x222076	H	74.88	4.83	59.38	7.39	45.61	67.81	9.88	53.36	49.23
	GAM	22.50	0.63	29.58	3.29	69.91	80.81	9.09	54.15	54.01
DZ-Cr-385 x acc-ck-15	H	62.03	46.17	58.52	25.81	17.53	46.12	-	17.88	67.80
	GAM	17.60	6.70	26.00	10.29	22.09	45.16	-	17.56	98.23

DPE= days-to-panicle emergence; DMT= days-to-maturity; PNG = panicle length (cm); PHT = plant height (cm); PDL= peduncle length (cm) ; TILL= number of productive tillers per plant; PNW = main shoot panicle seed weight of the main shoot (g panicle<sup>1</sup>); YLD= grain yield (g plant<sup>1</sup>); BM= biomass yield (g plant<sup>1</sup>); HI = harvest index; LOD= lodging index (%); RWC= relative water content (%) Dash line (-) indicates absence of heritability

Table 6.14 Broad sense heritability (H) and genetic advance as percent of mean (GAM) for 12 quantitative traits of the 17 F<sub>2</sub> tef populations tested under non-stressed conditions

Cross	Parameter	DPE	DMT	PHT	PLG	PDL	TILL	PNW	BM	YLD
9415 x 222076	H	47.36	56.01	59.51	77.22	41.94	13.97	74.74	60.10	48.49
	GAM	3.67	4.19	9.94	17.92	53.81	11.56	47.66	58.19	44.80
DZ-Cr-387 x acc-ck-15	H	78.10	73.09	24.60	73.42	45.34	51.74	48.17	72.77	69.01
	GAM	11.57	5.22	3.77	15.17	60.27	43.33	32.57	92.91	92.80
205896 x 207832	H	68.70	71.86	33.36	56.70	25.09	19.46	-	13.35	11.84
	GAM	7.43	6.95	7.14	14.43	23.79	13.69	-	12.80	11.41
207832 x 222076	H	79.36	43.36	-	47.50	-	67.34	-	-	-
	GAM	14.17	3.32	-	13.83	-	48.04	-	-	-
205896 x 9415	H	83.35	52.41	71.44	71.69	42.69	9.16	47.65	19.95	32.52
	GAM	9.05	3.47	13.80	17.22	46.16	6.20	30.07	17.87	33.49
DZ-Cr-387 x 207832	H	46.81	70.83	-	79.63	65.07	45.06	21.21	9.99	73.14
	GAM	4.50	6.08	-	20.51	78.16	38.32	19.97	8.77	92.26
9415 x Dschanger	H	69.68	25.48	48.41	75.98	49.52	72.29	16.78	9.68	10.51
	GAM	7.45	1.34	8.54	21.65	59.82	69.71	15.76	7.89	12.21
DZ-Cr-387 x Dschanger	H	54.04	58.45	-	70.02	55.68	-	42.18	47.41	41.32
	GAM	5.56	3.54	-	12.91	65.40	-	31.92	43.63	43.69
DZ-Cr-387 x 205896	H	70.07	51.69	20.02	75.40	56.75	-	69.78	-	61.81
	GAM	6.99	3.05	2.90	15.51	54.56	-	52.82	-	68.24
205896 x 222076	H	77.07	45.82	67.22	77.55	22.47	-	47.97	57.39	40.00
	GAM	8.36	3.18	15.82	20.79	42.36	-	47.39	53.82	37.64
Dschanger x 222076	H	62.18	69.24	-	59.57	39.97	61.06	21.14	58.33	30.01
	GAM	7.33	5.96	-	15.55	44.00	55.18	14.75	70.89	46.53
DZ-Cr-387 x 9415	H	76.03	58.00	75.98	90.54	55.39	-	59.18	31.08	49.22
	GAM	8.81	3.60	17.11	26.52	81.56	-	52.42	27.93	47.63
DZ-Cr-385 x 222076	H	50.77	80.53	10.20	69.95	-	74.95	19.39	35.35	32.62
	GAM	5.77	8.82	2.56	21.25	-	68.28	16.10	42.60	41.79
205896 x Dschanger	H	74.04	37.24	41.90	4.62	37.00	39.59	31.23	24.71	33.20
	GAM	8.66	2.20	9.30	0.82	29.14	30.36	27.38	25.58	37.99
DZ-Cr-387 x 222076	H	76.33	66.89	58.08	88.93	19.59	75.78	54.19	75.22	76.75
	GAM	10.69	5.78	17.57	38.03	25.33	78.00	52.17	102.03	96.23
acc-ck-15 x222076	H	66.29	67.75	51.07	76.08	30.76	81.25	23.77	59.80	31.28
	GAM	8.90	5.50	11.79	26.89	36.67	86.30	23.72	67.51	34.83
DZ-Cr-385 x acc-ck-15	H	70.81	80.14	30.77	75.58	-	66.37	24.54	52.10	75.56
	GAM	9.70	7.44	6.10	20.69	-	51.75	15.92	53.61	98.84

DPE = days-to-panicle emergence; DMT = days-to-maturity; PNG = panicle length (cm); PHT = plant height (cm); PDL = peduncle length (cm) ; TILL = number of productive tillers per plant; PNW = main shoot panicle seed weight of the main shoot (g panicle<sup>-1</sup>); YLD= grain yield (g plant<sup>-1</sup>); BM = biomass yield (g plant<sup>-1</sup>); HI = harvest index; LOD = lodging index (%); RWC= relative water content (%) Dash line (-) indicates absence of heritability

### 6.3.7 Selection of F<sub>2</sub> individuals through single seed descent

In this study high genetic advance values were estimated for important quantitative traits such as grain yield, biomass yield, number of productive tillers per plant, main shoot panicle seed weight and peduncle seed weight, and intermediate values were noticed for plant height and panicle length. These suggest the existence of heritable variation for continuous selection until complete homozygosity is reached. The single seed descent selection (SSD) method is one of the most efficient selection methods to fix additive genes, by advancing the early segregating generation through single seed harvested from each plant (Kang and Priyadarshan, 2007), and

has been recommended in tef for selection of quantitative traits such as yield (Tefera and Peat, 1997; Tefera, 2002).

The number of F<sub>2</sub> seeds selected from each cross combination to advance to the F<sub>3</sub> and later generations through single seed descent is presented on Table 6.15. These segregating populations will be further advanced and selected for drought tolerance incorporating farmer-preferred traits. Because of the tiny seed size of tef, which could be lost easily, F<sub>3</sub> seed of the main panicle were harvested from each plant, targeting one to three seeds for planting to obtain one F<sub>3</sub> plant.

Table 6.15 Newly developed 17 tef populations at the F<sub>2</sub> and number of individual plants selected for subsequent selection through single seed descent

No.	Cross	No of selected F2 plants	No.	Cross	No of selected F2 plants
1	9415 x 222076	180	10	205896 x 222076	150
2	DZ-Cr-387 x acc-ck-15	173	11	Dschanger x 222076	161
3	205896 x 207832	162	12	DZ-Cr-387 x 9415	154
4	207832 x 222076	138	13	Dz-Cr-385 x 222076	175
5	205896 x 9415	166	14	205896 x Dschanger	156
6	DZ-Cr-387 x 207832	110	15	DZ-Cr-387 x 222076	183
7	9415 x Dschanger	144	16	acc-ck-15 x 222076	187
		110	17	DZ-Cr-385 x acc-ck-15	159
8	DZ-Cr-387 x Dschanger				
9	DZ-Cr-387 x 205896	134			

#### 6.4 Conclusions

Under both the moisture-stressed and non-stressed conditions, genotypes, DZ-Cr-387 and 9415 were the best combiners for increased grain yield and yield components. Conversely, genotype 222076 was the best combiner for early maturity only. The selected complementary parents will be useful for tef breeding to improve grain yield and early maturity, which are desired traits for production in moisture-stressed tef growing areas in Ethiopia. The families of the crosses: DZ-Cr-387 x 207832 and DZ-Cr-387 x 222076 were selected for high grain yield and early maturity in both the moisture-stressed and non-stressed environments.

Overall, the present study found high genetic variation, heritability and genetic advance values for yield and yield components among 17 newly developed F<sub>2</sub> tef populations tested under moisture-stressed and non-stressed conditions. This would allow for continuous selection

through pedigree, bulk or single seed descent methods to develop improved tef varieties with enhanced grain yields and drought tolerance.

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Appendix 6.1 Monthly and total annual rainfall and mean minimum and maximum temperatures (2005-2015) of the Adi-gidad site

Year	Monthly rainfall (mm)												Total rainfall (mm)	Temperature (°C)	
	Jan	Feb	Mar	Apr	May	Jun	Jul	Aug	Sep	Oct	Nov	Dec		Mean max	Mean min
2005	0.0	0.0	54.6	12.0	52.1	123.2	264.1	194.7	204.7	5.6	3.4	0.0	914.4	28.0	15.0
2006	0.0	0.0	34.0	13.6	71.7	95.3	293.9	414.4	141.1	98.0	1.4	5.4	1168.8	27.4	14.2
2007	0.0	0.0	8.6	9.7	67.6	169.3	337.7	234.9	177.5	18.1	0.7	0.0	1024.1	27.4	14.4
2008	24.4	4.4	0.0	53.4	45.0	125.1	235.9	248.9	131.4	3.4	4.0	0.4	876.3	27.6	14.5
2009	0.0	0.0	24.8	8.1	0.0	129.4	286.8	332.8	64.7	41.6	1.8	0.0	890.0	29.1	15.7
2010	19.0	0.0	60.5	36.4	53.1	148.1	246.2	289.1	265.8	30.8	0.0	0.0	1149.0	27.7	14.9
2011	5.0	0.0	10.0	14.1	80.9	169.1	156.7	287.5	171.4	5.7	5.1	0.0	905.5	27.8	14.5
2012	0.0	0.0	0.0	5.8	14.6	211.4	405.4	428.7	232.4	0.2	12.6	0.0	1311.1	28.2	15.0
2013	0.0	0.0	2.7	8.0	58.4	214.5	138.7	333.2	60.7	60.7	22.1	0.0	899.0	28.3	14.4
2014	0.0	0.0	32.1	56.3	76.7	125.6	295.6	275.8	310.2	70.6	0.0	0.0	1242.9	28.0	13.9
2015	2.4	3.8	10.2	13.7	120.7	87.5	225.7	246.5	196.3	54.6	17.4	9.7	988.5	28.8	14.8

Appendix 6.2 Monthly and total annual rainfall and mean minimum and maximum temperatures (2005-2015) of Hastebo and Dura sites

Year	Monthly rainfall (mm)												Total rainfall (mm)	Temperature (°C)	
	Jan	Feb	Mar	Apr	May	Jun	Jul	Aug	Sep	Oct	Nov	Dec		Mean max	Mean min
2005	0.0	0.0	129.7	86.0	5.1	85.4	176.6	226.0	67.2	0.0	0.0	0.0	776.0	28.5	11.2
2006	0.0	0.0	0.0	31.1	61.0	86.5	230.7	240.6	123.9	9.5	0.0	30.5	813.8	28.0	10.8
2007	0.0	0.0	7.0	10.9	35.5	112.6	428.1	272.8	154.3	0.0	6.1	0.0	1027.3	28.0	10.6
2008	38.5	0.0	0.0	85.2	41.3	102.3	161.8	174.7	49.9	1.5	6.8	0.0	662.0	26.6	10.2
2009	0.0	0.0	0.9	6.0	8.6	35.6	231.9	288.6	1.8	0.0	2.9	0.0	576.3	27.5	11.3
2010	1.2	0.0	54.2	36.3	17.3	109.4	209.4	223.2	137.5	20.3	0.0	0.0	808.8	27.4	9.9
2011	1.2	0.0	1.9	7.9	58.7	13.5	204.2	151.1	93.7	3.4	11.8	0.0	547.4	26.4	10.0
2012	0.0	0.0	2.7	14.6	4.3	93.9	157.0	289.5	119.5	0.0	3.2	0.0	684.7	27.8	11.2
2013	0.0	0.0	0.0	34.4	20.9	84.8	166.0	248.2	7.5	10.2	0.0	0.0	572.0	28.9	12.7
2014	0.0	0.0	59.4	68.0	55.0	123.2	185.0	217.5	194.0	18.0	13.0	4.0	937.1	27.7	11.9
2015	0.0	0.0	16.7	7.0	31.9	50.0	191.7	233.4	44.0	6.8	7.5	26.8	615.8	28.6	12.9

## Overview of the Research Findings

### Introduction and objectives of the study

Tef is the major staple food in Ethiopia and is becoming globally popular for its gluten-free property. Tef is the leading cereal crop in Ethiopia in terms of area coverage. However, its productivity is the least, contributing to food insecurity in the country. Tef has an array of production problems, the major being lodging, moisture stress, shattering and poor pre- and post-harvest management practices. Typically, the crop is cultivated under rain-fed condition and its yield response is vulnerable to erratic and variable rainfall conditions frequently witnessed in northern Ethiopia. Developing farmer-preferred, moisture stress tolerant tef varieties with better grain yield performance and quality traits such as white seed colour and good *injera* making quality are the main goals of tef improvement. The success of developing improved varieties depends on proper evaluation and selection procedures to exploit the available genetic variability for targeted traits. This chapter highlights the study objectives with subsequent summary of major findings of each objective. Finally, the implications of the major findings for breeding moisture stress tolerant and better yielding tef varieties are presented.

The objectives of the study were

1. To assess tef production constraints, management practices, input use, farmers trait preferences and breeding priorities in northern Ethiopia, Tigray Region.
2. To determine the genetic diversity of tef genotypes for drought tolerance using phenotypic and molecular markers.
3. To estimate the level of genetic variation, inheritance and trait association of tef genotypes.
4. To determine gene action and inheritance of drought tolerance in tef and identify better performing populations for moisture-stressed and non-stressed environments and advance these through the single seed descent method.

### Research findings in brief

**Assessment of production status, constraints, management practices and farmer-preferred traits of tef [*Eragrostis tef* (Zucc.) Trotter] under drought-prone environments: Implications for future research directions**

Participatory rural appraisal was conducted to determine the current production and productivity status, input use and management practices and farmers' trait and variety

preferences of tef in moisture stressed growing areas. The study sampled 240 household farmers from four districts within three administrative Zones of the Tigray Region, northern Ethiopia. Primary data was collected through individual structured interview and secondary data was collected from the Bureau of Agriculture and Rural Development (BoARD). The main outcomes were as follows:

- The farmers allocated more than half of their land to tef production for its high value (relative to other cereal crops) for food, animal feed and cash income.
- High grain yield, increased panicle length and straw yield were the most important farmer-preferred traits. At Alamata district which is a severely moisture stressed area, early maturity was the third preferred trait while straw yield ranked fourth.
- Tef variety, Quncho (DZ-Cr-387), was most adopted by farmers in Ahferom, Laelay-Maichew and Medebay-Zana Districts. The variety, DZ-Cr-37, was popular in Alamata District for its better moisture stress tolerance.
- Transplanting maximized the yield potential of tef, but a cost-benefit analysis showed that direct row sowing was more profitable. Due to the difficulty to apply, associated high costs, and high human labour requirement, farmers are reluctant to apply these technologies for large-scale production.
- Moisture stress, weeds, shoot fly, lodging and seed shattering were rated as severe tef production constraints in the study areas. About 37-60% tef grain yield reductions were reported by farmers due to moisture stress.
- Farmers grow early maturing tef variety to avoid yield loss due late moisture stress, but this was at the expense of high grain yield.
- Farmers in the study areas demand a moisture stress tolerant tef variety with high grain yield.

#### **Performance of tef [*Eragrostis tef* (Zucc.) Trotter] genotypes for yield and yield components under drought-stressed and non-stressed conditions**

One hundred forty four tef genotypes were evaluated under moisture-stressed and non-stressed conditions. Morphological traits such as days to 50% panicle emergence, days to 75% maturity, grain filling period, grain yield, biomass yield, harvest index, lodging index, plant height, panicle length, number of productive tillers per plant and panicle seed weight from the non-stressed and moisture stressed environments were collected and subjected to multivariate analysis.

The main outcomes were as follows:

- Classification of genotypes for moisture stress tolerance were not consistent when they were evaluated in moisture stressed and non-stressed environments, indicating the different response of the genotypes to the different environment.
- Genotypes DZ-Cr-387, DZ-01-787, DZ-01-3186, 9432, 9403, 9415, 205917, 205896, 215678, 213237, Jano, Kaye-Agachew, Purpurea, Kaye-Murri and Dschanger, were selected with superior grain yields, reduced lodging, tall plant height and long panicles in the non-stressed environment.
- Genotypes DZ-Cr-385, DZ-Cr-37, HO-Cr-136, DZ-01-2053, Dabbi, 207832, Zagure and Shawa-Gemerra, were selected as superior genotypes for their early maturity and better yield performances under moisture stressed condition. This study demonstrated the usefulness of evaluation of diverse tef genotypes in moisture-stressed and non-stressed environments for selection of potentially high yielding and moisture stress tolerant genetic resources helpful for strategic breeding of tef. The selected tef genotypes are useful genetic resources when breeding for moisture-stress tolerance

#### **Genetic variation and trait association of tef [*Eragrostis tef* (Zucc.) Trotter] evaluated under optimal and moisture stressed environments**

Data collected from 144 tef genotypes evaluated under four experiments representing optimum moisture and moisture-stressed environments were subjected to genotypic and phenotypic coefficient of variation, heritability, genetic advance, correlation and path analyses. The aim was to determine the genetic variability present among 144 tef genotypes of varied population and to identify important agronomic traits with high heritability and correlations for effective breeding. The main outcomes were as follows:

- There existed substantial genetic variability within tef genotypes screened in optimal and moisture stressed environments useful for tef improvement.
- Heritability values of traits were higher in the optimal environments than they were in the moisture stressed environment due to the relatively high environmental variability.
- The relatively high genetic advance of grain yield and main shoot panicle seed weight under both optimum and moisture stressed conditions would be desirable for genetic gains through selection.
- Direct selection for high biomass, harvest index and late maturity could increase grain yield in optimal environments, while under moisture stressed conditions early

maturity, high biomass and harvest index were important direct selection criteria to use when breeding tef for drought tolerance.

### **Assessment of the genetic relationship of tef [*Eragrostis tef* (Zucc.) Trotter] using SSR markers**

Sixty tef genotypes (including landraces, improved varieties and accessions) were genotyped using 10 selected diagnostic and polymorphic simple sequence repeat (SSR) DNA markers. The main outcomes were as follows:

- The polymorphic information content (PIC) ranged from 0.64 for marker CNLTS11 to 0.94 (CNLTS136A/B) with a mean of 0.84.
- Analysis of molecular variance showed that 63% and 35% of the total variability could be attributed to differences within and among tef genotypes, respectively. The high level of genetic dissimilarity within the tested tef genotypes provides an opportunity for systematic selection and conservation.
- The SSR analysis identified distinct genotypes such as DZ-Cr-385, 222076 and 213237 which are known for their early maturity and good yields under moisture stress. The analysis also demarcated genotypes, DZ-Cr-387, 205896, 205917 and Dschanger, which are consistent to their unique agronomic attributes such as late maturity, high grain yields, relatively tall plant and long panicles under optimum rainfall conditions.
- The SSR analysis enabled in identification of distinct and agronomical complementary tef genotypes. Through designed hybridization, these genotypes could combine well and lead to transgressive segregants for selection of moisture stress tolerant and high yielding tef varieties. .

### **Gene action and early generation selection of yield and related traits among tef populations under moisture-stressed and non-stressed environments**

Eight parents consisting of five accessions (9415, 205896, 222076, acc-15-ck and 207832), a landrace (Dschanger) and two released varieties (DZ-Cr-387 and DZ-Cr-385) were crossed using a half-diallel mating design. Seventeen successful crosses along with the parents were evaluated in the F<sub>2</sub> generation under moisture-stressed and non-stressed conditions with the aim of determining gene action, genetic variation and inheritance of grain yield, and component traits of newly developed tef populations under moisture-stressed and non-stressed conditions for drought tolerance breeding. The main outcomes were as follows:

- Additive gene action was predominantly controlling inheritance of grain yield, biomass yield, harvest index, days to panicle emergence, days to maturity, panicle

length, plant height, main shoot panicle seed weight, number of productive tillers per plant and relative water content under both the moisture-stressed and non-stressed conditions.

- Genotypes, DZ-Cr-387 and 9415 were best combiners for increasing grain yield and major yield components except for days to maturity, on the contrary 222076 was better general combiner in reducing the maturity period.
- Use of diverse tef genotypes such as DZ-Cr-37, 222076 and 207832, has resulted crosses such as, DZ-Cr-387 x 207832 and DZ-Cr-387 x 222076, which combined high grain yield and early maturity under both moisture-stressed and non-stressed conditions.
- The genotypic and phenotypic variability was high for plant height, panicle length, peduncle length, number of productive tillers per plant, main shoot panicle seed weight, biomass yield and grain yield while low for days to panicle emergence and days to maturity for majority of the crosses under both moisture conditions, which shows the high opportunity for selection.

### **Implications of the research findings for breeding tef for moisture stress tolerance**

The following implications for breeding were noted:

- The high grain yield reduction due to moisture stress indicated by farmers implies the importance of developing moisture stress tolerant varieties for sustainable tef production.
- Consideration of identified farmer's variety selection criteria, such as early maturity, high grain and biomass yield and white seed colour, in the tef breeding program would have great implication for high adoption of improved variety in the moisture stressed areas.
- The wide genetic diversity for yield and moisture stress tolerance implies the potential for further improvement through selection and genetic recombination using controlled crosses.
- The high polymorphism of SSR markers used in the current study implies their usefulness in genetic studies and breeding program of tef.
- The distinctive DNA level classification of the tested tef genotypes would enable breeders to undertake designed crosses which could lead to transgressive segregants.

- The predominance of additive gene action in controlling inheritance of grain yield and its components under both the moisture stressed and non-stressed conditions implies that early and later generation selection would be effective.
- The genetic diversity of chosen parents was reflected in the high GCA and SCA effects for high grain yield and early maturity, which where a desired traits in breeding tef varieties with better yield performance for moisture stressed areas.
- The high genetic variability, heritability and genetic advance of grain yield for the segregating tef population implies greater opportunity for selection of high yielding varieties adaptable to both moisture stressed and non-stressed environments.
- Crosses such as DZ-Cr-387 x 222076 and DZ-Cr-387 x 222076 resulted from diverse parents has shown high variability for grain yield and early maturity, which would enable for selection of high yielding tef varieties for moisture stressed areas.