Integrating Sorghum [*Sorghum bicolor* (L.) Moench] Breeding and Biological Control Using *Fusarium oxysporum* Against *Striga hermonthica* in Ethiopia

By

Rebeka Gebretsadik Teshome

MSc. Plant Protection (Pathology) (Alemaya University, Ethiopia)
BSc. Plant Sciences (Alemaya University of Agriculture, Ethiopia)

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

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Thesis Abstract

Sorghum [*Sorghum bicolor* (L.) Moench] is a major food security crop for millions of people in sub-Saharan Africa and the fourth most important crop in Africa. The potential sorghum yields are limited due to a number of abiotic, biotic and socio-economic constraints. Among the biotic stresses is the parasitic weed, *Striga hermonthica*, which inflicts yield losses ranging from 30-100%. Various control options have been recommended to reduce levels of *Striga* damage. However, these techniques need to be integrated for effective control and to boost sorghum productivity. A series of experiments was conducted to integrate host resistance improvement and the use of a biological control agent, *Fusarium oxysporum* f.sp. strigae to control *Striga hermonthica*. These studies were also focused on improving breeders' awareness of the traits that farmers' desire, on the assumption that farmers' variety preference traits are the missing link in technology development and adoption process for *S. hermonthica* management.

The objectives of the study were to: 1) determine farmers' views on sorghum production opportunities; threats; indigenous knowledge and perceptions; breeding priorities; *Striga* infestation; and the coping mechanisms of farmers in the north eastern and north western Ethiopia, 2) evaluate sorghum genotypes for compatibility to *F. oxysporum* inoculation where grown in *Striga* infested soil in controlled environments, 3) determine field responses of sorghum genotypes and *F. oxysporum* compatibility for integrated *Striga* management (ISM), 4) determine the variability present among selected sorghum genotypes exhibiting *S. hermonthica* resistance, and compatibility with the biological control agent using phenotypic and simple sequence repeat (SSR) markers, 5) identify *F. oxysporum* compatible sorghum parents and hybrids with high combining ability for grain yield, yield components, and *Striga* resistance for ISM, and 6) undertake farmers' participatory assessment, and identify their preferred traits for sorghum genotypes under ISM, simultaneously with the breeders' evaluation.

A participatory rural appraisal (PRA) research was conducted involving 315 farmers in nine districts of three administrative zones within two provinces in Ethiopia. Sorghum landraces were preferred by >85% of participants rather than previously improved released varieties. The participating farmers listed and prioritized their sorghum production constraints. In the North Shewa and North Wello zones drought was the most important constraint, followed by *Striga*. In the Metekel zone *Striga* was the number one constraint followed by a lack of genotypes with high grain quality.

Controlled environment experiments were conducted involving greenhouse and laboratory tests in order to evaluate 50 sorghum genotypes for their compatibility with *F. oxysporum* and for possible deployment of the bio-control agent to control *Striga*. *Striga* population was reduced by 92% through the application of *F. oxysporum,* resulting in yield increment of 144%. Twelve sorghum genotypes were identified as promising parents for breeding and to control *Striga* through integration of host resistance and *F. oxysporum* seed treatment.

During field and sick plot plot evaluations differential responses to *F. oxysporum* application among the sorghum genotypes were observed for various attributes including *Striga* plant height. Most traits showed highly significant ($p<0.001$) genotype X site interactions. Similarly, the main effects of *F.oxysporum* application were highly significant ($p<0.001$) across sites for most of the traits. The genotype and genotype X environment biplot identified 13 genotypes that consistently performed well following *Fusarium* application.

The variability present among 14 selected sorghum genotypes exhibiting *S. hermonthica* resistance, and compatibility with a biological control agent, *Fusarium oxysporum,* were determined using phenotypic and 20 polymorphic simple sequence repeat (SSR) markers. Highly significant ($p<0.001$) differences were detected among genotypes for phenotypic traits. Principal component analysis showed three components that accounted for 73.99% of the total variability exhibited among genotypes. Cluster analysis allocated the genotypes into two major groups, one with a further two subgroups based on morphological traits, showing clear demarcations between the genotypes. The SSR markers revealed high levels of polymorphisms among genotypes, with the mean number of alleles per locus being 6.95 and the mean polymorphic information content being 0.80. The observed genetic diversity was relatively wide, with the allele sizes ranging from 203.6-334 bp. The SSR markers allocated genotypes into two distinct clusters close to the phenotypic markers.

Forty sorghum hybrids were developed through a line by tester mating design involving 10 lines selected for their compatibility with *F. oxysporum* and high agronomic performances and four *Striga* resistant tester parents. The F_1s and their parents were field evaluated with complementary *in-vitro* tests. Field evaluations were conducted at two locations: Kobo and Shewa Robit in Ethiopia, which are well known for their severe *Striga* infestation. Significant ($p<0.05$) general combining ability (GCA) effects were observed among testers and lines at both sites for days to 50% flowering and maturity, plant height, biomass, number of *Striga* plants and *Striga* plant height. Furthermore, significant ($p<0.05$) specific combining ability (SCA) effects were detected for days to 50% flowering, biomass, grain yield and number of *Striga* plants. From the complementary *in-vitro* experiment, highly significant variation ($p<0.01$) was exhibited due to line x tester interaction for maximum *Striga* germination distance. The study identified paternal parents with high GCA effects including SRN-39 and Birhan and maternals 235761, 2384443, IC9830, 235466, 237289,235763, and 235929 to be useful for breeding for ISM in sorghum. At Kobo, cross 235763 x N-13 and Shewa Robit IC9830 x SRN-39 had significantly negative SCA effects for the numbers of *Striga* plants. Progenies of these crosses will be selected in the *Striga* resistance breeding program.

In the participatory sorghum genotypes assessment, farmers were invited to assess and select the genotypes based on their preferences at maturity and harvesting. The standard agronomic traits and *Striga* parameters relevant for breeding were collected by the breeders. Earliness, *Striga* resistance, high yield and high grain quality and threshability were the most important farmers'-preferred traits for sorghum genotypes. Comparative analyses between farmers' and breeders' evaluations revealed highly significant correlations ($p<0.01$) except between *Striga* resistance and *Striga* damage and pest resistance and insect damage. Repeatability of scoring genotypes among farmers was consistent (>0.80) for all traits except *Striga* and pest resistance. The prioritized traits through farmers' participation are important for further breeding program. Overall, the study established farmers' preferred traits, the effectiveness of ISM to boost sorghum productivity, and identified useful parents and crosses for effective sorghum breeding to control *Striga* in Ethiopia.

Declaration

I, Rebeka Gebretsadik Teshome, 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:

    a. Their words have been re-written but the general information attributed to them has been referenced

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

5. This thesis does not contain text, graphics or tables copied and pasted from the Internet, unless specifically acknowledged, and the source being detailed in the thesis and in the references sections.

Signed

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Rebeka Gebretsadik Teshome

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

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Prof. Shimelis Hussein (Supervisor)

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Prof. Mark Laing (Co-Supervisor)

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Prof. Pangirayi Tongoona (Co-Supervisor)

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Dedication

This thesis work is humbly dedicated to my husband Sisay Lemma and our son Hibir Sisay, for your memorable sacrifice.

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Abbrevations

APPC	Ambo Plant Protection Centre/Ethiopia
BM	Biomass
CFU	Colony Forming Units
CSA	Central Statistics Agency
DAP	Diammonium Phosphate
DEM	Days to Emergence
DFL	Days to Flowering
DM	Days to Maturity
FAO	Food and Agricultural Organisation
GCA	General Combining Ability
GGE	Genotype and Genotype by Environment interaction
GY	Grain Yield
IBC	Institute of Biodiversity Centre/Ethiopia
ICRISAT	International Crops research Institute for the Semi-Arid Tropics
ISM	Integrated *Striga* management
MGD	Maximum Germination Distance
PCA	Principal Component Analysis
PDA	Potato Dextrose Agar
PHT	Plant Height
PL	Panicle Length
PRA	Participatory Rural Appraisal
SARC	Sirinka Agricultural Research Centre/Ethiopia
SCA	Specific Combining Ability
SNA	Special Nutrient Agar
SSR	Simple Sequence Repeat

Introduction to the thesis

Importance of sorghum

Globally, sorghum [*Sorghum bicolor* (L.) Moench] is the fifth most important staple food crop after wheat, rice, maize and barley (FAO, 2012). The crop is produced for its grain which is used for food, and stalks for fodder and building materials in developing countries. In developed countries, sorghum is used primarily as animal feed and in the sugar, syrup, and molasses industry (Dahlbert et al., 2004). The crop is widely grown in the semi-arid tropics because of its unique adaptation to harsh and drought-prone environments. Total sorghum production from all sorghum producing countries was 55.6 million tonnes in 2010. The world average annual yield for sorghum was 1.37 tonnes per hectare in 2010. FAO reported the United States of America as the top sorghum producer with a harvest of 9.7 million tonnes followed by India, Nigeria, Sudan, and Ethiopia (FAOSTAT, 2011).

The sub-Saharan Africa produces about 18 million tonnes of sorghum annually making it the second important cereal crop after maize (*Zea mays* L.) which is produced on about 27 million hectares (Haussmann et al., 2000a; Mutisya, 2004). Nigeria is the leading sorghum producer in Africa followed by Sudan, Ethiopia, and Burkina Faso. However, in terms of productivity, Egypt achieves the highest yields followed by Algeria, South Africa, Uganda, and Ethiopia (FAOSTAT, 2006).

Sorghum is the fourth primary staple food crop in Ethiopia after tef, maize, and wheat, both in area coverage, and production (CSA, 2012). In the country cereals comprise 78.23% (8.8 million ha) of the field crops of which sorghum accounts for 14.41%. In Ethiopia sorghum is grown in almost all regions occupying an estimated total land area of 1.6 million ha (CSA, 2012). The major sorghum production regions of the country are Oromia at 38.5%, Amhara (32.9%), Tigray (14.1%), and Southern Nations and Nationalities People (S.N.N.P.) region (7.6%).

The productivity of sorghum in Ethiopia is low when compared to other African countries (FAOSTAT, 2006). Ethiopia's average productivity of sorghum is <1.35 tons ha^{-1}, ranking it fifth in Africa. The global average yield of sorghum stands at 1.35 ton ha^{-1} (Geremew et al., 2004). The low national sorghum yield signifies the necessity of sorghum improvement to enhance productivity and achieve food security.

Sorghum production constraints

The potential productivity of sorghum is reduced due to a number of abiotic and biotic stresses. Paramounts among the abiotic factors are low soil fertility (nutrient deficiency) and drought. Important biotic constraints include the parasitic weed; *Striga* (*Striga* species), foliar and panicle diseases, stem borers, and shoot fly (Wortmann et al., 2006). Among the major sorghum diseases anthracnose, smuts and rusts account for substantial yield losses in the country.Sorghum production constraints vary from region to region within Ethiopia. However, drought and *Striga* are the most important problems across regions. Consequently, the present research focuses on integrated *Striga* management to enhance sorghum productivity in Ethiopia.

Striga hermonthica

Striga, is a parasitic weed belonging to the Orobanchaceae (formerly: Scrophulariaceae) family. It infests and significantly reduces yields of cereal crops including rice (*Oryza glaberrima* Steudel and *O. sativa* L.), pearl millet (*Pennisetum glaucum* [L.] R. Br. or *P. americanum* [L.] K. Schum), maize (*Zea mays* L.), and sorghum (*Sorghum bicolor* [L.] Moench) (Rich et al., 2004; Khan et al., 2005; Ejeta, 2007). The Orobanchaceae family includes 50 species, of these 11 are recognized as crop pests (Mohamed et al., 2007). *Striga* threatens the livelihoods of millions of smallholder farmers throughout the semi-arid Africa and parts of Asia. Continuous cropping and the extension of cultivation to marginal soils due to population pressure have resulted in the spread and intensification of the *Striga* problem (Parker, 1991). It has been estimated that $100x10^6$ ha of the African savannah zones are infested with *Striga* (Ejeta, 2007).

The giant witch weed, *Striga hermonthica,* is the most important species that cause severe yield reduction in cereals. Dogget (1975) reported a 70% yield reduction in sorghum due to this weed. Parker (1991), indicated yield losses ranging from 70% to total crop failure, depending on the severity of the infestation. However, accurate estimates of yield losses are compromised by the non-uniformity of natural infestations and by the difficulty of creating parasite free areas for comparison (Sauerborn, 1991).

In Ethiopia, *Striga* is widely found in the lowland areas where sorghum is the dominant crop (Figure 1). Based on its infestation level sorghum yield loss due to *Striga* damage varies from place to place. On average sorghum yield losses of 65% were estimated in moderate to heavy infestations (Tesso et al., 2007), however, the continental average is 40% (Lagoke et al., 1991). In the country *Striga* is also observed on maize, pearl millet, rice, tef and cowpea fields. Diagnostic surveys conducted in the past five years by Debrebirhan, Sirinka and Pawe agricultural research centers indicated that *Striga* is rapidly expanding in many parts of the country (unpublished survey reports).

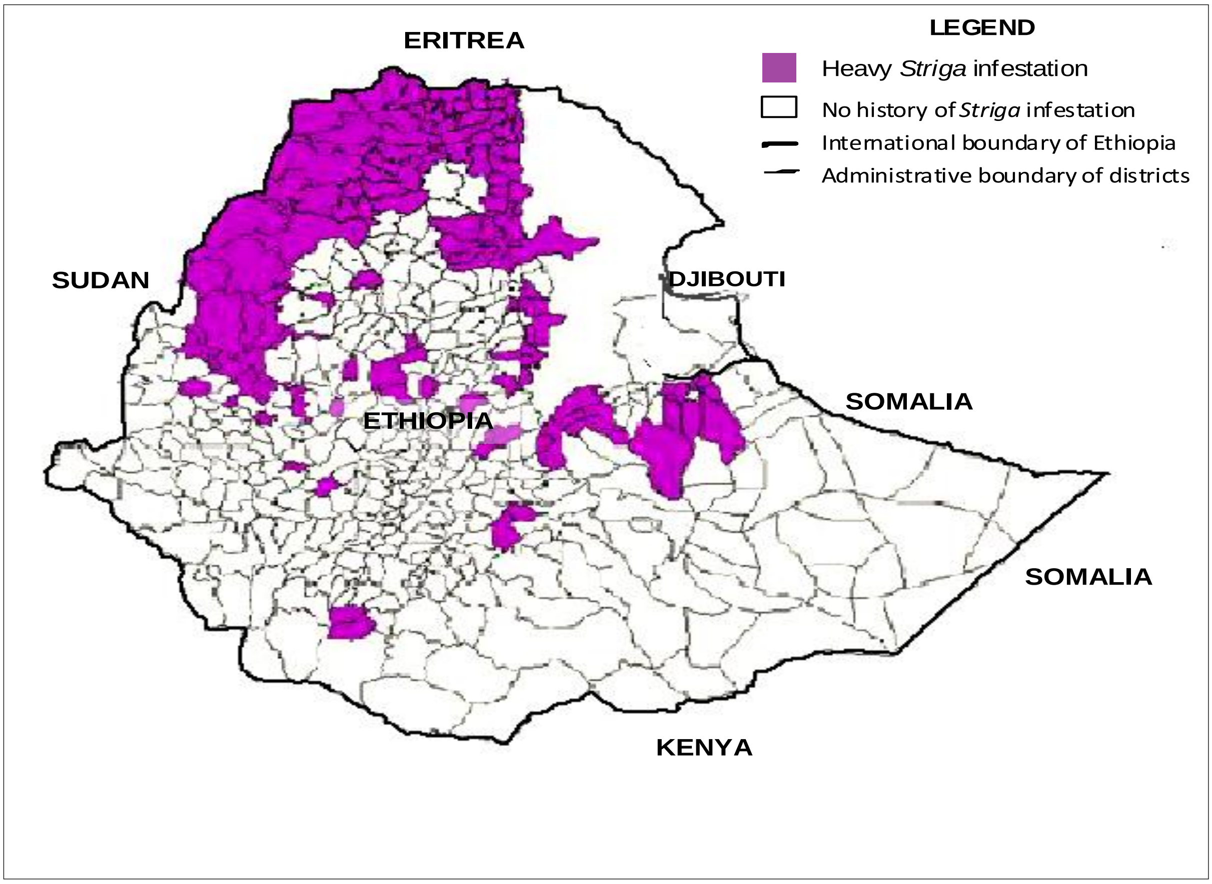

Figure 1. Historically known areas with heavy *Striga* infestation in Ethiopia (Tesso et al., 2007)

Control options

Striga control is more difficult and complicated than the control of other weeds. This is due mainly to its biology. The weed produces large quantities of very small seeds which can remain viable in the soil for up to 20 years (Gurney et al., 2003). A single plant can produce up to 50,000 seeds, which mature at different times. Because of their after ripening requirement, all seeds are not preconditioned for germination at the same time (Khan et al., 2005).

There is a need for an effective control method against *Striga* parasitism. Most control strategies work by disrupting the *Striga* life cycle. This requires understanding the physiological and metabolic interactions between host and parasite (Ejeta, 2007). *Striga* is primarily a problem of small scale subsistence farmers with limited options for external inputs. Thus control options must be low cost and easy to apply.

Over the past years' different control options have been recommended against *Striga hermonthica*. Despite the high potential of some of these solutions, no single option on its own has proven to be effective and durable for sorghum production for resource poor farmers. The best options for successful *Striga* control lies in an integrated *Striga* management (ISM) approach (Joel, 2000; Schulz et al., 2003; Hearne, 2009).

Rationale on integrated *Striga* management

There are various control options of *Striga* in sorghum production. These include the use of resistant cultivars, crop rotation, intercropping with legumes, late planting, use of trap crops, application of organic and inorganic fertilizers, herbicides, and biological control (Hearne, 2009). Host resistance is potentially an acceptable *Striga* control option to resource-poor farmers (Hess and Ejeta, 1992; Haussmann et al., 2000b; Gurney et al., 2003; Rich et al., 2004). However, reliance on host resistance alone is not ideal because so far complete resistance against *Striga* cannot be attained through breeding (Gurney et al., 2002), and usually the newly developed varieties may not fulfill farmers preference traits (Adugna, 2007). The development of partially resistant sorghum varieties, therefore, is an effective approach to reduce the impact of *Striga* as an important component of integrated *Striga* management (ISM).

Since the commencement of sorghum breeding for *Striga* resistance in Ethiopia, research activities were focused on adaptability and performance evaluation of sorghum genotypes from exotic sources (Adugna, 2007). Consequently, the available and farmers' preferred diverse sorghum landraces potential as a source of breeding material is not well exploited. Because farmers' have their own preferred traits for their growing environment, the majority of them become reluctant to grow the genotypes improved so far for *Striga* (Adugna, 2007; Wale and Yallew, 2007; McGuire, 2008; Sinafkish et al., 2010). Therefore, the knowledge about the farmers' variety preferences and variability present among the existing sorghum genotypes can be used as raw material by the breeder to launch an effective breeding program as an important component of ISM.

Biological control using microbes is also becoming a critical component of ISM. Biological control methods are reported to be economical, self-perpetuating and usually free from residual effects. Management of *Striga* through bio-control agents is also much safer and its usage is presumed to be less polluting to the environment than chemical pesticides (Abbasher et al., 1998; Charudattan, 2001; Fen et al., 2007; Rebeka, 2007). Among the microbes, fungi are preferred as bio-herbicides, given that they are usually host specific, highly aggressive, easy to mass-produce and diverse in terms of their genetic constitution (Ciotola et al., 2000). However, farmers rarely adopt *Striga* control methods, either due to limitations associated with the technology itself or because the technology is inaccessible or unaffordable or a lack of information about these control options (Oswald, 2005; Hearne, 2009). Also, these control options when applied individually are not effective and sometimes affected by environmental conditions. The combined use of resistant varieties with the application of *Fusarium oxysporum* as pest granules or as a seed coating was reported to be effective to controlling *Striga* (Marley et al., 2004; Julien et al., 2009). Thus several options need to be integrated in order to achieve sustained and successful *Striga* control.

Because cultivar development depends on the existence of the traits under consideration, diverse genotype collections were required to be evaluated for their compatibility to *Fusarium oxysporum*. Then, promising genotypes based on their response to the bioagent used as a parent to be crossed further with *Striga* resistance genotypes under the ISM breeding program. Research efforts in order to introgress *Striga* resistance genes into locally adapted sorghum genotypes at target environments has been limited due to the difficult nature of *Striga* physiology, reproduction and its complex interaction with the host plant (Ejeta et al., 1992; Haussmann et al., 2000b). However, information related to the general and specific combining abilities of the selected parental lines particularly related to traits associated to *Striga* resistance and yield is vital for this study. This can be obtained through combining ability studies after the systematic crossing of selected parents using different mating designs and further hybrid evaluation including the parents. Variation due to general combining ability is attributed to the presence of additive genes and that due to specific combining ability is attributed to non-additive gene action (Kenga et al., 2004).

Research objectives

The specific objectives of this study were to:

1. Determine farmers' sorghum production opportunities, threats, indigenous knowledge and perceptions, emphasising breeding priorities and *Striga* infestation, and the coping mechanisms of farmers in the north eastern and north western Ethiopia.
2. Evaluate sorghum genotypes for compatibility to *F. oxysporum* inoculation under *Striga* infested soil in controlled environments.
3. Determine field responses of sorghum genotypes *F. oxysporum c*ompatibility for ISM.
4. Determine the variability present among selected sorghum genotypes exhibiting *Striga hermonthica* resistance, and compatibility with the biological control agent, *F. oxysporum*, using phenotypic and simple sequence repeat (SSR) markers.
5. Identify *F.oxysporum* compatible parents and hybrids with high combining ability for grain yield, yield components, and *Striga* resistance for integrated *Striga* management (ISM).

6. Undertake farmers' participatory assessment and identify preferred traits of sorghum genotypes under integrated *Striga* management simultaneously with the breeders' evaluation.

Research hypotheses

This study was carried out to test the following hypotheses:

1. In sorghum growing areas of Ethiopia, smallholder farmers have different social, cultural, and economic factors associated with sorghum production opportunities and threats which can contribute to sorghum breeding.
2. Sorghum productivity can be improved through selection of *Striga* resistant genotypes that are compatibile with the bio-control agent, *Fusarium oxysporum.*
3. Phenotypic and SSR markers could be used in identifying variability in *Striga* resistance among sorghum genotypes.
4. Crosses between *Fusarium* compatible sorghum genotypes and *Striga* resistance sources could be exploited through identifying parents having high combining abilities for yield and *Striga* resistance.
5. Farmers' preferred traits of sorghum genotypes for effective ISM approach can be identified through farmers' participation starting from early breeding stage together with breeders' evaluation.

Outline of this thesis

This thesis consists of seven distinct chapters in accordance with a number of activities related to the above objectives. Chapters 2-7 are written 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 referencing system used in the chapters of this thesis is based on the Harvard system of referencing (De Montfort University), and follows the specific style used in

“Southern Forests: a Journal of Forest Science”. The exception to this is Chapters 2, which is in press to be published in “Agricultural Systems” and Chapter 3, which is published in the journal of “Crop Science” In this case, Chapter 2 and Chapter 3 have followed the referencing and formatting style used by “Agricultural systems” and “Crop Science”, respectively.

Chapter	Title
-	Introduction to thesis
1	A review of the literature
2	A diagnostic appraisal of the sorghum farming system and breeding priorities in *Striga* infested agro-ecologies of Ethiopia
3	Evaluation of sorghum genotypes compatibility with *Fusarium oxysporum* under *Striga* infestation
4	Field evaluation of sorghum genotypes against *Striga* through combined use of resistance and *Fusarium oxysporum* compatibility
5	Assessment of sorghum genotypes with *Striga hermonthica* resistance and *Fusarium oxysporum* compatibility using phenotypic and SSR markers
6	Combining ability for grain yield and *Striga* resistance in sorghum [*Sorghum bicolor* (L.) Moench]
7	Participatory assessment of farmers’ preferences of sorghum genotypes under integrated *Striga* management
8	An overview of the research findings

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Chapter 1
A review of the literature

1.1 Introduction

Sorghum [*Sorghum bicolor* (L.) Moench] is a major cereal in the semi-arid regions of the world where it is an important food and feed crop. However, its production and productivity is limited by a number of abiotic and biotic stresses. The parasitic weed, *Striga,* remains the major biotic stress factor inflicting significant yield loss in sorghum in the sub-Saharan Africa. Although various control options have been recommended to reduce *Striga* damage on sorghum and related cereals, these techniques need to be integrated for best results to enhance productivity. This review covers the current state of knowledge on sorghum production, the importance of *Striga* infestation and its control strategies. Further, the review provides background information on integrated *Striga* management, and highlights the challenges and opportunities for *Striga* control. Integrated use of *Striga* resistant sorghum cultivars with a *Striga*-specific *Fusarium oxysporum* seed treatment could provide a sustainable management approach to *Striga* control, and to improve sorghum productivity in smallholder farming systems.

1.2 Sorghum domestication and production

The geographic place of origin and initial domestication of sorghum is in Africa. Ethiopia is believed to be the centre of origin and domestication of sorghum [*Sorghum bicolour* (L.) Moench] (Vavilov, 1951; Doggett, 1988). This is owing to the existence of the largest diversity of sorghum in northeast Africa. Conversely, (Stemler et al., 1975) argued that none of the bio-geographical, morphological, historical, or evolutionary evidence supported the claim that sorghum was domesticated or originated in Ethiopia. However, in many crop species the wild ancestors are found at the centre of origins and they represent an important source of variation and adaptive traits for breeding. Unravelling the origin and distribution of sorghum diversity is potentially important to its future utilization and conservation.

Sorghums occur both as weedy species in Africa’s savannah ecosystems (Wood and Lenne’, 2001) and as a cultivated cereal. Cultivated sorghum species is classified as *Sorghum bicolour* L. Moench and comprises five major races (*Guinea,* *Durra, Caudatum, Kafir, and Bicolor*) based on spikelet and panicle characteristics (Harlan and Dewet, 1972). Four major races of the cultivated sorghum are grown in Ethiopia, with the exception being the *Kafir* race.

Following domestication, genetic admixture or introgression or hybridization events probably occurred between wild and cultivated species. Subsequently, thousands of years of selection in response to diverse physical environments and human needs, genetic drift, and natural inter-crossing among the different sorghum races have contributed to sorghum diversity. These have allowed sorghum to be grown in a variety of environments (Vavilov, 1951; Stemler et al., 1975; Doggett, 1988). At present, sorghum is an important cereal crop with remarkable genetic diversity with more than 22,000 accessions kept in the world sorghum collection in India (Kimber, 2000).

Globally, sorghum is the fifth most important staple food crop after wheat, rice, maize and barley (FAO, 2012). It supports about 500 million people serving as a source of food, feed, fibre, building material and bio-fuel. The world average annual yield for sorghum was 1.37 tonnes per hectare in 2010 (FAOSTAT, 2011). FAO reported the United States of America was the top sorghum producer with a harvest of 9.7 million tonnes followed by India, Nigeria, Sudan, and Ethiopia (FAOSTAT, 2011). In the sub-Saharan Africa sorghum is the second most important food crop after maize, and is predominantly grown by small-scale and subsistence farmers. More importantly, sorghum is the major food and cash crop for the most food insecure farmers in the semi-arid areas which experience low and unreliable rainfall patterns, and which are not suitable for most other crops, including maize (Mutisya, 2004).

In Ethiopia, sorghum is one of the primary staple food crops. It ranks fourth among the most important crops grown next to tef, maize, and wheat, both in area coverage, and production. Of the total area covered by field crops 78.23% (8.8 million ha) is under cereals, of which sorghum accounts for 14.41%. It is grown in almost all regions, with a total area of 1.6 million ha of land (CSA, 2009). The major sorghum production regions of the country include: Oromia at 38.5%, Amhara (32.9%), Tigray (14.1%), and Southern Nation and Nationality People (S.N.N.P) region (7.6%). At the national level, sorghum provides about 16.38% of the total 84.69% area allocated to cereal crop production (CSA, 2009).

1.3 Constraints to sorghum production

The livelihoods of millions of subsistence farmers depend on sorghum production. However, its productivity in Ethiopia is low at about 1.35 ton ha^{-1} (Geremew et al., 2004). This is attributed to a number of abiotic and biotic stresses. Yield reducing factors include low soil fertility (nutrient deficiency), drought, *Striga*, stem borers, and shoot fly (Wortmann et al., 2006). Although these constraints cause a significant loss of grain, the level of losses varies from region to region. In Ethiopia, *Striga*, is a major production constraint in most sorghum producing areas. The weed limits the productivity of the crop by allelopathy, competition for nutrients and limiting the expression of the full genetic potential of sorghum plants.

In Ethiopia, drought and *Striga* are reported to be important sorghum production constraints in the north and north eastern parts of the country whereas quelea birds are the major constraint in the Rift Valley and Southwest lowlands (Wortmann et al., 2006). Consequently, the current research was conducted in the north eastern, north western, and Eastern parts of Ethiopia, which represents Ethiopia's sorghum growing belts. Although both drought and *Striga* are equally important problems in sorghum, this research and review focused on *Striga.*

The domestication and spread of sorghum and *Striga* species are reported to have occurred together (Rao and Musselman, 1987). Consequently, *Striga* most probably evolved in association with sorghum. It is believed that *S. hermonthica* originated in the Sudano-Ethiopian region, where sorghum originated and spread into the rest of Africa and Arabia (Tadesse and Yilma, 1991). The co-evolution of cultivated or wild sorghum species with *Striga* could be a source of genetic variation for resistance breeding against *Striga*.

1.4 *Striga*: a parasitic weed of sorghum

The genus *Striga* belongs to the family *Orobanchaceae* (formerly: *Scrophulariaceae*). This genus parasitizes cereal crops such as rice (*Oryza glaberrima* Steudel and *O. sativa* L.), pearl millet (*Pennisetum glaucum* L. R. Br. or

P. americanum [L.] K. Schum), maize (*Zea mays* L.), and sorghum (*Sorghum bicolor* [L.] Moench) (Parker, 1991; Johnson et al., 1997). It also parasitizes many wild grass species in Africa. There are more than 50 species of *Striga*, with several species affecting the production of cereals and legumes in sub-Saharan Africa and Asia (Parker and Riches, 1993; Kiruki et al., 2006).

The *Striga* species are among the most specialized of all root-parasitic plant parasites (Parker and Riches, 1993). *Striga* combines the life styles of both a holo-parasite at the seedling stage and a hemiparasite as a green, chlorophyll-containing emergent plant (Mohamed et al., 2001). Of the parasitic species of *Striga*, *S. hermonthica, S. asiatica, S. aspera, S. forbesii* and *S. gesnerioides* are of particular economic importance as crop parasites in Africa (Mohamed et al., 2001). These species attack all the important tropical cereals except *S. gesnerioides* which parasitizes only dicotyledons.

S. hermonthica is perhaps the most destructive as compared to the other *Striga* species to cereal production. It attacks sorghum, maize, millet and rice (Abbasher et al., 1998). The plants are hairy with robust, quadrangular, fibrous stems. The leaves are linear-lanceolate to lanceolate, with a length of 2.5-7.5 cm and up to 2 cm wide. Flowers are usually bright pink, but many variants occur, and occasionally completely white flowers have also been observed (Mohamed et al., 2001).

1.5 Host-parasite association

Striga is an obligate hemi-parasite needing a host plant to fulfill its life-cycle. However, due to its chlorophyllous leaves, it undergoes photosynthesis and as such it does not entirely depend on its host for its metabolite requirements (Kuijt, 1969). Understanding the life cycle of *Striga* spp. and their interactions with their hosts allow plant breeders to exploit several different mechanisms of resistance against this parasite.

The life cycle of the parasite follows a series of developmental stages, from seed to seed producing plants. Like many other plant species, *Striga* seeds have a period of primary dormancy before the seeds are able to germinate. A second prerequisite for germination is the preconditioning of the seed, which requires about two weeks of humid and warm (25-35°C) conditions (Vallance, 1950; Parker and Riches, 1993).

Preconditioned *Striga* seeds will then need secondary metabolites (xenognosins), which are found in root exudates of their hosts, for germination (Vallance, 1950; Yoder, 2001).

Secondary metabolites serve to direct the radicle of the *Striga* seedling towards the host root (Williams, 1961 a and b). Within four days of germination, the *Striga* radicle needs to find a host root, form a haustorium, and penetrate the host root (Riopel and Timko, 1995). The haustorium is a specialized organ that connects the parasite to the xylem of the host root, enabling the transport of water and nutrients from the host. As a result, it affects the host plant growth and reduces the photosynthetic rate in the host (Ejeta and Butler, 2000).

During *Striga* infestations the symptoms on the crop plant resembles that of a disease. These include stunting of the host plant and the failure of panicle formation as a result of severe infestations. *S. asiatica* can cause the host plant to appear wilted with leaf rolling even though there may be adequate soil moisture. *S. hermonthica* can cause chlorotic lesions or yellowish spots on the host leaves (Ejeta and Butler, 2000).

1.6 *Striga* control methods

1.6.1 Breeding for *Striga* resistance

Plant breeding involves the development of new cultivars with desired balanced genetic constitution, expressing desirable traits consistently in target growing environments. Various research groups have described host plant resistance mechanisms based on their interaction between the parasitic weed and the host plant (Mati et al., 1984; Cherif-Ari et al., 1990; Ramaiah et al., 1990; Grenier et al., 2001; Mohamed et al., 2003; Ejeta, 2007; Amusan et al., 2008). A resistant host genotype may limit the number of *Striga* plants that infect each host plant (Ejeta et al., 1991), or they may reduce the impact of *Striga* on the host plants (Wilson et al., 2000). In contrast, tolerance is the ability of a host variety to support equally severe levels of infestations as other varieties of the same crop, without associated yield loss (Doggett, 1988).

Sorghum shows remarkable genetic diversity with more than 22,000 accessions systematically conserved in the world sorghum collection in India (Kimber, 2000).

Genetic admixtures, introgression of genes or hybridization events between wild and cultivated species probably lead to the development of *Striga* resistant sorghum genotypes (Gurney et al., 2002). Genetic markers including phenotypic, protein (biochemical) or DNA (molecular) markers help to identify characteristics of the specific sorghum genotype. Uses of phenotypic characteristics are a common and traditional approach because they form the most direct measure of the phenotype, readily available, relatively cheap to evaluate and requiring simple equipment (Harlan and DeWet, 1972). However, phenotypic markers are subject to environmental influences in the field that may mask the underlying genetic variation among genotypes. DNA based molecular markers are efficient for the analysis of large numbers of genotypes (Melchinger and Gumber, 1998; Reif et al., 2003). The combined use of phenotypic and molecular markers allows for estimation of genetic diversity more reliably and efficiently. Combined, they provide useful information for breeders to select appropriate parents for efficient breeding approaches, and to conserve novel genetic resources.

Among the breeding approaches, sorghum hybrid development is in its infancy in Ethiopia. Experimental hybrids are being developed using introduced cytoplasmic male sterile (CMS) inbred lines which have the genes for a semi-dwarf habit. These hybrids have been rejected by smallholder farmers because these farmers grow tall landraces, in order to produce highly valued biomass in the long stalks, as well as better grain yield (McGuire, 2008). This reflects the need for plant breeders to identify the crop traits that local farmers demand in new sorghum varieties at the start of a breeding programme, including the breeding of novel hybrids with *Striga* resistance.

Population breeding techniques involving recurrent selection have greater potential to improve selection responses to multiple traits. These techniques allow for the combining of useful alleles in each cycle of selection (Bhola, 1982; Hallauer and Darrah, 1985). Recurrent selection is effective for improving quantitative traits with low and intermediate heritability. This procedure involves systematic testing and selection of desirable progeny derived from a population, followed by recombination of the selected progeny to form an improved population. Menkir and Kling (2007) subjected tropical maize populations to six-cycles of recurrent selection to improve resistance to *S. hermonthica*, successfully improving the levels of *Striga* resistance, based on the accumulation of additive genes for *Striga* resistance..

1.6.1.1 Sources and genetics of *Striga* resistance

Resistance to *Striga* can be expressed at different stages of the *Striga* infection cycle. In some genotypes resistance can be expressed either before parasitic attachment (e.g., low germination stimulation, low haustorial development initiation). In other genotypes resistance can be expressed after attachment (e.g., a hypersensitive response). Rich and Ejeta (2007) reported that interrupting the parasite life cycle in one way or another could lead to a lack of access to the host plant and prevent parasite develpment. This would probably lead to the death of the parasite.

Various sorghum cultivars or breeding lines have been identified and characterized as being resistant to *Striga*. For example, sorghum cultivars that show post attachment resistance are Dobbs, and Framida (SRN 4841) (Mohamed et al., 2003). Among wild relatives, resistance has been expressed by reduced haustoria formation in various genotypes, such as P47121 (Mohamed et al., 2003; Rich et al., 2004). However, more information is needed on the various mechanisms of resistance. This will allow for the transfer of multiple mechanisms for *Striga* resistance into productive and well-adapted local genotypes.

The genetic basis of resistance to *Striga* parasitism has been reported by a number of researchers in this field (Ramaiah et al., 1990; Vogler and Ejeta, 1996; Haussmann et al., 2001). Ramaiah et al. (1990) and Vogler and Ejeta (1996) reported that there is a single recessive gene that confers resistance to *Striga* due to low stimulation of *Striga* seed germination. In contrast, Haussmann et al. (2000a) showed that different sets of genes are responsible for low stimulant production in sorghum cultivars, by analysing for general combining ability (GCA) effects for *Striga* maximum germination distance using the agar-gel assay. Information on combining abilities of genotypes is vital for efficient choice of promising parents and to develop hybrids or segregating generations for selection. This study will identify parents and hybrids which will express favorable gene combinations for yield and *Striga* resistance.

Diallel studies and line by tester analyses with sorghum have clearly indicated the presence of quantitative genetic variation, with a preponderance of additive genetic effects. These investigations were done for stimulation of *S. hermonthica* seed germination in the agar-gel assay; the number of aboveground *Striga* plants becoming established in pots; and the number of emerged *Striga* under field conditions (Haussmann et al., 2000a).

Studies have been conducted to examine the mode of inheritance of traits associated with resistance to *Striga*. Haussmann et al. (2000a) estimated the broad-sense heritability at 0.91 and 0.97 for germination distances in a diallel cross and parental lines, respectively, in an agar-gel assay. In field trials, combined across two locations, each in Mali and Kenya, Omanya et al. (2000) estimated broad-sense heritability in two sorghum recombinant inbred populations to be between 0.70 and 0.81 for three traits, namely, *Striga* counts, *Striga* severity, and the area under *Striga* severity progress curve (ASNPC).

Heterosis for *Striga* resistance is genotype-dependent and may be positive or negative (Ramaiah, 1984; Haussmann et al., 2000b). Sorghum hybrids derived from crosses between a resistant and a susceptible parent were reported to be susceptible (Obilana, 1984), suggesting partial or complete dominance of genes for susceptibility. It was concluded that both parents of a hybrid should be selected for *Striga* resistance.

In general, use of resistant varieties is the most cost effective and environmentally sound control method. The development of resistant sorghum varieties is contributing to an effective approach to reduce the impact of *Striga*. However, complete resistance through breeding alone has not been achieved (Gurney et al., 2002). Therefore, reliance on resistance alone for effective control of *Striga* is not 100% effective and it should be supplemented by other methods, including breeding for varieties that combine resistance with high levels of tolerance (Haussmann et al., 2001).

In Ethiopia for the last 15 years *Striga* resistant varieties have been tested on *Striga* infested experimental plots. Recently a few sorghum varieties were released for commercial production in *Striga*-prone areas. However, these resistant varieties have not been widely adopted by the farmers (Adugna, 2007). Thus, host plant resistance alone has been used with limited success because of the breeders have ignored farmers' trait preferences (Adugna, 2007). Farmers-preferences are critical for successful adoption of improved sorghum genotypes and their production packages such as the ISM. Farmers have complex and diverse sorghum selection criteria including high grain yield and resistance to *Striga*, among others (McGuire, 2008; Sinafkish et al., 2010). Farmers' participation in setting up research priorities and technology evaluation is crucial to researchers in order to design, test and recommend appropriate and new production technologies. This can be achieved through participatory research and evaluation that allows incorporation of farmers' indigenous technical knowledge, identification of farmers' criteria and priorities, and the definition of the research agenda. Participatory research will accelerate the acceptance and diffusion of developed technology by end users.

1.6.1.2 Mechanisms of *Striga* resistance

(i) Biochemical expressions

All host plants susceptible to *Striga* such as, sorghum, maize, pearl millet possess *Striga* growth initiation compounds in their root exudates. These compounds instigate either the germination of *Striga* seed or trigger the growth of the bridge organ, the haustoria, in its parasitic development (Pieterse and Pesch, 1983). Therefore breeding for the low or negligible production of these compounds is one of the approaches to develop *Striga* resistant/tolerant varieties (Ramaiah et al., 1990). One form of resistance in sorghum to *Striga* is via a reduced production of strigol (Ramaiah, 1987; Hess et al., 1992). The *Striga* seed requires preconditioning to be stimulated for germination, leading to respiration and the synthesis of proteins and hormones that would be involved in subsequent steps of parasitism (Joel et al., 2007). Under laboratory conditions, using agar-gel assay, the distance between the host rootlets and germinated *Striga* seed indicate the amount of germination stimulation (Haussmann et al., 2001).

Striga forms a haustorium in order to attach to its hosts. With haustorial formation the apical meristem of the *Striga* radicle switches from cell divisions in a longitudinal direction to radial divisions, resulting in a swelling and proliferation of hair-like projections. Chemical stimulants in the host rhizosphere, called haustorial initiation factors, trigger this development (Riopel and Musselman, 1979). Germinated *Striga* near the roots of sorghum that do not possess this trait normally do not form haustoria and therefore die due to their inability to attach to their potential host (Smith et al., 1990; Yoder, 1999). Although the need for chemical signals exudates by host and non-host plants to elicit *Striga* germination has been known for many years, evidence for the requirement of an additional host signal to encourage production of the haustorium to facilitate attachment to host roots only emerged in the early 1990s (Riopel and Timko, 1995; Jorgensen and Kuijt, 1995).

Striga seeds germinated using artificially synthesized germination stimulants *in vitro* will not develop beyond the formation of a radicle unless these are placed close to a developing root of a host or some non-hosts. Unlike the signals required for the germination of *Striga* seeds, host-produced compounds that are involved in haustorial formation have not been identified. However, it is known that the chemistry of haustorial induction is distinct from germination stimulants. A large number of phenolic compounds have been shown to function as haustorial initiators in *Striga*. A simple quinone, 2, 6-dimethoxy-p-benzoquinone (DMBQ), though not found in root exudates, has been shown to act as a strong haustorial initiating factor (Lynn and Chang, 1990).

(ii) Physiological expressions

Striga can penetrate the epidermis and shows sustained cellular development that allows intrusion to the point of vascular connection. This depends on a host supplied factor to activate post-attachment development of the parasite (Ejeta, 2007).

The hypersensitivity resistance reaction of the *Striga*-resistant plants is characterized by the expression of necrotic lesions at the point of haustorial attachment; these lesions prevent further penetration of the parasite into host roots (Mohamed et al., 2003). The hypersensitivity reaction is further associated with a release of phytoalexins that kill the attached *Striga* (Ejeta, 2007). Previous findings indicated that the wild sorghum genotype, P47121, show a more massive necrotic expression to *Striga* attack than the cultivated sorghum genotype, Framida (Mohamed et al., 2003). This suggests that wild genotypes possessing the hypersensitive reaction could greatly reduce the frequency of *Striga* attachment and parasitic association, relative to susceptible genotypes. Thus, such genotypes might be a source of resistant genes for pyramiding hypersensitivity reaction gene(s) for resistance in sorghum cultivars, despite the negative linkage to wild-type genes for traits such as shattering.

Some sorghum genotypes do not show any reaction to the toxin produced by the parasitic weed during its attachment and then *Striga* growth is terminated immediately after its first penetration (Grenier et al., 2001). On these genotypes, *Striga* plants that survive the early infection stage may not develop beyond the first emergence of the first leaves. Some *Striga* plants appear to develop normally at first but show signs of stunted growth (Matusova et al., 2005). This reaction is similar to that observed when *Striga* unsuccessfully infests non-host plants; thus the use of the term ‘incompatible response’ can be applied to insensitivity to *Striga* toxin (Ejeta, 2007).

(iii) Root morphology

Differences between the roots of the susceptible and the resistant sorghum cultivars may influence *Striga* resistance. Their root character and their rhizosphere also affect the soil microbial population which may be able to suppress *Striga* growth in various ways.

Damage to the root system caused by *Striga* results in a reduction in plant performance. Differences in root morphology may protect the root from *Striga* damage. Among the root characteristics that differ between varieties are the amount of lignin in the roots (Mati et al., 1984), the amount of cellulose deposition layers (Oliver et al., 1991), and encapsulation (Labrousse et al., 2001). These variations in root morphology contribute to host resistance to *Striga* because they affect its ability to penetrate the sorghum root endodermis. Mati et al. (1984) found that the roots of some resistant sorghum cultivars were much tougher to penetrate than in susceptible cultivars, where the *Striga* haustauria could easily penetrate the endoderm.

Amusan et al., (2008) found that resistant maize had fewer *Striga* attachments, delayed parasitic development, and a higher mortality of attached parasites compared with *Striga* development on a susceptible inbred. On the susceptible inbred, *Striga* penetrated the xylem and showed substantial internal haustorial development. Haustorial penetration into the resistant inbred was often stopped at the endodermis. The few parasitic plants that were able to reach the xylem vessels of resistant host plants showed diminished haustorial development, relative to those invading susceptible roots. These results suggest that the resistant inbred expressed both a developmental barrier and an incompatible response against *Striga* parasitism.

1.6.2 Cultural practices

A number of cultural practices have been recommended for *Striga* control such as crop rotation (Oswald and Ransom, 2001); intercropping (Udom et al., 2007); transplanting (Oswald et al., 2001); soil and water management (Van Delft et al., 2000; Reda and Verkleij, 2007); use of fertilizers (Jamil et al., 2011); and hand weeding (Ransom 2000) to reduce the production of further *Striga* seed. These methods should also reduce the density of *Striga* seeds already in the soil seed bank (Reda and Verkleij, 2007). Some of these practices improve soil fertility, which will stimulate the growth of the host but also adversely affects germination, attachment and subsequent development of the juvenile *Striga* plants (Reda and Verkleij, 2007). However, this approach has only limited success for small-scale farmers, largely due to socio-economic and financial constraints that prevent the use of adequate amount of nitrogen fertilization.

1.6.3 Chemical control method

Herbicides tested for the selective control of *Striga* mostly acts through the foliage, although some have soil residual effects. Among the herbicides tested, 2, 4-D has been the most selective and is the cheapest. MCPA (2-methyl-4-chlorophenoxyacetic acid), a compound closely related to 2, 4-D, has also been effective especially when mixed with bromoxynil (Ejeta et al., 1996). Many herbicides are useful in preventing the build-up of *Striga* seeds in the soil but may not prevent damage prior to their emergence (Gworgwor et al., 2002; Kanampiu et al., 2003). Research efforts should therefore be directed towards identifying herbicides that persist in the soil, allowing the germination of *Striga* seeds but killing the seedlings before attachment to the host. However, these herbicide options are mostly unaffordable for resource poor farmers.

Development of transgenic herbicide resistant sorghum is reported as an alternative way for the use of herbicide application to immediate *Striga* management through treatment of crop seeds with herbicide as a low cost solution (Haussmann et al., 2000a; Joel, 2000; Kanampiu et al., 2003). For instance, herbicide seed treatment using imazapyr or 2, 4-D appears to be a promising approach for the control of *Striga* in maize or sorghum (Kanampiu et al., 2001; Dembele et al., 2005). Ndung'u (2009) has also reported coating sorghum seed with herbicide reduced *Striga* infestation, *Striga* flowering and *Striga* seed set, and it is considered as the most effective approach as it does not affect sorghum biomass.

1.6.4 Biological control

Most organisms have natural enemies that balance their populations, avoiding excessive abundance (Templeton, 1982). The basis of biological control is the exploitation of natural enemies of pest species. A prerequisite for the assessment of the prospects for biological control include knowledge of natural enemies and their effect on the population dynamics of the host (Templeton, 1982).

Biological control is particularly attractive in suppressing root parasitic weeds in annual crops because of the intimate physiological relationship with their host plants makes it difficult to apply conventional weed control measures. Currently, biological control using microbes is becoming a critical component of integrated management of *Striga*, given that the bio-control agents are usually host specific, highly aggressive, easy to mass produce and diverse in terms of the number of isolates (Ciotola et al., 1996). Biological control methods are also relatively economical, may be self-perpetuating and are usually free from negative residual effects. Management of *Striga* through bio-control agents is also much safer and less polluting to the environment than the use of chemical pesticides, especially the phenoxy herbicides which are associated with non-target drift problems (Abbasher et al., 1998). The dynamics of both the biotic and abiotic components of the rhizosphere affect *Striga* parasitism, and the efficacy and persistence of bio-control agents (Fen et al., 2007).

Various fungal species are reported to infect *S. hermonthica* plants. Specific isolates of *Fusarium* species are among the most prevalent pathogens and may be highly pathogenic to *S. hermonthica* (Abbasher et al., 1998). Extensive surveys in Burkina Faso, Mali and Niger also demonstrated the occurrence of highly pathogenic and *Striga* specific isolates of *F. oxysporum* (Ciotola et al., 2000). Among this isolate virulent isolate of *F. oxysproum* M12-4A provided more than 90% control of *Striga*, and a three-fold increase in sorghum biomass (Ciotola et al., 1996). The use of a myco-herbicide, i.e., *Fusarium oxysporum* coated seeds and host plant resistance reportedly reduced *Striga* emergence by 95% and increased sorghum yield by 50% (Franke et al., 2006).

Little research has been conducted in Ethiopia to study the use of bio-herbicides to control *Striga*. Preliminary studies revealed the occurrence of four distinct pathogenic races in species *Fusarium oxysporum.* The races are indigenous to Ethiopia and can be found in the major sorghum production areas, associated with *Striga* infestations. The races are virulent and are capable of attacking *Striga* before it penetrates into the roots of sorghum (Rebeka, 2007). Although the potential of bio-herbicidal activity is obvious, research efforts should be strengthened to utilize antagonistic *Fusarium* isolates as a key component of the integrated management of *Striga* in Ethiopia and the many other agricultural systems affected by *Striga* species.

1.6.5 Integrated *Striga* management (ISM) approach

Striga is difficult to control effectively because most of its damage to the host plant occurs underground before the parasitic plant emerges (Rich et al., 2004). The integration of multiple control options is suggested as a better approach to combat *Striga* problem (Lagoke et al., 1994; Kuchinda et al., 2003; Schulz et al., 2003, Aliyu et al., 2004; Temam, 2006; Tesso et al., 2007). Many research findings claim that the integration of multiple control methods provides advantages over the application of each method in isolation. Research findings reported the effectiveness of the combined use of trap-cropping, fertilization and host plant resistance to control *S. hermonthica* (IITA, 2002; Tesso, et al., 2007).

Various *Fusarium* spp. and vesicular arbuscular mycorrhizal (VAM) fungi have been found which can reduce *Striga* infestations significantly on sorghum and maize when used together with resistant host (Abbasher et al., 1998; Ciotola et al., 2000;

Lendzemo et al., 2005; Franke et al., 2006). Integrated *Striga* management approach relies on the use of resistant sorghum genotypes and *Striga* pathogenic *F.oxysporum* application to control *S. hermonthica* emergence and growth lead to effective results (Hearne, 2009; Julien et al., 2009). However, the proposed ISM approaches have not been assimilated easily yet at the farm level, probably because of the complexity and investment of labour that the multiple methods would require.

1.7 Challenges and opportunities

Much research has been conducted on the control of *Striga* species. However, each of these management approaches has their own technical and adoption challenges (Hearne, 2009).The biology of *S. hermonthica* contributes to the difficulty of developing effective control methods. Each *Striga* plant has incredible reproductive capacity, producing numerous small and very light seeds which can be easily dispersed by wind, water, animals and agricultural implements (Ejeta, 2007). These seeds remain viable for five to ten years, and possibly longer (Parker, 1991; Gbehounou and Adango, 2003). This undermines the efficiency of current control technologies. A second major problem is that conventional hand or mechanical weeding do not control the weed because the parasite causes its greatest damage to the crop before emerges above ground (Ejeta, 2007). Availability and unaffordable costs of herbicides and inorganic fertilizers for poor African farmers is another major obstacle to minimizing damage due to *Striga*. Thus, there is a need to look for alternative strategies to efficient and economical control of *Striga* to the poor small scale farmers.

An integrated *Striga* management approach was recommended as comparably cheap enough and technically simple for low-input, small scale farmer of Africa to be able to adopt it (Joel, 2000). The use of resistant crop cultivars is one of the most economically feasible and environmentally friendly means of *Striga* control (Hearne, 2009). Host resistance is believed to reduce *Striga* seed production, through reducing the rate of *Striga* development or *Striga* numbers (Haussmann et al., 2000b). A reduction in aboveground *Striga* numbers, caused by resistance, does however not necessarily lead to a reduction in *Striga* seed production. Therefore supplementing this measure by other easily available control options can contribute for the reduction of *Striga* seed bank and its devastative impact. As one of the alternative approach to be integrated with host resistance, use of bio-control agents such as *F. oxysporum* would minimize *Striga* damage by suppressing its growth and subsequent development (Fen et al., 2007). Such ISM is a user-friendly approach because the bio-agent can be supplied as seed based technology together with the resistant germplasm for better control of *Striga* (Ciotola et al., 2000).

1.8 Conclusion

Sorghum is an important food security crop in the sub-Saharan Africa, growing in harsh environments that are unsuitable to other cereal crops. However, its production and productivity is low because of various yield limiting factors, including *Striga* infestation. Unlike other weed species *Striga* is difficult to control and limited technologies are available that has been assimilated by small scale farmers in Africa to reduce its devastating effect.

Although various control options (cultural, chemical, biological, and use of resistant varieties) exist to reduce *Striga* damage on sorghum, these techniques need to be integrated for best results on the improvement of sorghum productivity. Currently in Ethiopia, *Striga* resistant variety development relies mainly on introduced genetic resources. However, adoption of exotic cultivars by farmers remains a major problem. In Ethiopia, landraces are available that possesses enormous genetic potential which may be useful in sorghum breeding programme. Hence, evaluation of these landraces and knowledge of their resistance level to *Striga* would be highly significant. Though the use of resistant cultivars is one of the most robust and effective approach to control *Striga*, use of mycoherbicide coated resistant cultivar seeds is a more effective and novel approach to diminish *Striga* damage and improve sorghum production. Therefore, profound knowledge would be valuable on the benefit gained by inoculating sorghum with *Fusarium oxysporum,* which is pathogenic to *Striga,* on the reduction of the performance of *Striga.*

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CHAPTER TWO

A diagnostic appraisal of the sorghum farming system and breeding priorities in *Striga* infested agro-ecologies of Ethiopia

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CHAPTER 2

A diagnostic appraisal of the sorghum farming system and breeding priorities in *Striga* infested agro-ecologies of Ethiopia

2.1 Abstract

Sorghum [*Sorghum bicolour* (L.) Moench] is a globally important food security crop, particularly in arid and semi-arid environments. Sorghum productivity is low in subsistence farming systems due to biotic, abiotic and socio-economic constraints. The objective of this study was to determine farmers' sorghum production opportunities, threats, indigenous knowledge and perceptions with a focus on breeding priorities *Striga* infestations and the farmers' coping mechanisms in different agro-ecologies in Ethiopia. A multistage cluster sampling method was employed to interview 315 households selected from nine districts of three administrative zones within two regional states. Participatory rural appraisal tools including a structured questionnaire, pair-wise ranking, focus group discussion, and observations through a transect walk were used to collect data. The results showed that the majority of the participant farmers, (86%) were involved in sorghum production. In all study areas sorghum landraces were preferred by >85% of respondents rather than improved released varieties. Farmers listed and prioritized several sorghum production constraints based on importance and severity. The constraints varied among the study areas due to the diversity of agro-ecologies and cropping systems. Results from the pair-wise ranking showed that farmers' have variable preferences for sorghum varieties. At the north Shewa and north Wello zones drought resistance was the most farmers-preferred trait, followed by *Striga* resistance. In the Metekel zone *Striga* resistance was the number one farmer-preferred trait, followed by grain quality. The prioritised traits will form the basis for farmer-oriented sorghum breeding.

Key words: Agro-ecology, landraces, PRA, sorghum, *Striga*

2.2 Introduction

Sorghum (*Sorghum bicolour* L. Moench, 2n=2x=20) is the fifth important grain crop providing food, fodder and bio-energy feedstock (Poehlman, 1994; FAO, 2012). Sorghum is a critical food security crop for more than 100 million people in Africa. It predominantly grows in low-rainfall, arid to semi-arid environments due to its excellent tolerance to drought, high temperature stresses and low soil fertility. The crop displays relatively high water use efficiency compared to other cereals such as maize and wheat (Doggett, 1988; Blum, 2004). It is believed that cultivated sorghum (*S. bicolour*) was first domesticated in north-eastern Africa. Vavilov (1951) described Ethiopia as a centre of origin of sorghum due to the presence of wide genetic variation. The crop has been adapted to a range of biotic and abiotic stresses, resulting in the evolution of many landraces cultivated in various sub-regions (Rao et al., 2002).

The most important staple cereal crops grown in Ethiopia include tef [*Eragrostis tef* (Zucc.) Trotter.], maize (*Zea mays* L.), sorghum and wheat (*Sorghum bicolour* L. Moench and *Triticum aestivum* L.). On average these crops account for 24.66, 17.56, 16.84, and 14.04% of the total cereal crop cultivated area over the last five years, respectively (CSA, 2011). In Ethiopia, sorghum is cultivated in almost all regions by subsistence farmers for various uses such as food, animal feed and to prepare local beverages. Further the stalk is also used for animal feed and for house and fence construction (McGuire, 2008). Despite its versatility and economic value in the livelihoods of millions of subsistence farmers, sorghum productivity is low, estimated at 1.35 ton ha^{-1} (Geremew et al., 2004). Important yield reducing factors are abiotic (low soil fertility and drought) and biotic (infestation by *Striga*, stem borers and shoot fly) (Wortmann et al., 2006). These factors cause significant grain yield losses but their relative importance varies between regions within the country.

Striga (*Striga hermonthica*), an obligate, root hemi-parasitic, noxious weed, is one of the major biotic constraints in most sorghum growing areas. Gressel et al. (2004) reported that *Striga* species are native constraints and reach their greatest diversity in the tropics where they have co-evolved with cereals, especially sorghum, millets and upland rice. The weed is endemic to sub-Saharan Africa and infests about 26 to 50 million hectares causing annual crop losses ranging from 30 to 90%, and sometimes leading to complete crop loss (Watson, 2007). *Striga* reduces yield and quality through parasitic competition.

Various control options (cultural, chemical, biological, and use of resistant varieties) have been developed to reduce *Striga* damage on sorghum. These approaches need to be integrated to improve sorghum productivity and quality. Currently in Ethiopia, development of *Striga* resistant varieties relies mainly on introduced genetic resources. However, adoption of exotic cultivars by farmers' has been negligible mainly because they do not possess farmers' preferred traits (Adugna, 2007). Different reports are available on the low adoption rate of improved sorghum varieties by resource poor farmers in Ethiopia (McGuire, 2008; Sinafkish et al., 2010). For instance, McGuire (2008) indicated that despite 25 years of sorghum breeding in Ethiopia most of the released varieties had been poorly adopted by the small-scale farmers. Reasons for slow adoption rate include lack of effective seed production and delivery mechanism and the introduced germplasm do not fulfil farmers' preferred traits. In Ethiopia, farmers' variety preferences is not only grain yield but also straw yield for livestock feed and other related social values. Wale and Yallew (2007) indicated improved variety development lacks fitness attributes to the farmers' preference traits. Also harsh growing conditions hinder the adoption rate of the breeders developed varieties due to less adaptation when compared to landrace varieties. Thus, a balance between farmers-preferred traits and solutions to production constraints should be the breeders' goal in order to enhance cultivar uptake by farmers. Sorghum landraces are invaluable sources of genetic variations for different socio-economic traits which include pest and disease resistance, early maturity, yield potential and other desired traits. These genetic resources have long agricultural histories and have co-evolved with different pests and disease. *Striga* resistance could possibly be selected from landraces, cultivated and wild sorghum species for further exploitation in sorghum breeding programs.

Participatory rural appraisal (PRA) tools are utilized to document farmers' traditional knowledge and experiences to mitigate food insecurity and improve their livelihood (Chambers, 1992). Adoption of improved sorghum technologies by smallholder subsistence farmers has been very low because the new technologies did not meet the requirements of farmers (Singh and Morris, 1997). Conventional breeding programs should be reformed to incorporate farmers preferred traits in varietal development programs (Ceccarelli et al., 2001). Therefore, in order to develop and enhance the acceptability and adoption of new varieties and improved production technologies the farmers' actual production constraints and varietal preferences should be well-known (Soleri et al., 2000; Ceccarelli et al., 2001).

The objective of this study was to determine farmers' sorghum production opportunities, threats, indigenous knowledge and perceptions, emphasising breeding priorities and *Striga* infestation, and the coping mechanisms of farmers in the north eastern and north western Ethiopia.

2.3 Materials and Methods

2.3.1 Description of study sites

The survey was undertaken in three administrative zones of Ethiopia, namely the north Shewa and north Wello zones of the Amhara Regional State and the Metekel zone of Benshangul-Gumez Regional State. North Shewa and north Wello are located in north eastern Ethiopia representing semi-arid to arid lowland agro-ecologies. Metekel has a humid lowland agro-ecology and is situated in north western Ethiopia, bordering the Sudan. The geographical locations of the study zones are shown in Figure 2.1 and their main agro-ecological features are summarized in Table 2.1.

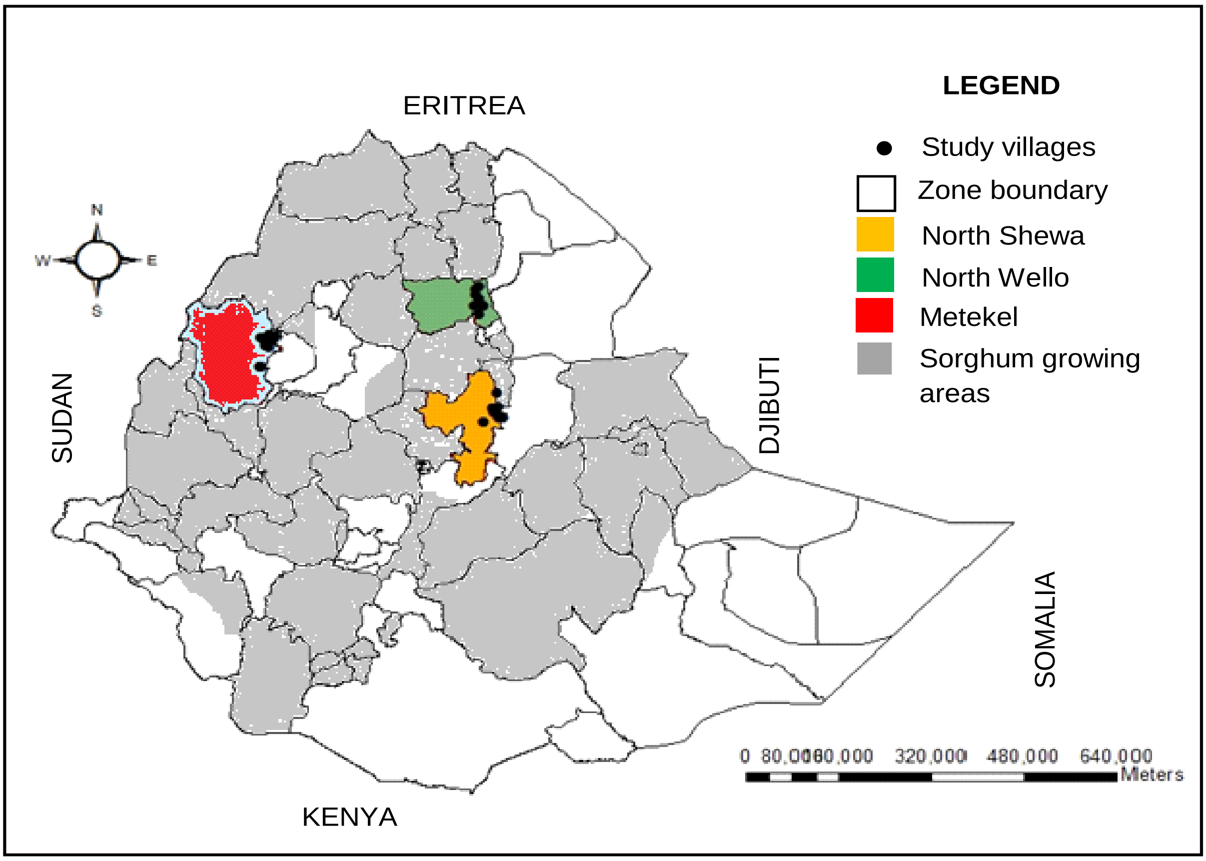

Figure 2.1 Sorghum production map of Ethiopia and location of study zones

Table 2.1 Major agro-ecological characteristic of the study zones

Study zone	Moisture regime	Altitude (masl)[a]	Geographic position	AGP (days)[b]	Temperature (°C)	
					Min	Max
North Shewa	Semi-arid	1200-1800	09° 59′ 98″ N 29° 53′ 90″ E	106-119	17.5	25.0
North Wello	Semi-arid	2000-2400	11° 50′ 02″ N 39° 36′ 37″ E	66-106	17.0	22.5
Metekel	Humid	700-2800	11° 19′ 31″ N 36° 28′.22″ E	150-165	16.0	32.0

[a] masl = meters above sea level

[b] AGP = Average growing period

In all study zones mixed crop and livestock farming is the predominant mode of agricultural production. Sorghum, *tef* and maize are the major cereal crops, together with pulses, oil seeds and some cotton. The study zones are believed to be the sub-diversity centres for sorghum in Ethiopia. Metekel zone is near to the Sudan-Ethiopia border region where sorghum originated and spread into the different parts of Africa and Arabia. The zone is also considered to be the centre of origin of *S. hermonthica* (Tadesse and Yilma, 1991).

2.3.2 Sampling method

A multistage cluster sampling method was used, conforming to the hierarchical administrative set-up of the study sites. A total of nine districts (three districts per zone) were chosen, based on prior information on the relative importance of sorghum, and severity of *Striga* infestation. Subsequently, villages were sampled within each district based on their accessibility. Overall, the survey was conducted in 28 villages selected from nine districts (10 villages from three districts of north Shewa and north Wello and eight villages from three districts of Metekel). A total of 315 farmers that cultivated sorghum during the study period participated in the study. Zone level agricultural experts and district agricultural development offices assisted with the identification of the various sampling districts and villages, and participated in the field data collection. The study was conducted between February and April 2011.

2.3.3 Data collection and analysis

Data were collected through interviews, pair-wise ranking, observations made by transect walks across selected villages, and discussions with focus groups. Semi-structured questionnaires were designed on topics related to the general socio-economic characteristics of the household, varietal development, production constraints, *Striga* infestation, and coping mechanisms towards *Striga* management. In order to understand farmers' preferences in varietal selection and utilization, sorghum traits were predefined through discussion with farmers and ranked by farmers using a pair-wise matrix ranking method. Sorghum varieties being used by farmers were identified by their local names, along with their merits and demerits.

In each district, discussions were held among a group of about 10 key informants and the proceedings were recorded. Key informants were selected among elders, elected council members and farmers renowned within the community. The discussion focused on check-lists set through discussion with farmers. Complementary information was recorded through personal observations in a transect walk through each of the sampled villages. During the transect walk observations were made on crop lands where sorghum has been planted during the growing season, as well as on the different uses of sorghum grain (for food, local beverages) and straw (for feed and construction) across the villages.

Quantitative and qualitative data collected through the questionnaire were coded and subjected for statistical analyses using the Statistical Package for Social Sciences computer software (SPSS Inc., 2005). Descriptive statistics and cross tabulations were performed for data summary. Data were tested for normality and homogeneity of variances prior to test the significance difference. Rank data that violated the assumption of normality was subjected to the non-parametric (Kruskal-Wallis) procedure to test significance differences.

2.4 Results and discussion

2.4.1 Household and demographic characteristics

A total of 315 smallholder farmers (101 in north Shewa, 110 in norh Wello and 104 in Metekel) that had sorghum planted in 2010 cropping season were interviewed for the household survey. Sex, family size, age, education background and off-school training of the respondents are summarized in Table 2.2.

Table 2.2 Proportion (%) of respondents per sex, family size, age group, level of education and off-school training status of the at the 3 study zones (N = 315)

Variable	Zones			Total
	North Shewa	North Wello	Metekel	
Sex				
Male	97.0	93.6	98.1	96.2
Female	3.0	6.4	1.9	3.8
Family size				
< 3	23.8	20.0	15.4	19.7
3-6	49.5	58.2	45.2	51.1
> 6	26.7	21.8	39.4	29.2
Age (years)				
< 30	19.8	21.8	24.0	21.9
30-50	54.5	52.7	64.4	57.1
> 50	25.7	25.5	11.5	21.0
Education				
Illiterate	40.6	54.5	32.7	42.9
Read and write	33.7	20.0	20.2	24.4
Grade 2-6	17.8	20.0	28.8	22.2
Above grade 6	7.9	5.5	18.3	10.5
Off school training				
No	65.3	76.4	90.4	77.5
Yes	34.7	23.6	9.6	22.5

The sample population contained a greater proportion of males (96.2 %) than females (3.8 %), but attempt was made to accommodate women in the informal group discussion. The mean family size of the sampled population was 5.44 (SD = 2.27) and about 80% of interviewed farmers had family sizes greater than 3 persons per household. Family size has a direct implication for the availability of human labour available to the farming system. The majority (78%) of the household heads were above 30 years of age and about 57% of them were able to write and read, or had attended formal school. The proportion of farmers that had attended off-school training given by government agricultural extension officers, non-government organizations and others was higher in north Shewa (34.7%) and north Wello (23.6%) than in Metekel (9.6%). Education levels and experience are expected to influence knowledge and the farming system followed in rural areas. Farmers having sufficient experience and training could provide reliable information about the production systems in each area, which is useful in gathering local and indigenous knowledge and in implementing participatory technology development.

2.4.2 Farming systems

All the sampled farmers practiced both crop and livestock production as major sources of food, feed, and income. Household total crop land in the study season ranged from 0.25 to 20 hectares, with mean farm size of 2.34 hectare (SD = 2.11) per household. Most farmers had a mix of livestock including at least one or two oxen, a few other cattle, and a few sheep or goats, donkeys, chicken and beehives. Crops grown in the study zones included sorghum, *tef*, maize, finger millet, oil crops (sesame and groundnuts), and pulses (soybean and mungbean). The majority of the interviewed farmers allocated most of their land for sorghum as the number one crop during the study season followed by *tef* in north Shewa and north Wello whereas sesame was the second important crop grown in Metekel. Proportion of land allocated for the different crops during the study season is shown in Figure 2.2.

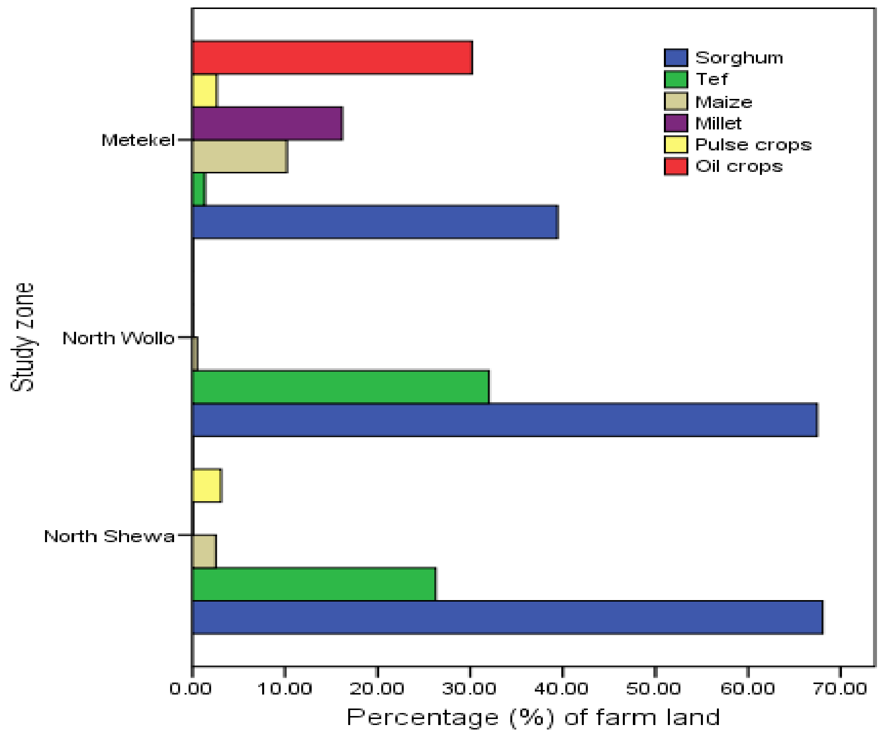

Figure 2.2The proportion of farm land allocated for different crops grown in 2010/11 cropping season in the three study zones

In all study zones sorghum monocropping was the predominant cropping system. Among the interviewed farmers 62.2% in north Shewa, 89.3% in north Wello, and

85.7% in Metekel practice sorghum sole planting and the remaining farmers intercropped sorghum with sesame or sunflower. This cropping system is vulnerable to a high build up and infestation of *Striga* and degradation of soil fertility, which results in yield reduction. According to data gathered during group discussions, proper planting time following the onset of rain is one of the basic success factors to achieve high yield in all study areas. Farmers practice planting immediately after the commencement of the main rain to capture the scarce soil moisture. In the main cropping season (April to December), sorghum planting time varied according to the farmer's choice of variety. Some famers' preferred late maturing landraces planting takes place between April and May, whereas planting of early maturing landraces is carried out in July with the subsequent rains or residual soil moisture. However, late maturing landraces are increasingly being withdrawn from production because of their requirement for long growing period, a negative selection criterion in these highly drought-susceptible areas.

2.4.3 Sorghum productivity

Proportion of interviewed farmers that used various inputs for production of the different crops is summarized in Table 2.3. The predominant seed source for crop cultivation particularly in north Shewa and north Wello zones was seeds of local varieties (landraces or famers' varieties) retained by farmers themselves from previous harvests. A few of the interviewed farmers acquired improved varieties from agricultural offices, research centres or local markets. For example, nearly 13% of farmers in north Shewa, 9% in north Wello and 16% in Metekel have grown improved sorghum varieties during the study season. The present study confirmed that farmers have a deep rooted custom of conserving their own varieties (landraces) for subsequent uses and consequently for transferring to generations with limited adoption of improved varieties.

The use of fertilizer is one of the most widely accepted practices for increasing yield and farm profits in crop production. In all study zones the majority of farmers (74.8 %) do not use either inorganic or organic fertilizer for sorghum production. In Metekel a significantly higher number of farmers (58.1%) grew improved maize varieties. Consequently greater proportion (67%) of farmers applied inorganic fertilizer on their maize farms at the recommended rate. Farmers believed that the use of fertilizer for sorghum production is important. However, unaffordable high costs, the low fertilizer response of landraces and the extreme drought at the end of growing seasons limit the use of inorganic fertilizer by famers. Instead, some farmers, particularly in north Shewa and north Wello, use organic fertilizers such as manure, compost and crop residues, or practice crop rotation with legumes to enhance soil fertility.

Use of pesticides is one effective option to control diseases, insect pests and weeds. In general, the number of farmers using pesticides against crop pests in the study zones was low. Of the respondent farmers 20.8% in north Shewa and 27.7% in Metekel used pesticides on *tef* and finger millet, respectively. During group discussion, the high price of pesticides, limited accessibility and lack of technical knowledge were identified as main reasons for the limited use of pesticides.

Table 2.3 Proportion of farmers that used various inputs for cereal crops production in the study zones

Zones	Crops	Seed type (%)		Fertilizer (%)			Pesticide (%)	
		Local	Improved	Inorganic	Organic	None	Yes	No
North Shewa	Sorghum	87.1	12.9	21.8	17.8	60.4	7.4	92.6
	Tef	100.0	0.0	47.7	13.6	38.6	20.8	79.2
	Maize	100.0	0.0	25.0	0.0	75.0	0.0	100.0
North Wello	Sorghum	91.0	9.0	13.7	18.3	67.9	0.0	100.0
	Tef	96.5	3.5	12.5	25.0	62.5	6.2	93.8
	Maize	100.0	0.0	0.0	0.0	100.0	0.0	100.0
Metekel	Sorghum	83.7	16.3	2.9	1.0	96.2	0.0	100.0
	Tef	50.0	50.0	0.0	25.0	75.0	0.0	100.0
	Maize	41.9	58.1	67.6	24.3	8.1	5.1	94.9
	Finger millet	84.2	15.8	6.5	8.7	84.8	27.7	72.3

The mean seed rate used by farmers, and yields of the different cereal crops in the study areas are shown in Table 2.4. Sorghum seed rates varied depending on soil type, weather condition and tillering capacity of varieties. The overall mean with standard deviation of sorghum seed rate used by farmers was 11.6 ± 9.4 kg ha^{-1}, with a range from 1 to 60 kg ha^{-1}. Compared to the recommended sorghum seed rate of 10 to 15 kg ha^{-1}, about 52% of the interviewed farmers used low sorghum seed rates of 1-10 kg ha^{-1} and 20% of the farmers used higher seed rates of 15 to 60 kg ha^{-1}. Farmers prefer to use higher seeding rates for adequate germination, better stand, and to be safe from losses incurred by different stresses. Lack of information about location specific seeding rate recommendation to farmers identifies the need for further research to assess the economics of using higher seed rates.

The overall mean sorghum yield achieved by farmers during the study season was 1200 kg ha^{-1}, which was lower than the estimated national average yield of 2000 kg ha^{-1} for the same season (CSA, 2011). Furthermore, sorghum yield obtained by farmers exhibited great variability, ranging between zero to 7500 kg per ha. Yield levels vary depending on varieties as well as climatic factors, especially drought, combined with other stresses. Despite the high genetic potential of sorghum for yield (Fisher and Wilson, 1975), the average sorghum yield obtained in Africa has been the lowest globally and is declining (Dogget, 1988). The results of this survey found similar trends in the study zones. The reasons for the decline of yield are mainly because resource-poor smallholder farmers still not have access for production inputs and information, and sorghum production relies on landrace cultivars. These landraces have been selected for their local adaptation, high grain quality and resistance to pre- and post-harvest pest and disease losses rather than their yield potential.

Table 2.4 Mean, standard deviation and range of seed rate and grain yield (kg/ha) of the major cereal crops during the survey season at the three study zones

Zones	Statistics[a]	Sorghum		Tef		Maize		Finger millet	
		Seed rate	Yield	Seed rate	Yield	Seed rate	Yield	Seed rate	Yield
North Shewa	Mean	10.3	1264.4	30.7	668.9	12.1	1380.0	-	-
	SD	6.9	1042.2	19.4	618.1	8.4	1543.4	-	-
	Range	1.0-48.0	0.0-6000.0	0.7-80.0	0.0-3200.0	0.8-24.0	100.0-4000.0	-	-
North Wello	Mean	9.9	1089.6	22.3	353.0	12.0	100.0	-	-
	SD	6.8	1025.2	16.7	363.9	0.0	0.0	-	-
	Range	2.3 - 48	100.0-7500.0	6 -75	0-1600	12.0-12.0	100.0-100.0	-	-
Metekel	Mean	14.5	1277.5	33.3	512.0	27.1	2080.0	36.6	1038.0
	SD	12.6	889.8	9.4	466.1	23.0	2465.0	22.8	712.1
	Range	2.7-60.0	20.0-5200.0	20.0-40.0	200.0-1200.0	3.3-125.0	0.0-15000.0	2.7-100.0	200.0-4800.0
Total	Mean	11.6	1207	25.9	481.7	24.4	1966.0	36.6	1038.0
	SD	9.4	988.9	18.1	502.9	21.7	2371.8	22.8	712.1
	Range	1.0-60.0	0.0-7500.0	0.7-80.0	0.0-3200.0	0.8-125.0	0.0-15000.0	2.7-100.0	200.0-4800.0

[a]SD= Standard deviation

2.4.4 Sorghum production constraints

The major production constraints of sorghum identified by farmers in the study zones are summarized in Table 2.5. In all of the study zones farmers faced crop production challenges due to numerous biotic and abiotic stresses. Farmers identified production constraints such as drought, *Striga*, stalk borer, low soil fertility, unavailability of production inputs (improved seed, fertilizers and pesticides), and storage insects. These constraints were identified as reducing the yield potential of sorghum over time. The Kruskal-Wallis test showed that the relative importance of constraints varied significantly between study zones. *Striga* infestation was identified by 88% of the interviewed farmers as the most severe pest across all zones, causing devastating losses in sorghum. *Striga* severity in the study zones varied considerably from nil to high infestation levels. The variations could be attributed to seed movements and extreme levels of soil infertility that increase *Striga* seed banks in the soil. In sum, the north eastern and north western parts of Ethiopia are the major *Striga* prone areas, affected by recurrent droughts and low soil fertility. Wortmann et al. (2006) reported that the paramount yield reducing factors in sorghum in eastern Africa are low soil fertility (nutrient deficiency), drought, *Striga*, stem borers, and shoot fly.

Following *Striga*, moisture deficit was the most severe constraint identified by 66 and 72% of farmers in the semi-arid to arid areas of north Shewa and north Wello zones, respectively. With recurrent and drastic changes of weather conditions, drought has become a normal phenomenon in north eastern Ethiopia, hampering crop production and restraining the livelihood of many subsistence smallholder farmers. Drought has forced farmers to change their cropping system, abandon some agricultural lands and has forced the most preferred landraces out of production.

Table 2.5 Proportion of farmers rated the severity of major sorghum production constraints across the three study zones

Constraint	**North Shewa**			**North Wello**			**Metekel**			**Kruskal-Wallis test (p=0.05)**
	HS[a]	MS	LS	HS	MS	LS	HS	MS	LS	
Moisture deficit	66.3	17.8	15.8	71.8	10.0	18.2	1.0	8.7	90.4	0.000
Soil fertility	10.9	25.7	63.4	2.7	24.5	72.7	12.5	22.1	65.4	0.170
Production inputs	13.9	15.8	70.3	3.6	16.4	80.0	4.8	18.3	76.9	0.141
Lack of improved variety	29.7	34.7	35.6	9.1	28.2	62.7	29.8	36.5	33.7	0.000
Striga	88.1	6.9	5.0	92.7	5.5	1.8	83.7	6.7	9.7	0.099
Other weeds	15.8	15.8	68.3	15.5	28.2	56.4	14.4	13.5	72.1	0.102
Stalk borer	49.0	25.0	26.0	69.1	14.5	16.4	5.8	12.5	81.8	0.000
Storage insects	24.8	12.9	62.4	25.5	10.9	63.6	35.6	25.0	39.4	0.002
Others disease and pest	16.8	12.9	70.3	1.8	14.5	83.6	8.7	5.8	85.6	0.005
Birds	35.6	25.7	38.6	14.7	17.4	67.9	25.0	26.0	49.0	0.000

[a] HS = highly severe; MS = moderately severe; LS = less severe.

2.4.5 *Striga* infestation and coping mechanisms

Farmers in all study zones indicated that all the major cereal crops are infested by *Striga*. The extent of *Striga* infestation varied between the study zones and crop types (Table 2.6). Sorghum, maize and finger millet were severely infested crops by *Striga*. Many farmers stated the level of infestation on *tef* is low. Farmers in all study areas considered the levels of *Striga* infestations to be increasing in sorghum (Figure 2.3). Relative to the non-*Striga* infested farms, sorghum yields declined progressively by 22.8 and 28.3% as a result of moderate and severe infestations, respectively. An estimated sorghum yield loss of 65% due to moderate to heavy *Striga* infestations was reported in an on-farm integrated *Striga* management study (Tesso et al., 2007).

With regard to the history of *Striga* appearance, farmers in north Shewa and Metekel witnessed its presence since about 20 years ago, primarily in sorghum fields. Later the weed started to infests *tef* fields. The majority of interviewed farmers (> 79%) in north Wello believed that *Striga* existed since time immemorial and were unable to guess the date for first appearance of *Striga* on their farms. According to farmers, *Striga* emerges from degraded lands and the weed is said to be a curse from God. Consequently, farmers are suspicious of the effectiveness of recommended coping mechanisms against the weed. Continuous cropping and extension of cultivation on marginal soils due to population pressure have resulted in the spread and intensification of the *Striga* problem (Parker, 1991). Welsh and Mohamed (2011) reported that the rift valley areas could be the source of *Striga* diversity and persistence presence in alignment to this study. Farmers in north Wello have a very long tradition of crop cultivation, which has been characterized by severe land degradation and drought events that favour *Striga* infestation. During group discussions most farmers in north Shewa suspected that *Striga* might have come from north Wello to their farms through seed exchange.

To reduce *Striga* infestations farmers use hand weeding, burning and manure application. In north Shewa and north Wello 75% of farmers considered *Striga* hand weeding at its flowering stage to be the most effective practice to reduce its impact in subsequent cropping seasons. This practice is known to preclude *Striga* seed setting and addition to the seed bank. In Metekel only 50% of the respondents thought the hand weeding were successful in reducing levels of *Striga* while the remaining 50%

claimed it to be ineffective. Research findings in different areas show that traditional control options such as hand weeding are insufficient to eradicate *Striga* once it is well established in the field (Woomer et al., 2004).

Farmers in the study areas have limited awareness of the recommended solutions against the noxious parasitic weed. A few of the farmers adopted use of improved technologies such as growing *Striga* resistant sorghum varieties, intercropping with legumes, crop rotation and herbicide application. Reasons given for the very limited adoption by farmers of *Striga*-controlling technologies were lack of information, fear of risks associated with the novel technologies, financial limitations, their preferences for traditional control methods and a lack of access to seed of resistant varieties.

Table 2.6 Farmers' assessment of levels of *Striga* infestation on the major cereal crops in three study zones

Crop	Extent of *Striga* infestation	North Shewa	North Wello	Metekel	Total
Sorghum	None	34.6	14.3	19.8	22.9
	Mild	29.1	41.1	25.4	31.9
	Severe	36.2	44.6	54.8	45.2
Maize	None	10.2	20.5	35.7	22.1
	Mild	48.0	41.1	27.8	39.0
	Severe	41.7	38.4	36.5	38.9
Tef	None	55.9	57.1	61.9	58.3
	Mild	24.4	22.3	12.7	19.8
	Severe	19.7	20.5	25.4	21.9
Finger millet	None	-	-	19.8	19.8
	Mild	-		50.8	50.8
	Severe	-	-	28.6	28.6

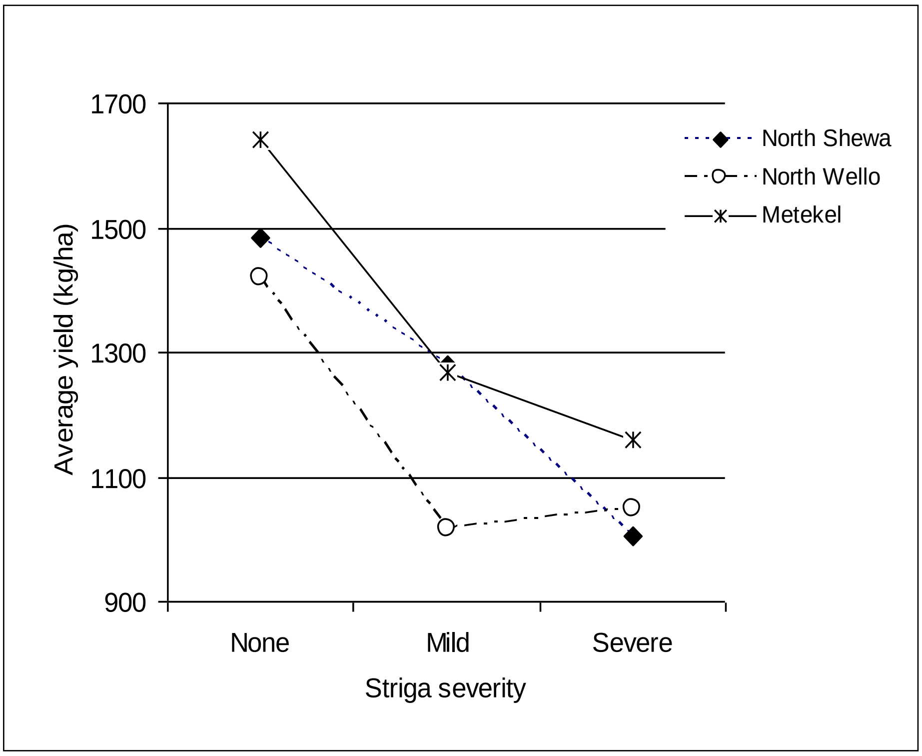

Figure 2.3 Estimates of sorghum yields (kg ha^{-1}) as affected by *Striga* infestation in the three study zones

2.4.6 Sorghum genetic diversity and farmers' preferred traits

Sorghum varieties that were well-known among the community in the study zones were recorded by their local names together with their attributes described by farmers themselves (Table 2.7). Farmers mentioned a broad range of sorghum landraces that had been grown in the area and maintained for generations. Some of the landraces were commonly recognized by most farmers within a study zone and across zones, whereas some were rare varieties known only by a handful of farmers. The large number of landraces shows the existence of diverse genetic resources which have evolved under different agro-ecologies, stressful climatic conditions and low input management systems by smallholder farmers. The diverse agro-ecologies along with farmers' consistent selection and crop management practices have contributed to the existing sorghum diversity in Ethiopia (McGuire et al., 1999; Ayana and Bekele, 2000; Tesso et al., 2008).

Only a small number of improved sorghum varieties were mentioned by farmers. These varieties had low adoption in all the study zones. Farmers associate particular varieties with qualities such as market value, cooking or fodder quality. It is noted that some are resistant to *Striga* and some are susceptible. The present study revealed that famers prefer to cultivate landraces selected among sorghum populations based on their adaptation to moisture stress, resistance to different biotic and abiotic stresses and food quality characteristics. Many farmers in Metekel adopted an improved sorghum variety (*Emahoy*) which was developed for its high yield and *Striga* resistance (Table 2.7). This suggests that crop improvement programs should incorporate farmers' desired traits for successful adoption of improved cultivars. Previous reports are available that focused on sorghum genetic diversity, characterization, and utilisation of the landraces for breeding (Kebede and Menkir, 1987; Benor and Sisay, 2003; Teshome et al., 2007). The advantages with landraces are well recognized mainly because they are best adapted to the local conditions however landraces also possess farmers' preferred attributes, despite their low productivity which should be considered as a source of novel genes for sorghum breeding programmes.

Table 2.7 List of sorghum varieties grown in the study zones and their associated characteristics as described by farmers

Zones	Varieties/local name	Suggested traits
North Shewa	Bedeno, Gambela, **Hormat** ,Esmile	*Striga* and stalk borer resistant, high yielder
	Afeso, Cherekit, Dalgom, Degalit, Dikugn, Gurade, Gurenjo, Hagosaraya, Jeru, Kancha, Keteto, Mayete, Mokake, Mugeayfere, **Serina**, Wediaker, Wegere, Wencho, Zelena, Zetere	Susceptible to *Striga*
	Gurade, Wencho, Wegere, Zelena	High yielder
	Hagosaraya, Jeru, Mayete	Better food quality
	Mayete, Dalgom, Degalit	Bird resistant
	Dikugn, **Esmile**, Mokake, Serina, Wediaker	Early maturing
	Afeso, Cherekit, Degalit, Jeru, Keteto, Mayete,	Susceptible to stalk borer
North Wello	**Birhan**, **Hormat**, white America, white jegurte	Resistant to *Striga*
	Abayere, Abola, Bukasie, Bunegn, Degalit, Hagosaraya, humera, Jamyu, Jegurte, Jeru, Mera, red America, wediaker, Yeju	Susceptible to *Striga*
	Abola, Bukasie, Bunegn, Degalit, Hormat, Yeju	High yielder, resistant to stalk borer
	Abayere, Bukasie, Degalit, Hagosaraya, mera, white America	Better food quality
	Abayere , Bunegn, Hormat, Humera, Jegurte, Mera, white America	Early maturing
Metekel	Birhan, **Emahuye**, **Gobeye**, redgobe	Resistant to *Striga*, early maturing
	Abayere, Cherekit, Dasech, white gobe	Susceptible to *Striga*, better food quality
	Emahuye, Birhan	High yielder, early maturing
	Abayere, Birhan, Cherekit, Emahuye, Dasech	Stalk borer resistant

- Bold faced scripts denote improved varieties

Farmers' ranking of preference traits in sorghum varieties in the study zones is presented in Table 2.8 In north Shewa and north Wello drought resistance, earliness and *Striga* tolerance were the major farmer-preferred traits for sorghum varieties. In the relatively wet Metekel zone, *Striga* resistance, grain quality and drought resistance were the most preferred traits for farmer's choice of sorghum varieties in order of importance.

The notable reasons of such preferences include the occurrence of frequent droughts, erratic rainfall and high *Striga* infestation experienced by farmers over the many decades. In these zones, farmers are shifting to planting early maturing and *Striga* tolerant sorghum varieties. Most of the reported landraces i.e., 80.5% in north Shewa and 72.1% in north Wello are slow maturing and suffer severe losses from drought, and are susceptible to *Striga*. Consistent with the present study Sinafkish et al. (2010) reported that farmers have special preferences of *tef* and sorghum varieties with wider environmental adaptability and yield stability than high grain yield *per se* in north Wello. Mekbib (2006) also noted reasons why farmers prefer specific sorghum landraces included tall plant height, high biomass and grain yield. In the present study most of the sorghum diversity observed in various agro-ecologies is associated with their genetic potential to withstand environmental calamities and to satisfy farmers' needs over time and space.

Findings from this result help to direct the focus of future sorghum breeding programs encompassing farmers-preferred traits. Gyawali et al. (2007) and Mekbib (2006) suggested that farmers' participation in the breeding of crop varieties for low resource farmers is necessary to guarantee acceptance and eventual adoption. Studies by Nkongolo et al. (2008) emphasised the use of farmers' participatory tools to gather farmers' indigenous knowledge on sorghum genetic resources for efficient characterisation and further improvement.

Table 2.8 Preference traits mean scores and ranks in sorghum among three study zones

Preference trait	Zones		
	North Shewa	North Wello	Metekel
Grain yield	4.32 (4)[1]	3.57 (5)	3.46 (6)
Straw yield	1.18 (7)	1.18 (7)	0.71 (7)
Plant height	0.59 (8)	0.36 (8)	0.46 (8)
Disease and pest resistance	3.40 (6)	4.03 (4)	3.91 (5)
Striga resistance	4.74 (3)	4.81 (3)	5.42 (1)
Drought resistance	5.05 (1)	5.48 (1)	5.06 (3)
Grain quality	3.65 (5)	3.49 (6)	5.07 (2)
Earliness	5.07 (2)	5.08 (2)	3.91 (4)

[1] Numbers in parenthesis indicate trait ranks based on 1 to 8 scores. 1 is good and 8 is bad

2.5 Conclusion

Sorghum is the most important crop for food security in harsh environments in Ethiopia, where it is difficult to grow other food crops. In this study a diagnostic appraisal of sorghum production in smallholder subsistence farming system was conducted in three representative sorghum growing administrative zones (north Shewa, north Wello and Metekel) of Ethiopia. The study identified farmers' sorghum production opportunities, threats, indigenous knowledge and perceptions, and priority traits. Drought, *Striga* infestation, declining soil fertility, lack of access to improved varieties and other production inputs, and bird damage identified by farmers as the most important constraints limiting sorghum production. The relative importance of constraints varied considerably within and between the study zones. Drought and *Striga* infestation were the most severe constraints in the semi-arid to arid north Shewa and north Wello zones, while *Striga* was the most severe constraint in the relatively wet Metekel zone. In north Shewa and north Wello zones farmers preferred drought resistance as the most important sorghum varietal selection criterion followed by earliness and *Striga* resistance, whereas in Metekel farmers preferred sorghum varieties with *Striga* resistance as the number one criterion followed by grain quality. These variations in varietal preferences across zones are attributed to differences in moisture regime, soil fertility status, crop management, and genetic diversity, among others.

Farmers in the study zones preferred to grow sorghum landraces despite low yield levels compared to improved released varieties. Farmers indicated that landraces have wide adaptation to their farming system, with relatively high level of drought tolerance to *Striga* and high pest resistance. Thus, sorghum improvement should be directed in developing superior sorghum cultivars with enhanced yield potential combined with *Striga* and drought resistance using farmers-preferred traits.

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CHAPTER THREE

Evaluation of sorghum genotypes compatibility with *Fusarium oxysporum* under *Striga* infestation

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CHAPTER 3

Evaluation of sorghum genotypes compatibility with *Fusarium oxysporum* under *Striga* infestation

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3.1 Abstract

Combined use of host resistance and *Fusarium oxysporum* as biocontrol agent may serve as a viable option in integrated management of *Striga* in sorghum (*Sorghum biocolor* L. Moench). The objective of this study was sorghum genotypes evaluation for *F. oxysporum* compatibility and farmers' preferred traits under *Striga* infestation. Fifty sorghum genotypes were evaluated in the greenhouse on *Striga* infested soils with and without inoculation by *F. oxysporum*. A supplementary laboratory study was conducted to investigate the growth and proliferation of *F. oxysporum* in the soil and sorghum roots. Experiments were conducted at Ambo Plant protection Center/Ethiopia in 2011. Data were recorded on crop growth parameters, *Striga* incidence and colonization and persistence of *F. oxysporum* in the soil and plant roots using samples taken at 45 and 60 days after planting. Inoculation with *F. oxysporum* significantly enhanced flowering, maturity compared to non-inoculated plants. The plants were taller in treated pots; panicle length, biomass and seed yield per plant were also higher in *F. oxysporum* treated pots. Likewise, *Striga* emergence was delayed and vigour and overall incidence of the parasite was significantly reduced in *Fusarium* treated pots. The number of *Fusarium* colony forming units obtained from soil and plant samples were significantly different between genotypes. Three principal components (PC) contributed to 67.31% of the total variation among the genotypes. PC1, PC2, and PC3 contributed 27, 23, and 18%, respectively, to the total variance. Days to *Striga* emergence, count and height correlated with PC1 while sorghum panicle length and plant height with PC2 and days to sorghum flowering and maturity with PC3.Thus, twelve promising sorghum lines were identified with considerable level of resistance to *Striga* and *F. oxysporum* compatibility which is valuable in the strategic resistance breeding of sorghum through integrated use of host resistance and *F. oxysporum* inoculation.

Key words: *Fusarium oxysporum*, integrated *Striga* management, sorghum, *Striga*

3.2 Introduction

Striga hermonthica is an obligate root hemi-parasitic weed which belongs to the family *Orobanchaceae* (formerly S*crophulariaceae*). It inflicts a significant yield reduction in cereal crops including sorghum, millets, maize and rice. The weed is endemic to sub-Saharan Africa and infests about 26 to 50 million hectares causing annual crop losses ranging from 30 to 90%, and sometimes leading to complete yield loss (Watson et al., 2007). Its effect depends on the crop genotype, degree of infestation, rainfall pattern and fertility of the soil (Aly, 2007; Ejeta, 2007; Watson et al., 2007). *Striga* remains increasingly a major crop production constraint mainly for subsistence farmers that effective, affordable and sustainable control options are needed to enhance small scale sorghum productivity in areas where the parasite occurs.

Several control options have been recommended to reduce *Striga* damage such as the use of resistant cultivars, crop rotation, intercropping with pulse crops, late planting, deep planting, using trap crops, use of organic and inorganic fertilizers, herbicides, and biological control (Hearne, 2008). Although the level of *Striga* infestation and damage is increasing, farmers rarely adopt *Striga* control methods either due to limitations associated with the technology itself, access and costs of the technology or due to lack of information about available technology options (Oswald, 2005; Hearne, 2008). Furthermore, available options when applied individually are not effective and sometimes affected by environmental conditions. Thus several options need to be integrated in order to achieve sustained and successful *Striga* control.

Biological control of *S. hermonthica* using *Fusarium oxysporum* is considered as one of the novel management strategies (Sauerborn et al., 2007). Fungi are preferred to other microorganisms as bio-herbicides because they are usually host specific, highly aggressive, and easy to mass produce and are genetically diverse (Abbasher et al., 1998; Ciotola et al., 2000). Field and laboratory tests showed that *F. oxysporum* is highly effective in hindering germination, growth and development of *Striga* and thus may lead to reduction of *Striga* seed bank in the soil (Ciotola et al., 2000). Therefore the use of *F. oxysporum* in combination with other cost effective control methods may provide an effective and sustainable control option for subsistence farmers.

The use of resistant varieties has been highlighted as the most effective and environmentally sound method for the control of *Striga*. This has been demonstrated in multi-location field tests conducted in Ethiopia and Tanzania (Mbuwaga et al., 2007; Tesso et al., 2007). In Ethiopia, about 30% of low altitude (<1500 masl) areas where sorghum is the predominant staple crop are infested with *Striga* (Watson et al., 2007) which causes yield losses ranging from 50 to 100% (Hussien, 2006; Tesso et al., 2007). Recently, few high yielding *Striga* resistant sorghum varieties have been introduced and released in the country (Adugna, 2007; Ejeta, 2007). These varieties when deployed along with moisture conservation practices and soil amendment inputs can dramatically reduce *Striga* infestation and increased sorghum yield by up to 400%. However, adoption of these varieties has been slow primarily due to the introduced germplasm do not fulfill farmers' preferred traits (Adugna, 2007), and lack of effective seed production and delivery mechanism.

Previous studies have indicated that *F. oxysporum* can reduce *Striga* incidence by attacking the parasite both above and below ground without affecting the host. Thus besides the use of resistant sorghum varieties, inoculation with the effective (virulent) strain of *F. oxysporum* can be considered as an additional component of the existing integrated *Striga* management package. In order to exploit the potential synergy from this integrated option, research efforts should be directed at developing superior *Striga* resistant sorghum varieties compatible with *F. oxysporum*. The rhizosphere of the ideal variety should favour active growth of *F. oxysporum* and at the same time the fungus should be virulent to the parasite and harmless to the host plant. Therefore, the objective of this study was to identify sorghum genotypes compatible with *F. oxysporum* and determine the potential of deploying this bio-agent on various genotypes for controlling *Striga* using artificially infested soils.

3.3 Materials and methods

3.3.1 Plant material

The study involved fifty sorghum genotypes obtained from the Institute of Biodiversity Center (IBC)/Ethiopia and International Crop Research Institute for the Semi-arid Tropics (ICRISAT). Table 3.1 presents list and sources of sorghum genotypes included in the study. Genotypes from the IBC were collection from low altitude areas (<2000 masl) in the north eastern region of Ethiopia. The ICRISAT genotypes were *Striga* tolerant and they have medium maturity .Shewa Robit local variety which is susceptible to *Striga* and Teshale, improved variety which is tolerant to stalk borer were also included in the study. Seeds of *S. hermonthica* were collected from similar areas where the IBC sorghum genotypes come from in the previous growing season (2010) and used in the experiments after tests for their viability and preconditioning.

Table 3.1 List and sources of sorghum genotypes used in the study

Entry	Accession number/Name	Source	Entry	Accession number/Name	Source
1	23840	IBC	26	238421	IBC
2	69310	IBC	27	238436	IBC
3	212541	IBC	28	238437	IBC
4	234095	IBC	29	238439	IBC
5	235464	IBC	30	238441	IBC
6	235466	IBC	31	238445	IBC
7	235467	IBC	32	238447	IBC
8	235761	IBC	33	238448	IBC
9	235763	IBC	34	238449	IBC
10	235921	IBC	35	239210	IBC
11	235924	IBC	36	239235	IBC
12	235925	IBC	37	239236	IBC
13	235926	IBC	38	243681	IBC
14	235927	IBC	39	243684	IBC
15	235929	IBC	40	244711	IBC
16	235930	IBC	41	244712	IBC
17	235931	IBC	42	244713	IBC
18	237256	IBC	43	2384442	IBC
19	237263	IBC	44	2384443	IBC
20	237267	IBC	45	IS 9830	ICRISAT
21	237283	IBC	46	ICB 587	ICRISAT (*Striga* tolerant and medium maturity)
22	237289	IBC	47	ICSB 570	ICRISAT(*Striga* tolerant and medium maturity)
23	238400	IBC	48	ICSB 576	ICRISAT(*Striga* tolerant and medium maturity)
24	238402	IBC	49	SR local	Shewa Robit district farmers and susceptible to *Striga*
25	238420	IBC	50	Teshale (3443-2-OP)	Improved variety adapted to lowland areas (resistance to major diseases and stalk borer)

3.3.2 *F. oxysporum* inoculum

F. oxysporum was isolated from severely diseased *Striga* plants collected from sorghum fields in north eastern lowlands of Ethiopia. Taxonomic identification of the isolate was confirmed by the Phytomedicine Department of Humboldt University in Berlin, Germany. Pathogenicity and host specificity of the isolate to *Striga* was confirmed in our earlier study (Rebeka, 2007). The isolate was maintained on Special Nutrient Agar (SNA) medium at -40^{o}C. Pure *Fusarium* chlamydospores from cultures grown on potato dextrose agar (PDA) were sampled and mass produced at Plant Health Products (pty) Ltd, Kwazulu-Natal, South Africa.

3.3.3 Greenhouse experiment

Greenhouse experiment was conducted to evaluate sorghum genotypes for *Striga* resistance with and without application of *F. oxysporum*. The greenhouse experiment was established at Ambo Plant Protection Centre (APPC)/Ethiopia using 300 plastic pots of 10 l capacity filled with sterilized black, red and sand soil in the ratio of 2:1:1, respectively. All the pots were artificially and uniformly infested with 20 milligrams of surface sterilized viable *Striga* seeds on the top 5 cm soil. The *Striga* seed was collected and stored at the weed science section of APPC. Briefly, *Striga* seeds were sterilized with 1% NaOCl for 10 minutes and washed with suction pump using distilled water and air dried. Half of the pots (150), three for each genotype, were planted to sorghum seeds dressed with 75 mg of *F. oxysporum* chlamydospores while the other half were planted to each genotype without fungal inoculation. All pots were planted after the *Striga* seeds have been preconditioned in the soil for 15 days. After emergence the sorghum plants were thinned to one seedling per pot. Treatments were laid out in a factorial randomised complete block design in three replications. After the experiments *Striga* infested soil was dip buried to prevenet further dispersal.

3.3.4 Laboratory experiment

A laboratory study was conducted to observe sorghum genotypes rhizosphere variation for the active growth of *F. oxysporum*. Sorghum seeds dressed with *F. oxysporum* were planted in completely randomized design with three replications similar to the greenhouse experiment. After 45 and 60 days after planting, soil and plant root samples were taken to monitor changes in *F. oxysporum* propagule densities in the soil to evaluate their persistence under sorghum rhizosphere environments. The *Fusarium* propagules per gram of soil were determined using a modified serial dilution plate technique as described by (Nash and Synder, 1962; Stapleton and Devay, 1982). A cylindrical corer, 10 cm in depth and 1 cm in diameter, was used to remove sub-samples of soil from the infested top soil of each pot. Three subsamples, one from the centre and two from opposite sides of each pot collected and were mixed together to form a composite sample. The samples collected from each pot were air dried, crushed, mixed thoroughly and sieved using a 600 mm mesh. The samples were then diluted in 0.05% water agar and 1 ml aliquots spread onto three peptone-pentachloronitrobenzene agar (PPA). The cultures were incubated for seven days, and then the concentration of colony forming units (CFU) per gram of sample determined.

The compatibility of *F. oxysporum* with sorghum roots was studied using root samples collected from each pot. The roots were cut into small pieces and surface sterilized in 5% sodium hypochlorite for 3 minutes. Then the samples were placed into petri dishes containing potato dextrose agar (PDA). The plates were incubated at 25^{o}C for seven days for visible fungal growth. After the incubation period, microscopic observation was carried out to observe the number of colony forming units (CFU).

3.3.5 Data collection and analysis

Data from the greenhouse experiment was collected on both the sorghum and *Striga*. Data on sorghum include number of days to 50% emergence, days to 50% flowering and maturity, plant height (in centimetres) at 50% flowering, panicle length (in centimetres), fresh biomass (in gram per pot), and seed yield (in gram per pot). Data on *Striga* include days to first *Striga* emergence, number of emerged *Striga* plant, and *Striga* height (in centimetres) at seven weeks after planting. For the laboratory study, data were collected on CFU by counting the colonies generated from the soil and plant root samples.

For statistical analysis, all data collected from the greenhouse experiment were subjected to the standard analysis of variance procedures using the SAS statistical program (SAS, 2002). The two-way ANOVA was applied to evaluate the individual and interaction effects of the two factors (genotypes and *Fusarium* treatment).

Independent samples t-test was used to assess the difference between the coated and uncoated sorghum genotypes performance and *Striga* emergence and growth. Significant differences between the mean values were determined by Fisher's Least Significant Differences (LSD) at a significance level of $p \leq 0.05$. The Principal Component Analysis (PCA) was conducted on the average values obtained from the 50 sorghum accessions in three replications using SPSS computer package (SPSS, 2005). For the PCA analyses, entry values were used as rows of the input matrix and *Fusarium*, sorghum and *Striga* traits as column variables of the matrix. *Fusarium* CFU count data from the laboratory study were log transformed and then subjected to analysis of variance.

3.4 Results and discussion

3.4.1 Greenhouse experiment

Coating sorghum seeds with *F. oxysporum* significantly affected both crop and Striga parameters except no genotype differences on days to emergence of the sorghum crop (Table 3.2). *F. oxysporum* appears to have markedly suppressed the establishment of the parasite such that the number of emerged *Striga* and mean Striga height were significantly ($p<0.001$) reduced under *Fusarium* inoculation compared to the non-inoculated treatments (Table 3.2). Likewise, sorghum grain yield was significantly different with grains harvested from pots planted to *Fusarium* coated seeds being 144% higher than those harvested from uncoated seeds. Crop maturity was delayed by an average of 14days in uncoated treatments as compared to those with coated seeds (Table 3.3). This seems to be *F.oxysporum* dramatically affect the adverse effect of the parasite on sorghum growth and development.

Table 3.2 Analysis of variance on traits measured from sorghum and *Striga* among 50 sorghum accessions tested with and without *F.oxysporum* application

Sources of variation	Traits										
	Sorghum [a]									*Striga*[b]	
	DF[c]	DEM	DFL	DM	PHT	PL	BM	GY	DEM	CNT	PHT
Replication	2	4.16	201.99	460.39	3129.46	18.98	5962.16	2188.37	2056.20	529.46	391.90
F. oxysporum	1	14.45**	474.78	6808.22**	2546.46	424.83*	3905.30	7240.29*	102490.08***	9097.01***	3080.65***
Variety	49	1.69	290.34*	432.79**	5187.35*	73.42	4802.97*	191.22	275.19	107.14	67.32
F. oxysporum x variety	49	1.48	101.69	213.24*	2722.78	57.38	2315.80	249.56	571.64**	112.65	57.35
Error	177	1.85	183.02	127.48	2946.46	57.62	3074.88	219.37	336.23	97.66	52.94

*, **and ***= significantly different at 0.05, 0.01, and 0.001 probability levels, respectively.

[a] DEM = days to emergence; DFL = days to flowering; DM = days to maturity; PHT = sorghum plant height; PL = sorghum panicle length; BM = sorghum biomass; GY = sorghum grain yield;

[b] DEM= *Striga* days to emergence; CNT= number of *Striga*; PHT= *Striga* plant height;

c DF=Degrees of freedom.

Table 3.3 Mean days to emergence, maturity, and seed yield in sorghum and *Striga* count and height when tested with and without *Fusarium* treatments

Treatment	Sorghum			*Striga*	
	Emergence (days)	Maturity (days)	Seed yield (g/plant)	Count/pot	Height (cm)
With *Fusarium*	4.2	104.8	16.6	0.8	0.9
Without *Fusarium*	4.7	118.1	6.8	11.9	7.4
t-value	2.08	5.19	5.73	9.09	7.03
Significance level	NS[†]	***	***	***	***

[†] NS, non-significant

***, significant differences at 0.001 probability levels

The number of *Striga* count on untreated pots was very high. Contrastingly, *Fusarium* dressing significantly suppressed *Striga* germination and emergence. The effects of *Fusarium* application on sorghum seed yield and *Striga* plant count per pot in the fifty sorghum genotypes is displayed in Figure 3.1. The figure indicated discrepancies observed among the sorghum genotypes both under *Fusarium* inoculated and un-inoculated treatments. Most of the treated sorghum genotypes seed yield was considerably higher than the untreated genotypes. In contrast few treated sorghum genotypes, their yield performance were not substantially different from the untreated genotypes. These results were due to less number of *Striga* infestations on the *Fusarium* treated pots as compared to the high number of *Striga* on the untreated sorghum (Figure 3.1).

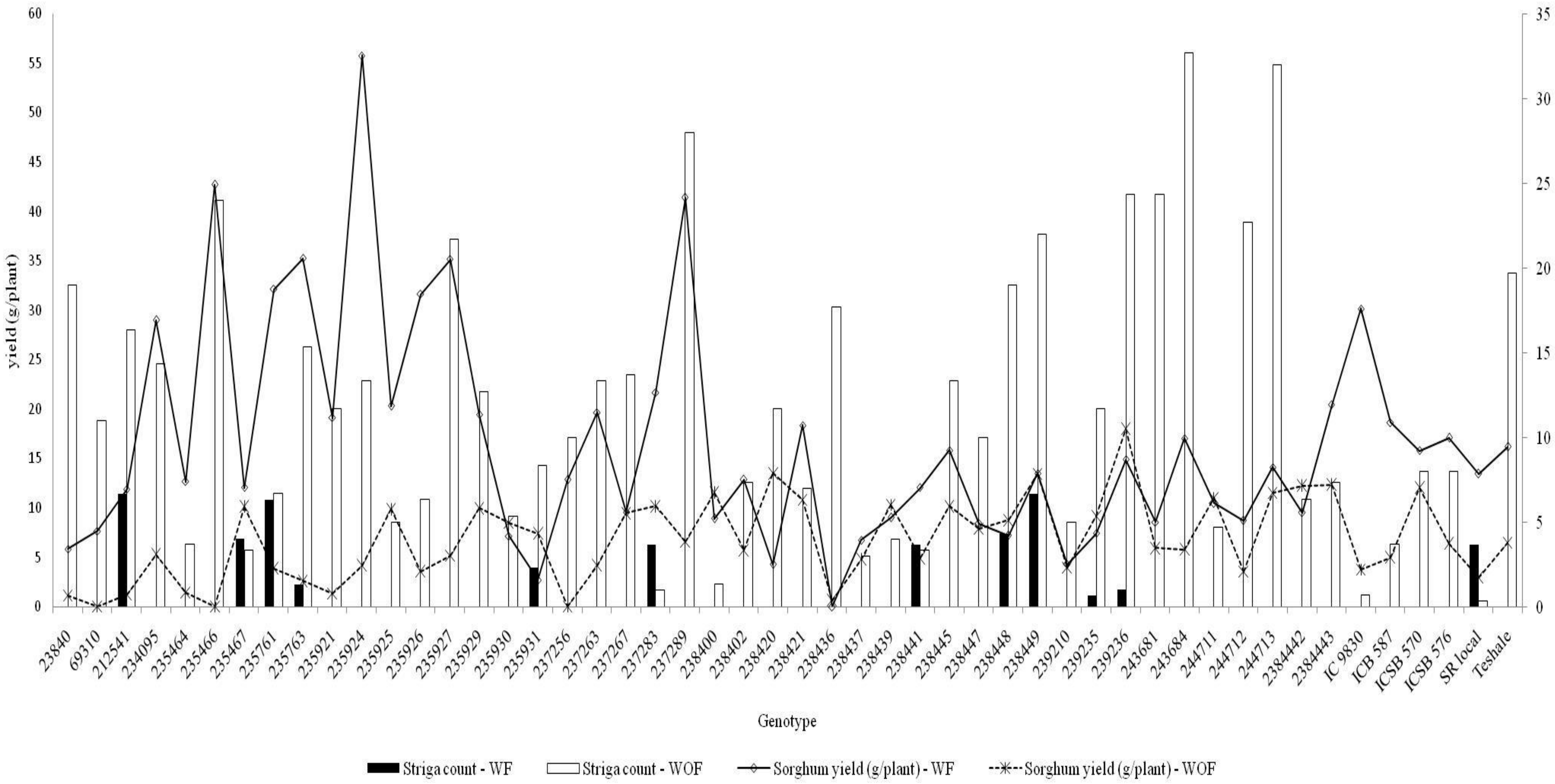

Figure 3.1 Effects of *Fusarium* application on sorghum seed yield (g/plant) and *Striga* count per pot on 50 sorghum genotype

Fusarium treatment for *Striga* control show differential responses on sorghum genotypes tested to *Fusarium* applications (Table 3.4). *Fusarium* dressed sorghum on average matured 13 days earlier than undressed seeds. *Fusarium* application reduced days to maturity in 39 sorghum varieties by 0.17 to 41 days but days to maturity were delayed in 7 of the varieties tested by 1 to 37 days. Compared to untreated seeds, *Fusarium* dressing improved sorghum panicle length and seed yield by 24.8 % and 14.50 %, respectively. Significant variations existed in days to 50% flowering (49-87), plant height (67-155 cm), biomass (38-170 g/plant) and seed yield (2.7-56 g/plant) (Table 3.4.

Fusarium inoculation significantly reduced the *Striga* emergence (11-44 days), incidence (0-7) and height (1-11 cm) (Table 3.5). *Striga* plants emerged in only 12 *Fusarium* dressed sorghum varieties three weeks after sowing while *Striga* emerged in all *Fusarium* undressed sorghum varieties six weeks after sowing ranging from 15-66 days. *Fusarium* application significantly reduced *Striga* count at 92.6% and height at 86.9% (Table 3.5).

Table 3.4 Mean responses measured on sorghum agronomic characters among 50 sorghum genotypes with and without *F. oxysporum* application

NO	Accession number	DEM		DFL		DM		PHT		PL		BMS		GY	
		+	-	+	-	+	-	+	-	+	-	+	-	+	-
1	23840	5.3	4.7	79.9	68.2	117.6	116.8	66.7	23.3	8.8	2.7	61.3	29.0	5.8	1.1
2	69310	5.0	4.7	62.6	85.6	88.0	-	92.3	93.3	8.7	-	76.1	93.1	7.6	-
3	212541	4.3	4.3	84.3	82.1	111.0	140.2	86.7	111.6	17.0	7.7	163.6	173.6	11.9	1.2
4	234095	4.3	4.3	80.3	86.4	108.3	141.6	183.0	141.6	16.7	9.0	84.9	124.4	29.0	5.3
5	235464	4.3	4.3	64.7	77.9	88.3	114.9	114.3	130.0	14.3	10.3	95.7	132.4	12.7	1.4
6	**235466¶**	**4.3**	**3.9**	**76.3**	**-**	**104.0**	**-**	**135.3**	**-**	**18.6**	**-**	**115.9**	**-**	**42.7**	**-**
7	235467	6.7	4.7	80.0	78.6	104.0	113.5	110.3	135.0	18.2	15.0	105.9	110.7	12.1	10.1
8	**235761**	**4.0**	**3.9**	**77.1**	**72.9**	**93.5**	**101.4**	**94.3**	**61.6**	**9.0**	**5.6**	**137.0**	**122.8**	**32.1**	**3.9**
9	**235763**	**5.7**	**5.0**	**85.6**	**87.1**	**119.0**	**115.2**	**97.7**	**57.6**	**13.3**	**4.3**	**119.5**	**90.4**	**35.3**	**2.6**
10	**235921**	**4.0**	**4.0**	**55.4**	**57.4**	**95.6**	**100.2**	**146.7**	**89.7**	**11.0**	**5.3**	**95.9**	**76.9**	**19.1**	**1.3**
11	**235924**	**4.3**	**4.3**	**70.3**	**88.0**	**110.3**	**134.4**	**155.0**	**157.7**	**19.0**	**12.3**	**153.9**	**118.9**	**55.8**	**4.2**
12	235925	6.7	4.0	78.9	86.3	106.6	115.3	73.3	102.7	8.7	12.3	86.5	121.1	20.3	9.9
13	**235926**	**4.0**	**5.1**	**57.7**	**78.7**	**88.3**	**120.9**	**131.7**	**81.7**	**13.16**	**4.7**	**129.6**	**77.1**	**31.6**	**3.6**
14	**235927**	**4.0**	**4.0**	**81.0**	**79.9**	**115.3**	**114.4**	**128.3**	**135.0**	**15.3**	**14.6**	**139.0**	**119.4**	**35.2**	**5.2**
15	**235929**	**4.0**	**4.3**	**59.0**	**65.9**	**88.7**	**110.4**	**123.3**	**127.0**	**10.7**	**7.3**	**132.8**	**122.6**	**19.4**	**9.9**
16	235930	4.4	4.0	56.4	63.9	84.8	102.9	90.0	112.3	10.8	9.0	51.5	81.4	7.2	8.4
17	235931	4.4	3.9	61.7	71.9	91.9	111.9	88.3	106.7	4.7	8.7	40.2	41.1	2.7	7.4
18	237256	4.3	4.4	85.7	-	112.0	-	135.7	-	11.0	-	137.1	52.3	12.9	-
19	237263	4.3	4.6	78.3	84.2	101.3	118.8	138.3	137.6	15.3	3.3	137.7	109.1	19.7	4.2
20	237267	4.7	6.0	75.6	87.6	114.5	119.3	138.3	145.0	9.0	15.3	130.3	151.6	9.6	9.5
21	237283	4.0	4.0	75.0	70.0	101.3	128.0	101.0	118.7	14.0	16.0	122.4	93.0	21.7	10.2
22	**237289**	**4.3**	**4.3**	**78.3**	**82.1**	**110.6**	**113.0**	**154.3**	**81.7**	**17.0**	**10.3**	**128.3**	**63.8**	**41.4**	**6.6**
23	238400	4.3	4.0	84	86.3	120.5	130.0	115.0	133.0	10.0	11.7	125.1	135.5	8.9	11.6
24	238402	7.0	4.0	75.0	83.6	113.3	122.5	119.0	70.3	12.0	11.3	109.6	63.9	12.9	5.7
25	238420	5.3	4.0	85.9	78.3	113.9	126.0	138.7	158.3	5.7	17.7	134.2	138.8	4.3	13.5
26	238421	4.3	4.0	87.0	68.3	118.3	117.3	164.3	125.7	17.3	15.7	105.5	113.3	18.4	10.8
27	238436	4.4	4.0	66.9	93.1	-	122.2	65.0	98.3	-	6.7	90.7	114.6	-	0.6
28	238437	4.0	3.9	83.9	75.1	130.5	125.5	119.7	98.3	14.0	7.0	114.7	84.4	6.8	4.8
29	238439	3.9	4.0	56.9	74.9	89.8	113.9	80.0	152.0	9.8	12.2	70.9	88.5	9.0	10.3
30	238441	4.3	4.0	74.7	77.9	109.0	120.6	147.7	121.7	14.0	8.3	70.0	93.8	12.1	4.9
31	238445	4.3	4.0	49.3	61.0	75.0	98.3	129.3	157.3	14.7	16.0	124.4	108.0	15.8	10.2
32	238447	4.0	4.3	70.4	68.6	112.4	114.5	90.0	141.0	8.7	9.7	54.0	111.3	8.4	7.8
33	238448	3.9	3.9	68.9	67.1	103.4	125.5	61.7	89.3	6.7	11.0	106.4	68.6	7.2	8.8
34	238449	6.7	4.3	74.4	67.0	103.1	106.0	107.3	138.3	11.3	15.7	105.3	103.0	13.5	13.4
35	239210	3.9	4.0	80.2	72.1	121.8	116.8	96.7	139.3	2.0	2.7	121.0	172.7	4.3	3.9
36	239235	4.4	4.0	81.7	64.1	118.9	111.0	48.3	130	6.7	9.3	68.8	122.7	7.4	9.2
37	239236	4.0	4.3	59.3	72.0	118.3	135.7	91.7	98.3	16.0	19.3	81.8	72.0	14.9	18.0
38	243681	4.4	4.6	79.9	77.4	113.8	118.9	104.3	88.3	11.3	8.3	48.9	55.0	8.6	5.9
39	**243684**	**4.7**	**3.9**	**70.0**	**79.1**	**99.0**	**136.8**	**125.7**	**76.3**	**20.3**	**5.7**	**161.1**	**106.2**	**17.0**	**5.8**
40	244711	7.0	4.3	64.9	66.6	97.6	119.5	186.7	170.3	8.7	14.3	69.2	100.5	10.5	11.0
41	244712	4.6	4.0	63.9	66.6	110.6	119.5	111.7	116.6	11.0	12.6	80.3	116.8	8.7	3.5
42	244713	4.3	4.0	74.0	73.0	101.0	126.0	199.0	152.6	20.0	12.3	109.2	143.9	14.1	11.5
43	2384442	7.9	4.0	66.9	68.0	96.3	111.0	112.3	158.6	12.3	13.3	40.8	83.4	9.5	12.3
44	**2384443**	**4.3**	**4.3**	**62.7**	**66.7**	**82.1**	**110.0**	**133.7**	**120.3**	**9.0**	**13.0**	**137.8**	**104.0**	**20.4**	**12.3**
45	**IS 9830**	**4.0**	**4.0**	**72.3**	**78.4**	**109.0**	**131.6**	**132.7**	**106.3**	**15.0**	**5.7**	**152.3**	**53.1**	**30.2**	**3.7**
46	ICB 587	4.1	4.3	60.1	59.4	102.2	120.9	36.7	66.7	5.0	6.3	38.3	60.4	18.7	5.0
47	ICSB 570	6.7	4.3	60.9	74.3	91.1	120.0	78.3	100.7	12.3	15.7	105.8	93.1	15.8	12.0
48	ICSB 576	4.3	4.7	86.0	94.4	115.0	141.6	96.0	74.0	13.3	6.3	115.1	73.2	17.1	6.4
49	SR local	4.3	3.9	75.0	77.2	116.0	75.8	171.0	110.0	13.0	5.3	170.0	133.0	13.5	2.9
50	Teshale	4.3	4.0	72.7	55.9	107.7	87.1	154.7	141.7	14.7	13.0	105.8	82.8	16.2	6.5
Significance level		NS		**		**		**		*		**		**	
	LSD			3.47		3.28		13.15		1.85		12.59		3.83	

†DEM, days to emergence; DFL, days to flowering; DM, days to maturity; PHT, plant height; PL, panicle length; BM, biomass; GY, grain yield; NS, Non-signficant

** denotes significant differences at 0.01 probability level; ‡ + , with *Fusarium;* -, without *Fusarium*; ¶, bold genotypes are selected genotypes

Table 3.5 Mean responses on measured on sorghum agronomic characters among 50 sorghum genotypes with and without *F. oxysporum* application

No	Accession number	SDEM		CNT		SPHT	
		+	-	+	-	+	-
1	23840	-	59.7	0	19.0	-	7.9
2	69310	-	33.0	0	11.0	-	3.1
3	212541	29.7	43.0	6.7	16.3	10.7	4.9
4	234095	-	61.3	0	14.3	-	6.2
5	235464	-	35.7	0	3.7	-	5.7
6	**235466¶**	**-**	**15.3**	**0**	**24.0**	**-**	**1.7**
7	235467	17.0	31.0	4.0	3.3	2.3	3.1
8	**235761**	**44.0**	**21.0**	**6.3**	**6.7**	**10.0**	**1.8**
9	**235763**	**11.0**	**36.7**	**1.3**	**15.3**	**1.0**	**1.1**
10	**235921**	**-**	**65.7**	**0**	**11.7**	**-**	**1.9**
11	**235924**	**-**	**48.7**	**0**	**13.3**	**-**	**7.8**
12	235925	-	36.3	0	5.0	-	4.3
13	**235926**	**-**	**34.7**	**0**	**6.3**	**-**	**2.7**
14	**235927**	**-**	**50.3**	**0**	**21.7**	**-**	**4.7**
15	**235929**	**-**	**47.7**	**0**	**12.7**	**-**	**4.7**
16	235930	-	51.7	0	5.3	-	11.6
17	235931	15.0	15.3	2.3	8.3	2.4	4.3
18	237256	-	49.3	0	10.0	-	7.0
19	237263	-	55.7	0	13.3	-	18.6
20	237267	-	45.3	0	13.3	-	2.5
21	237283	26.7	19.3	3.6	1.0	1.0	0.2
22	**237289**	**-**	**54.7**	**0**	**28.0**	**-**	**12.8**
23	238400	-	43.7	0	1.3	-	2.0
24	238402	-	37.3	0	7.3	-	1.9
25	238420	-	29.7	0	11.7	-	2.7
26	238421	-	52.7	0	7.0	-	4.4
27	238436	-	46.3	0	17.7	-	10.2
28	238437	-	55.3	0	3.0	-	3.5
29	238439	-	46.3	0	4.0	-	8.3
30	238441	10.7	16.0	3.6	3.3	1.7	2.3
31	238445	-	53.3	0	13.3	-	10.9
32	238447	-	53.7	0	10.0	-	9.3
33	238448	12.7	37.0	4.3	19.0	7.3	13.9
34	238449	16.0	47.7	6.7	22.0	4.6	23.2
35	239210	-	35.3	0	5.0	-	2.2
36	239235	16.0	50.7	0.6	11.7	3.0	17.5
37	239236	21.0	49.3	1	24.3	3.6	23.9
38	243681	-	49.3	0	24.3	-	15.3
39	**243684**	**-**	**49.3**	**0**	**32.7**	**-**	**8.4**
40	244711	-	50.3	0	4.7	-	5.4
41	244712	-	49.3	0	22.7	-	18.9
42	244713	-	47.0	0	32.0	-	14.3
43	2384442	-	34.3	0	6.3	-	4.8
44	**2384443**	**-**	**44.0**	**0**	**7.3**	**-**	**9.7**
45	**IS 9830**	**-**	**15.7**	**0**	**0.6**	**-**	**0.7**
46	ICB 587	-	15.7	0	3.6	-	12.0
47	ICSB 570	-	47.7	0	8.0	-	7.3
48	ICSB 576	-	32.7	0	8.0	-	4.2
49	SR local	16.0	17.3	3.7	0.3	1.2	0.3
50	Teshale	-	65.7	0	19.7	-	13.3
Significance level		**		**			**
LSD			4.79		2.58		1.84

[†]SDEM, *Striga* days to emergence; CNT, number of *Striga*; SPHT, *Striga* plant height;

** denotes significant differences at 0.01 probability level;

[‡] + , with *Fusarium;* -, without *Fusarium*; ¶, bold genotypes are selected genotypes

3.4.2 Laboratory experiment

Spore formation of *F. oxysporum* was influenced by sorghum root rhizosphere and varied due to sorghum varieties (Table 3.6). The number of colony forming units counted from soil and plant samples showed significant interactions between sorghum varieties and count intervals ($p<0.05$) i.e. 45 and 60 days after sorghum planting. The difference between sampling intervals was not significantly different (Table 3.6). CFU count at 45 and 60 days after sowing indicated the persistence of the bio-agent on the sorghum root and its rhizosphere and reduced the chance of successful attachment with *Striga* rendering less or negligible number of *Striga* emergence on the treated pots. Average CFU counts from soil and different sorghum genotype root samples are displayed in Figure 3.2. The observed CFU counts were shown the genotypes contribution is different on the applied *Fusarium* persistence and multiplication.

Table 3.6 Analysis of variance with mean square and degrees of freedom on colony forming units (CFU) of *F. oxysporum* collected at two intervals from soil and plant samples among 50 sorghum genotypes

Source of variation	DF	CFU ($X10^3$)g^{-1} soil	CFU($X10^3$) from plant
Rep	2	8.08	5.82
Variety	49	2.48**	3.79**
Interval	1	0.02	1.57
Variety*Interval	49	1.59*	1.48*
Error	196	1.63	2.54
CV (%)		77	74

** and * denotes significant differences at 0.01 and 0.05 probability levels respectively

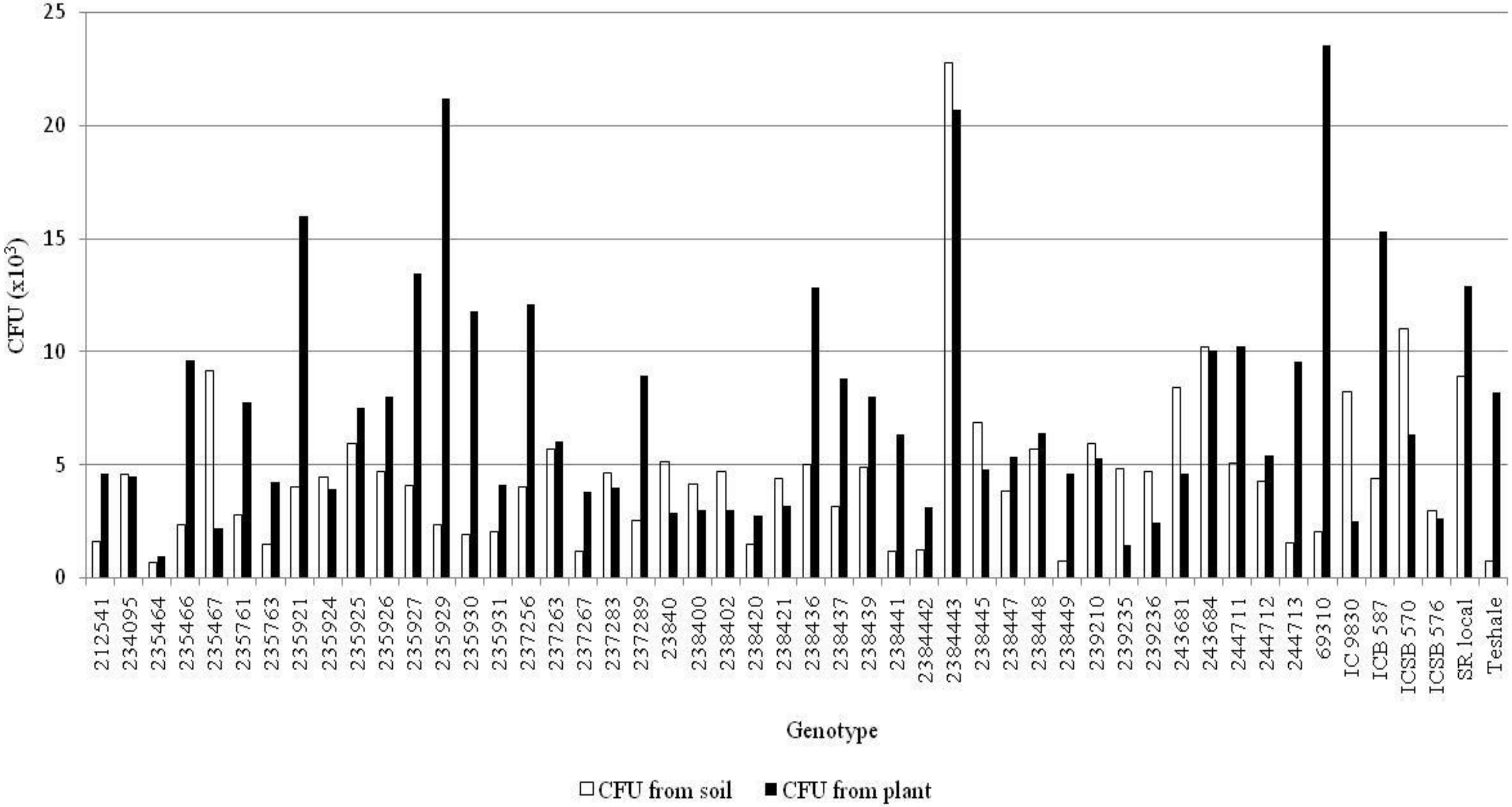

Figure 3.2 Average CFU count from soil and plant samples of 50 sorghum genotypes

3.4.3 Multivariate analysis

Principal factor analysis, after varimax rotation with Kaiser Normalization, provided three principal components (PC) with distinctive sets of groups of highly correlated characters i.e. $r \geq 0.7$ (Table 3.7). The magnitude of component loadings that specified the degree of correlation between a variable and a given factor was used to classify variables into distinct groups (Table 3.7). The three components together explained 67.31 % of the total variance in the original 10 characters measured on sorghum and *Striga*. The first principal component comprising three traits (days to first *Striga* emergence, *Striga* count and *Striga* height) explained 26.95 % of the total variance that related to *Striga* infestation and incidence factor. The second component comprising four sorghum related traits (plant height, panicle length, biomass and seed yield) explained 22.81 % of the total variance followed by PC3 comprising two traits (days to flowering and maturity) contributing 17.54 % of the variance. Based on this component loadings, twelve sorghum genotypes (235763, 235927, 237289, 235466, IC9830, 235924, 235921, 2384443, 235929, 243684, 235761, and 235926) having better compatibility with *Fusarium*, supporting no or minimum number of *Striga*, with relatively higher seed yield and biomass were retained as promising breeding genotypes for integrated *Striga* management study.

Table 3.7 Component loadings and eigenvalues of the principal factor components

Character	Component loadings		
	1	2	3
Days to sorghum emergence	-.415	-.153	.024
Days to sorghum flowering	-.068	.049	**.934**
Days to sorghum maturity	.318	-.088	**.814**
Sorghum plant height	.059	**.780**	.009
Panicle length	.001	**.802**	-.125
Sorghum biomass	.014	**.685**	.314
Sorghum seed yield	-.303	**.688**	-.188
Days to Striga emergence	**.876**	-.159	.192
Striga count	**.870**	-.149	.180
Striga plant height	**.893**	-.080	-.012
Eigenvalue	2.70	2.28	1.75
Cumulative eigenvalue (%)	26.95	22.81	17.54
Proportion of total variance (%)	26.95	49.76	67.31

This study revealed coating the sorghum seeds with *Striga* pathogenic *F. oxysporum* chlamydospore didn’t have adverse effect on the sorghum seed germination and emergence. The t- test has confirmed this hypothesis since there was no statistically significant difference between the treated and untreated sorghum seeds emergence. Other studies (Elzein et al., 2010; Elzein et al., 2006) indicated that sorghum seed germination and emergence was not adversely affected by coating sorghum seeds with *F. oxysporum* chlamydospore propagules. The present study found that seed dressing is beneficial since it is the place where the host and the parasite attachment occur. The bio-agent well pertained on the sorghum root and its rhizosphere and reduced the chance of successful attachment with *Striga* then resulted in less or negligible number of *Striga* emergence on the treated pots. These results also align with the experiment results as the fungus cause severe reduction of *Striga* seedlings, whereas most of *Striga* seedlings were healthy and vigorous on the untreated sorghum (Elzein et al., 2010; Hassan et al., 2009).

Previous studies (Julien et al., 2009; Marley et al., 2004; Yonli et al., 2004) suggested that the application of *F. oxysporum* significantly increased the sorghum seedling establishment and perform considerably better than the undressed genotypes. The effects of *Fusarium* application could be explained due to a combination of its direct and indirect effects on the growth of sorghum. The direct effect of *Fusarium* could be through reduction of *Striga* infestation. Whereas, indirectly either the sorghum might use the fungus growth regulating hormones as a bio-fertilizer or the disease that occurs on the *Striga* seedlings helps the crop to escape from its parasitic impact (Sugimoto et al., 2002). The present data demonstrate the use of *Fusarium* was able to reduce the number of *Striga* by 92.52% by attacking the parasite at its different growth stage before emergence as well as before flowering. Thus this contributes to the reduction of *Striga* seed bank with impact of enhancing future crop yield. The present study results agree with Julien et al., (2009) who reported the synergistic effects between the *Striga* resistant maize line and *Fusarium oxysporum f.sp Strigae* that led to over 90% reduction in *Striga* emergence.

Different studies (Hassan et al., 2009; Julien et al., 2009) reported combined use of resistant variety and the fungus that decreased the number of *Striga* plants. Julien et al., (2009) reported 98% control of *Striga* in maize fields using a resistant sorghum variety and use of Foxy 2 isolate of *Fusarium*. In this particular experiment, variation among varieties in reduction of *Striga* infestation due to *Fusarium* suggests the possibility of an integrated *Striga* management comprising host resistance together with *Fusarium* application. This would provide adequate level of *Striga* control.

Fen et al., (2007) pointed out the dynamism between biotic and abiotic environment of the rhizosphere in affecting the *Striga* parasitism and the efficacy and persistence of bio-control agents. Since microbial communities can have different impacts on the parasitism of cereals by *Striga*, the modification of microbial communities can improve parasitism. Among these biotic factors altering the different varieties of the host could have an effect on the differential response on the level of *Striga* management using *F. oxysporum*. Therefore, this experiment targeted at identifying genotypes which can be used in combination with the bio-agent and consequently as parent materials for further resistance breeding. The use of resistant varieties is cost effective and environmentally friendly. Biological control in conjunction with *Striga* resistance suppresses root parasitic weeds in annual crops where the intimate physiological relationship with their host plants makes it difficult to apply conventional weed control measures such as post emergent herbicides. This is observed in the current study where the use of *F. oxysporum* and host resistance was useful components that can be applied in the integrated *Striga* management in sorghum production.

3.5 Conclusion

The present study found a significantly reduced *Striga* count by 92% and identified 12 sorghum genotypes with suitable seed yield and agronomic traits under controlled studies with application of the bio-agent, *F. oxysporum.* These results illustrate the potential of an integrated management strategy that incorporates host plant resistance and biological control using *F. oxysporum* as an effective means of *Striga* control. Further breeding is required to develop sorghum varieties with *F. oxysporum* compatibility, *Striga* resistance and enhanced yield with farmers preferred traits.

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CHAPTER 4

Field evaluation of sorghum genotypes through *Fusarium oxysporum* application against *Striga hermontica*

4.1 Abstract

The parasitic weed, *Striga hermonthica*, inflicts significant yield losses in sorghum production. Integrated *Striga* management (ISM) has become one of the promising approaches for effective *Striga* management in sorghum. The objective of this study was to determine responses of sorghum genotypes using *Fusarium oxysporum* against *Striga* for ISM under field condition. Experiments involving 50 sorghum genotypes were conducted with and without a *F. oxysporum* f.sp.strigae seed dressing in Ethiopia in 2011. Data were collected on sorghum days to 50% emergence, flowering and maturity, plant height, panicle length, biomass, head weight, and grain yield, days taken to first *Striga* emergence, *Striga* count, and *Striga* plant height. There were significant differences among genotypes for *Fusarium* application across experiments affecting sorghum head weight, seed yield, days taken to first *Striga* emergence, *Striga* count and *Striga* plant height. There were significant genotype by site interactions (p<0.001) for all traits except days taken to 50% sorghum flowering and days to maturity. The main effect of *Fusarium* application was highly significant (p<0.001) across sites for all traits except days to flowering and sorghum plant height. Slight range of variations was detected across test environments without *Fusarium* application, including sorghum head weight and seed yield except for *Striga* number. The genotype and genotype by environment (GGE) biplot identified sorghum genotypes 235929, 2384443, 235921, 235761, 235924, 235926, IC9830, 235927, 237289, 243684, 235763, 238441, and 235466 as the best performing across all sites after *Fusarium* application. The three environments under *Fusarium* application were highly correlated and discriminated high performing genotypes adequately. The study demonstrated the effectiveness of *Fusarium* application with compatible sorghum genotypes inorder to suppress *Striga* infestation under field growing conditions which will be useful for ISM.

Key words: *F. oxysporum* f.sp.strigae, genotype, genotype by environment (GGE) biplot, integrated *Striga* management, sorghum, *Striga hermonthica*

4.2 Introduction

The hemiparasitic weed, *Striga* [*Striga hermonthica* (Del.) Benth], is a major biotic constraint to the production of sorghum [*Sorghum bicolour* (L.) Moench], maize (*Zea mays* L.), and millet [*Pennisetum americanum* (L.) K. Schum] in Sub Saharan Africa. *Striga* causes severe crop damage of course often leading to yield losses of 30% to 100%, depending on the level of infestation and susceptibility of the crop (Ast et al., 2005; Ejeta, 2007). In Ethiopia, sorghum is a major subsistence crop providing food and feed for smallholder farmers particularly in the low altitude, moisture stressed areas where continuous mono-cropping and low soil fertility are common features.

The complex biology of the parasite and its intimate physiological interaction with sorghum limits the development of practical control methods, especially for resource-poor subsistence farmers (Parker and Riches, 1993; Elzein and Kroschel, 2003). Farmers still depend on using traditional *Striga* control methods such as hand removal and hand hoeing. However, desired levels of control are seldom achieved by these traditional practices due to the prolific reproductive nature of the parasite, which generates huge numbers of minute seeds that can remain viable for over a decade (Odhiambo and Woomer, 2005). Also the parasite causes significant damage to the host before it emerges from the soil. Consequently, there is a continued effort towards developing integrated *Striga* management using a combination of control measures to reduce *Striga* infestations (Franke et al., 2006; Hearne, 2008).

Several approaches for controlling *Striga* have been reported. These include cultural practices such as hand weeding, crop rotation (Schulz et al., 2003; Oswald, 2005), trap cropping (Hess and Dodo, 2003), intercropping (Khan et al., 2008), improving soil fertility (Kim et al., 1997), and the use of resistant or tolerant crop varieties (Rodenburg et al., 2006; Grenier et al., 2007). Resistant cultivars could have a major impact in reducing *Striga* damage in sorghum but there is need to integrate this with farmers' preferred varieties and high yielding genotypes. Also adequate knowledge is required on the host-parasite interaction at the different growing stages of both plants, and the complex genotype by environment interaction (Haussmann et al., 2000). Since so far there are difficulties to attain complete *Striga* resistance through breeding (Gurney et al., 2002), it indicates the limitation of the presently available recommended control options.

Some isolates of *Fusarium oxysporum* f.sp. strigae show high pathogenic potential to attack *Striga* at its different growth stages, starting from seed germination (Abbasher et al., 1998; Ciotola et al., 2000). Biocontrol can contribute to enhanced crop yields and several research reports have recorded its efficacy in controlling *S. hermonthica* under controlled and field conditions (Abbasher et al., 1995; Ciotola et al., 2000; Marley and Shebayan, 2005; Yonli et al., 2005; Schaub et al., 2006; Sauerborn et al., 2007; Rebeka, 2007). Research in this area has advanced to the level of commercialization of *F. oxysporum* based mycoherbicidal products (Marley and Shebayan, 2005; Elzein and Kroschel, 2006). Further, the use of bio-control to complement host resistance has gained considerable support for its role as a component of integrated *Striga* management (Marley et al., 2004; Fen et al., 2007; Venne et al., 2009).

A preliminary study conducted in Ethiopia under controlled conditions revealed the potential of the synergistic effect that the biocontrol agent (*Fusarium oxysporum*) and compatible sorghum genotypes, have to control *Striga* (Rebeka et al., 2013). However, these results need to be verified under natural growing conditions for further development and utilization of the technology by sorghum growers. The objective of this study was to determine field responses of 50 sorghum genotypes using *Fusarium oxysporum* against *Striga* for ISM.

4.3 Materials and methods

4.3.1 Study sites

Field experiments were conducted at three sites, namely, in a farmer's field in Shewa Robit, in Sirinka and Kobo on research stations during the main cropping season in Ethiopia (June-November) in 2011. The sites are located along the Rift Valley escarpments in northeastern region of Ethiopia at the geographical location of latitude 10° 00' and 12°30' North and longitude 39°30' and 40°00' East. The Kobo research site is situated at an altitude of 1400 meters above sea level (masl). It receives a mean annual rainfall of approximately 650mm with the length of growing period 60-120 days. Shewarobit is found at an altitude of 1300 masl and its annual average rainfall is 1023.8 mm with the length of growing period of 108-138 days. The predominant mode of agricultural production is subsistence, smallholder, mixed crop andlivestock system. At these sites sorghum and *Tef* (*Eragrostis tef*) are the major crops grown under rainfed conditions with minimal external inputs such as fertilizer. Drought, *Striga* infestation, pests and low soil fertility are the major constraints for sorghum production in the region. The experimental sites represent typical sorghum growing dry land agro-ecologies in Ethiopia and have been characterized as 'hot-spots' for *Striga* infestation.

4.3.2 Sorghum genotypes

Fifty sorghum genotypes were used in this study. The genotypes were sourced from IBC-Ethiopia (the Institute of Biodiversity Conservation of Ethiopia) and ICRISAT (the International Crops Research Institute for the Semi-Arid Tropics). The 50 genotypes were selected for their adaptability to the specific environment of the study area as well as on their variable levels of resistance and tolerance to *Striga*.

4.3.3 *Fusarium oxysporum* isolate

The isolate of *Fusarium oxysporum* used in this study was collected in Ethiopia from severely diseased *Striga hermonthica* plants by Rebeka (2007). Taxonomic identification was confirmed by the Department of Phytomedicine, Humboldt University of Berlin, Germany. The isolate was used for mass production of chlamydospores in powder at the University of KwaZulu-Natal, South Africa. The biocontrol agent inocula consisting of a large amount of chlamydospores were used to treat seeds of the sorghum genotypes. For seed coating, 0.216 g of dried fungal chlamydospores was used to coat 4.5 g of sorghum seed. Seed coating technique has been found to be efficient and facilitates uniform incorporation of fungal chlamydospores on to the seeds (Elzein et al., 2010).

4.3.4 Experimental design and management

Field trials were arranged in a randomized complete blocks design with three replications per site. Treatments were constituted in factorial combinations of 50 sorghum genotypes, with or without *Fusarium oxysporum* application. Sorghum seeds were planted in 2 rows of 3 meters length each with a space of 0.75 meters between rows and 0.30 meters between plants. The experiments in Shewa Robit and Sirinka were undertaken under natural *Striga* infestation using farmers' fields while in Kobo a *Striga* sickplot field was used. Pre-conditioned *Striga hermonthica* seeds were used to provide supplementary infestation in Kobo, which were collected in the previous cropping season from the surrounding sorghum fields. Diammonium phosphate and urea fertilizers were applied at the recommended rates of 46 kg P_2O_5 ha^{-1} and 54 kg nitrogen ha^{-1}, respectively. The plots were hand weeded as frequently as needed without affecting *Striga* development.

Data were recorded on the agronomic performances of sorghum and on the growth of *Striga.* For all sorghum genotypes, five randomly selected plants were used to measure sorghum plant height, panicle length, biomass, head weight and grain yield. Data on days taken for 50% plant emergence, flowering and maturity were recorded on whole plot basis. The time of emergence of the first *Striga* shoots in the plot, number of *Striga* plants per plot and their plant height were also recorded.

4.3.5 Data analysis

Data collected were subjected to analysis using the general linear model (GLM) procedure of the Statistical Analysis Systems statistical package (SAS 9.1, 2003). Data were tested for normality and homogeneity of variances prior to analysis. Data on numbers of *Striga* plants counted per plot violated the assumption of homogeneity of variances so they were subjected to logarithmic transformation using the function $Y = \log(X+1)$, where X represents the initial *Striga* count data. The fixed effects of block, site, genotype, application of *Fusarium oxysporum* and their 2-way and 3-way interactions were fitted into the statistical model. The significant differences between means were tested by the Tukey’s multiple comparison procedure at a 5% level of significance.

The genotype and genotype by environment (GGE) biplot analysis of the Breeding View program of Genstat (BV 1.1) was used to visualize the relative yield performance of sorghum genotypes over the different environments. For the GGE biplot analysis, the factorial combinations of the 3 levels of the site with 2 levels of *Fusarium oxysporum* application were considered to represent six separate environments.

4.4 Results and discussion

4.4.1 Performance for sorghum agronomic traits

Mean squares of the combined analysis of variance for sorghum traits are presented in Table 4.1 All sorghum agronomic traits measured in this study were significantly affected by site, genotype, and site x genotype interaction (P < 0.001). The interaction between *Fusarium* treatment and site had significant effects on most sorghum traits except plant height (P < 0.001). The box plots in Figure 4.1 (a), (b) and (c) show a larger range of variation of some sorghum genotypes performances following *Fusarium* application compared to untreated plots across sites. *Fusarium* application at Kobo reduced the days to sorghum flowering marginally (6%) but not at Shewa Robit and Sirinka. This implies that at Kobo *Fusarium* application has slightly contributed for earliness of the genotypes as compared to untreated controls. Head weight varied due to *Fusarium* application between 25 to 104% and seed yield between 33 to 146% at the three sites.

Sorghum head weight and seed yield were significantly influenced by the interaction between *Fusarium* treatment and sorghum genotype (P < 0.001), and also by the interaction of site x genotype x *Fusarium* treatment (P < 0.05). The bar charts in Figure 4.2 summarize mean head weight and seed yield performances of the fifty sorghum genotypes, with or without *Fusarium* application, across sites. Both head weight and seed yield increased significantly in most sorghum genotypes due to *Fusarium* application and both traits showed a similar trend of variation reflecting significant correlation (r= = 0.94; P < 0.001).

Field evaluation of sorghum genotypes for compatibility to *F. oxysporum* application against *S. hermonthica* resulted in promising outcomes for the use of the biocontrol in ISM. From a previous study undertaken in the greenhouse and laboratory tests using similar sorghum genotypes, *Fusarium* compatible and phenotypically superior candidate genotypes were selected for breeding (Rebeka et al., 2013). The present field experiments confirmed the preliminary studies showing the beneficial effects of *Fusarium* application and compatible sorghum genotypes on suppressing the *Striga* infestations under field conditions. Treatments resulted in significant improvement of most of the sorghum growth and yield across the three study sites. Previous research findings reported similar achievements on the improvement of sorghum growth when *F. oxysporum* was used as the bio-agent for the control of *Striga* (Ciotola et al., 2000; Schaub, et al., 2006; Venne et al., 2009). The *Fusarium* treatment had remarkable positive effect on the earliness of sorghum genotypes. Similar observations were made by previous workers (Marley and Shebayan, 2001; Venne et al., 2009; Elzein, et al., 2010) who also reported the growth promoting effect of *Fusarium* species treatments on sorghum. This was manifested at the Kobo site of the current study. Earliness is a desirable attribute in drought stressed areas where *Striga* exerts its devastating and pronounced effect. Overall, the use of *Fusarium* combined with *Fusarium* compatible sorghum genotypes helped to suppress *Striga* infestation and presumably contributed to the enhanced growth in the treated sorghum allowing it to escape the late season drought stress which would otherwise have led to serious yield reduction.

Table 4.1 Analysis of variance for sorghum agronomic traits measured across three environments

Sources of variation	DF	Days to emergency	Days to flowering	Days to maturity	Plant height (cm)	Panicle length (cm)	Biomass (t ha^{-1})	Head weight (t ha^{-1})	Seed yield (t ha^{-1})
Replication(nested within site)	2	0.103	131.08	212.77	21402.90	304.89***	207.98*	1.12	0.91
Site (S)	2	5268.643***	1958.93***	17591.43***	84115.76***	770.41***	8962.98***	41.57***	16.95***
Genotype (G)	49	1.618***	223.27***	446.20***	11560.99***	89.14***	124.87***	20.75***	14.45***
Fusarium (F)	1	4.134*	579.51**	247.81	667.20	1.44	5658.49***	674.09***	481.24***
S x G	98	1.489***	108.06***	191.79***	3889.41***	35.72***	115.08***	7.64***	5.09***
S x F	2	6.774***	471.56***	989.32***	723.70	121.45***	2362.52***	55.71***	43.01***
G x F	49	0.604	68.17	99.77	701.61	8.62	45.94	21.63***	15.01***
S x G x F	98	0.659	64.67	62.90	776.92	8.41	37.18	3.37*	1.96*
Error	598	0.613	39697.68	83.27	932.53	11.65	57.11	2.51	1.39
R-square (%)		96.70	48.32	64.91	70.11	63.27	60.37	72.83	76.66
CV (%)		8.69	10.66	7.40	16.76	15.68	45.23	38.25	42.09

DF = Degrees of freedom.

*, **, *** = Significant at 5%, 1% and 0.1% level, respectively.

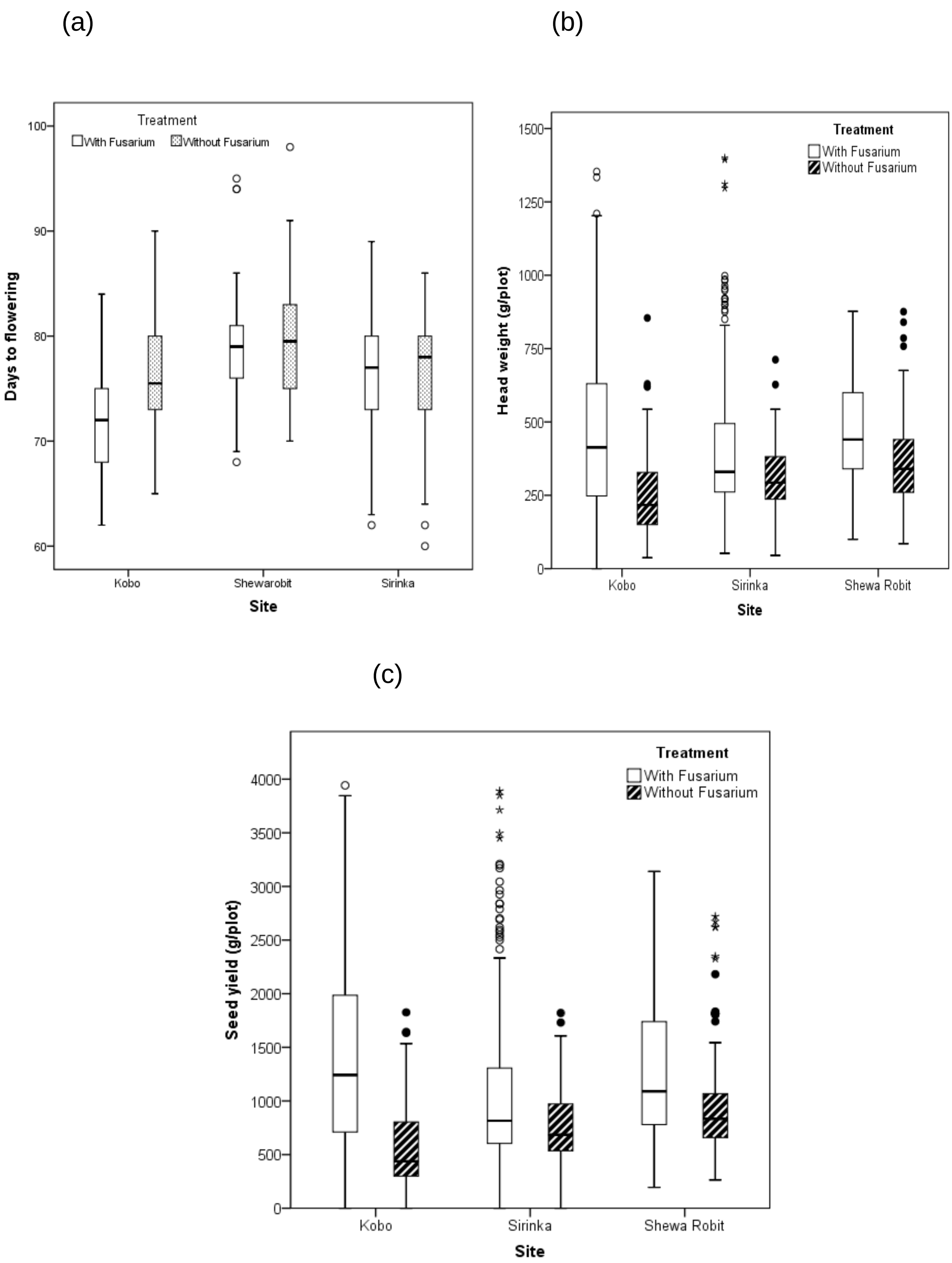
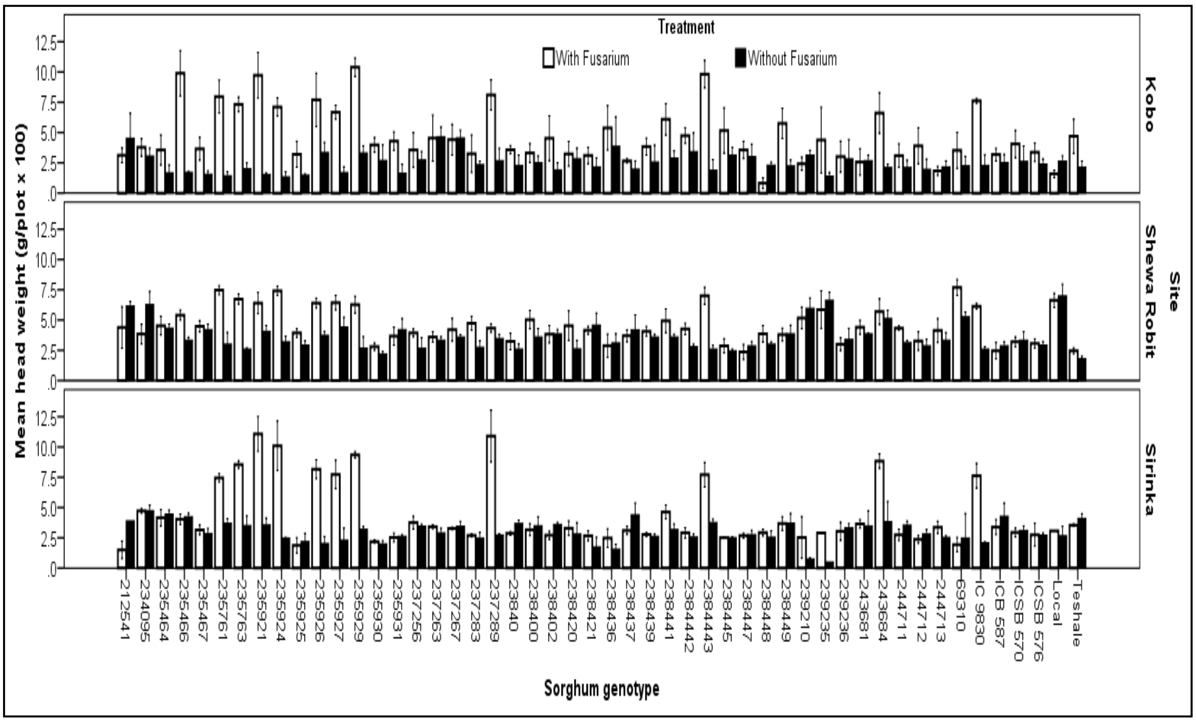


Figure 4.1 Box plots for days to flowering (a), head weight (b) and seed yield (c) of 50 sorghum genotypes tested, with or without *Fusarium* seed treatments, at three sites in Ethiopia

(a)



(b)

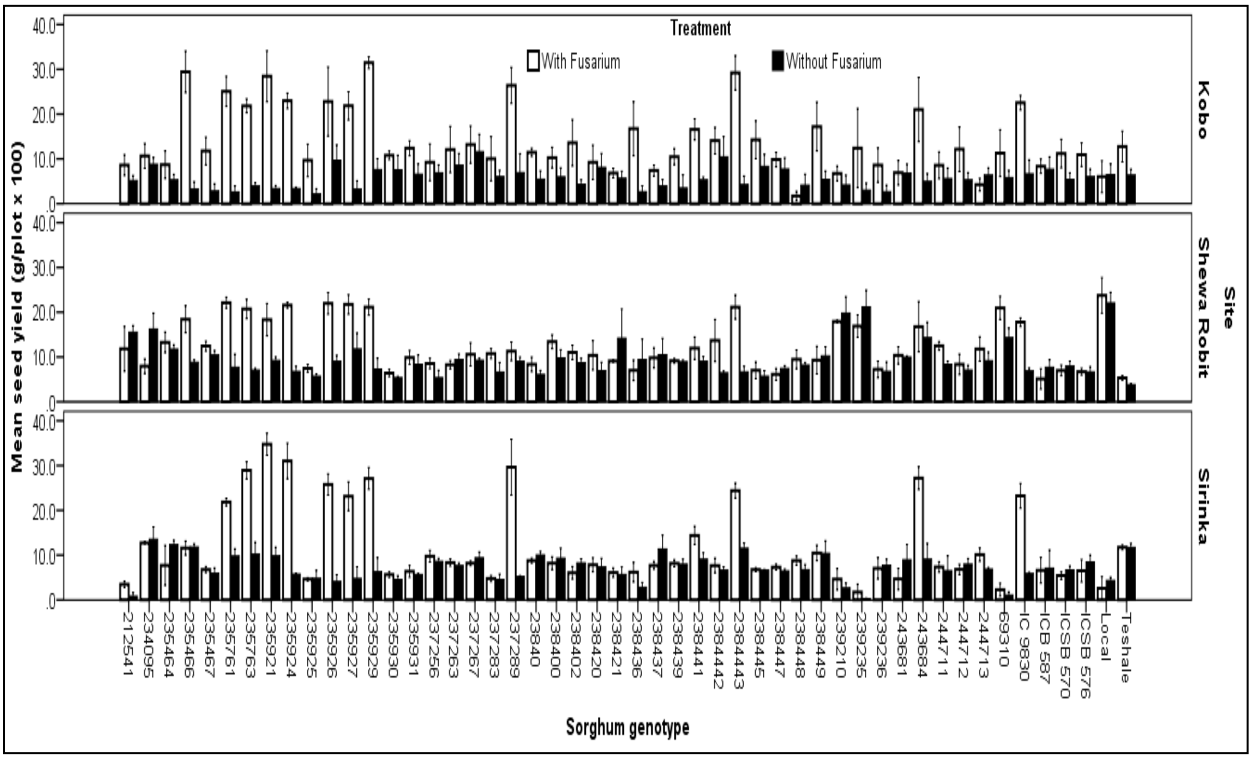


Figure 4.2 Mean head weight (a) and seed yield (b) of fifty sorghum genotypes tested with or without *Fusarium* seed treatments at three sites in Ethiopia

4.4.2 Differences in *Striga* parameters

Analysis of variance (Table 4.2) showed that time of *Striga* emergence, number of *Striga* plants/plot and *Striga* plant height were significantly affected by the main effects of site, genotype and *Fusarium* treatment (P <0.01). On sorghum genotypes treated with *F. oxysporum* days to *Striga* emergence were decreased with mean (± standard error) of 16.2±1.70 days in treated plots compared to 35.6±1.76 days in untreated plots. *Fusarium* application reduced mean numbers of *Striga* plants (0.50±0.07 versus 3.14±0.26 plants/plot). Growth of *Striga* plants was retarded, with a mean plant height of 3.80±0.47 cm compared to 11.95±0.66 cm in untreated plots. Furthermore, variations in all *Striga* parameters considered in the study were significantly affected by site x genotype and site x *Fusarium* interactions (P <0.001), but not by genotype x *Fusarium* treatment interaction. Time of *Striga* emergence (P <0.05) delay and *Striga* plant height (P <0.01) reduction were significantly affected by the effects of site x genotype x *Fusarium* treatment interaction. The significant interaction effects observed in the study could be attributed to the high heterogeneity of the level of *Striga* infestation both between sites and between plots within sites at the beginning of the experiment. Means and dispersion measurements of *Striga* parameters in *Fusarium* treated and untreated plots across the three sites are summarized in Table 4.3.

Table 4.2 Combined analysis of variance for *Striga* parameters across sites

Sources of variation	DF	Days to *Striga* emergence	Number of *Striga*/plot	*Striga* plant height
Replication(nested within site)	2	17170.99***	0.97***	1089.02***
Site (S)	2	6797.73**	2.51***	1340.16***
Genotype (G)	49	2240.39***	0.15***	241.04***
Fusarium (F)	1	84448.36***	17.38***	14983.90***
S x G	98	1788.75***	0.14***	186.95***
S x F	2	42796.84***	5.69***	4818.46***
G x F	49	938.48	0.09	124.71
S x G x F	98	1326.45*	0.10	162.10**
Error	598	1026.53	0.08	110.64
R-square (%)		52.52	59.33	55.27
CV (%)		123.82	123.59	133.50

1= Nested effect of replication within each site

Table 4.3 Means, standard deviations (SD) and ranges of *Striga* measurements with (WF) and without *Fusarium* (WOF) across sites (N = 150 plots per treatment within site)

Variable	Statistic	Kobo		Shewa Robit		Sirinka	
		WF	WOF	WF	WOF	WF	WOF
Days to *Striga* emergence	Mean	21.13	39.09	4.83	48.76	22.61	18.83
	SD	45.21	37.74	19.30	32.96	36.42	34.52
	Range	0-165	0-120	0-95	0-95	0-120	0-128
Number of *Striga*/plot	Mean	0.41	3.69	0.37	5.21	0.72	0.51
	SD	1.01	4.83	1.85	7.44	1.60	1.14
	Range	0-5	0-23	0-14	0-35	0-9	0-7
Striga plant height	Mean	1.92	17.72	1.17	10.08	8.33	8.05
	SD	5.16	15.84	5.55	8.99	14.33	14.64
	Range	0-33	0-43	0-43	0-37	0-61	0-52

Seed treatment of the host crop with the biocontrol offers a simple and easy delivery system to attack *Striga* effectively (Elzein et al., 2006). Use of Striga resistant sorghum genotypes combined with a pathogenic strain of *Fusarium* isolate was able to achieve up to 100% reduction of *Striga* emergence using different compatible sorghum genotypes. The biocontrol agent has to be applied in the root zone of the crop in order to be in close proximity to easily penetrate and disintegrate *Striga* seeds (Saureborn, 1996; Elzein et al., 2010), and to subsequently prevent the *Striga* attaching to sorghum roots. This leads to disruption of parasitic growth of the weed and its inability to emerge following *Fusarium* treatment. Similar achievements using *Fusarium* isolates as a biocontrol agent have been reported by various authors, from both laboratory and field studies. These reports have indicated that *Striga* emergence was reduced by more than 90% (Ciotola et al., 2000; Marley and Shebayan, 2005; Venne et al., 2009; Rebeka et al., 2013). In addition to the reduction of the *Striga* number and its emergence, it was also observed that on *F. oxysporum* induced disease on emerged *Striga* plants, which died before flowering. The current results are in agreement with other similar studies done elsewhere in Africa (Schaub et al., 2006; Zahran et al., 2008) which found that the combination of

F. oxysporum with *Striga* resistant sorghum genotypes provided superior control of *Striga*.

Figure 4.3 displays plots of sorghum genotypes on the axes as defined by their response to *Fusarium* application in seed yield performance and reduction of numbers of *Striga* per plot, fitted between –1 and +1. The response to *Fusarium* application showed wide variation among sorghum genotypes, both within and between sites. The relative differences in seed yield between *Fusarium* treated and untreated sorghum genotypes ranged from -58% to 857% at Kobo, -51% to 220% at Shewa Robit and -48% to 835% at Sirinka. Overall 22 sorghum genotypes that were showed responsiveness values equal to or above +0.5 to *Fusarium* application (the right side of the dashed line in Figure 4.3 perpendicular to the seed yield axis) were identified as the most compatible with *F. oxysporium*. These genotypes included 235763, 235924, and 235929 at all three sites; 235921, 235927, 237289, 239235, 243684, and IC9830 at the Kobo and Sirinka sites; and genotype 235761 and 2384443 at the Kobo and Shewa Robit sites. Genotypes 235466, 235467, 235925, 238402, 238436, 238439, 238441, 238449, 239236 showed higher responses in Kobo site only and those that showed response at the Sirinka site only were 212541 and 235926. The mean seed yield of the 22 genotypes with *Fusarium* application was about 3.8 times greater than that obtained without *Fusarium* application (mean ± SD of 5.87 ± 2.22 and 1.38 ± 0.66 t/ha, respectively). Reduction in *Striga* count due to *Fusarium* application ranged from 33 to 100% at Kobo and Shewa Robit sites, while at Sirinka the effect of *Fusarium* was highly expressed not directly through *Striga* count reduction but through the sorghum growth performance improvement. This might be the *Fusarium* effect reflected by its killing the *Striga* seeds before emergence.

The magnitude and direction of response expressed by various genotypes to *Fusarium* application indicated variations among genotypes in their level of compatibility with the biocontrol agent in different environments. The synergistic effect of host resistance and *Fusarium* application was expressed through drastic reductions in *Striga* counts after *Fusarium* treatment, and sorghum grain yield improvement because of the absence of *Striga* infestation. This tritrophic interactive effect between the *Fusarium* compatible sorghum genotypes, *F. oxysporum* and *S.*

hermonthica clearly indicate their potential in an integrated *Striga* management approach.

The destructive effects of *Striga* start before its emergence while it is underground. *Fusarium* is able to attack, all stages of the Striga life cycle, from *Striga* seed all the way to the mature plant (Abbasher et al., 1998; Ciotola et al., 2000; Fen et al., 2007; Venne et al., 2009). At the three study sites, for instance at Sirinka, the number of *Striga* count was not as expected. However the yield differences between treatments with *Fusarium* and without *Fusarium* were highly significant. This indicates the influence of *Fusarium* through sorghum growth and grain yield improvement as compared to without *Fusarium* treatment.

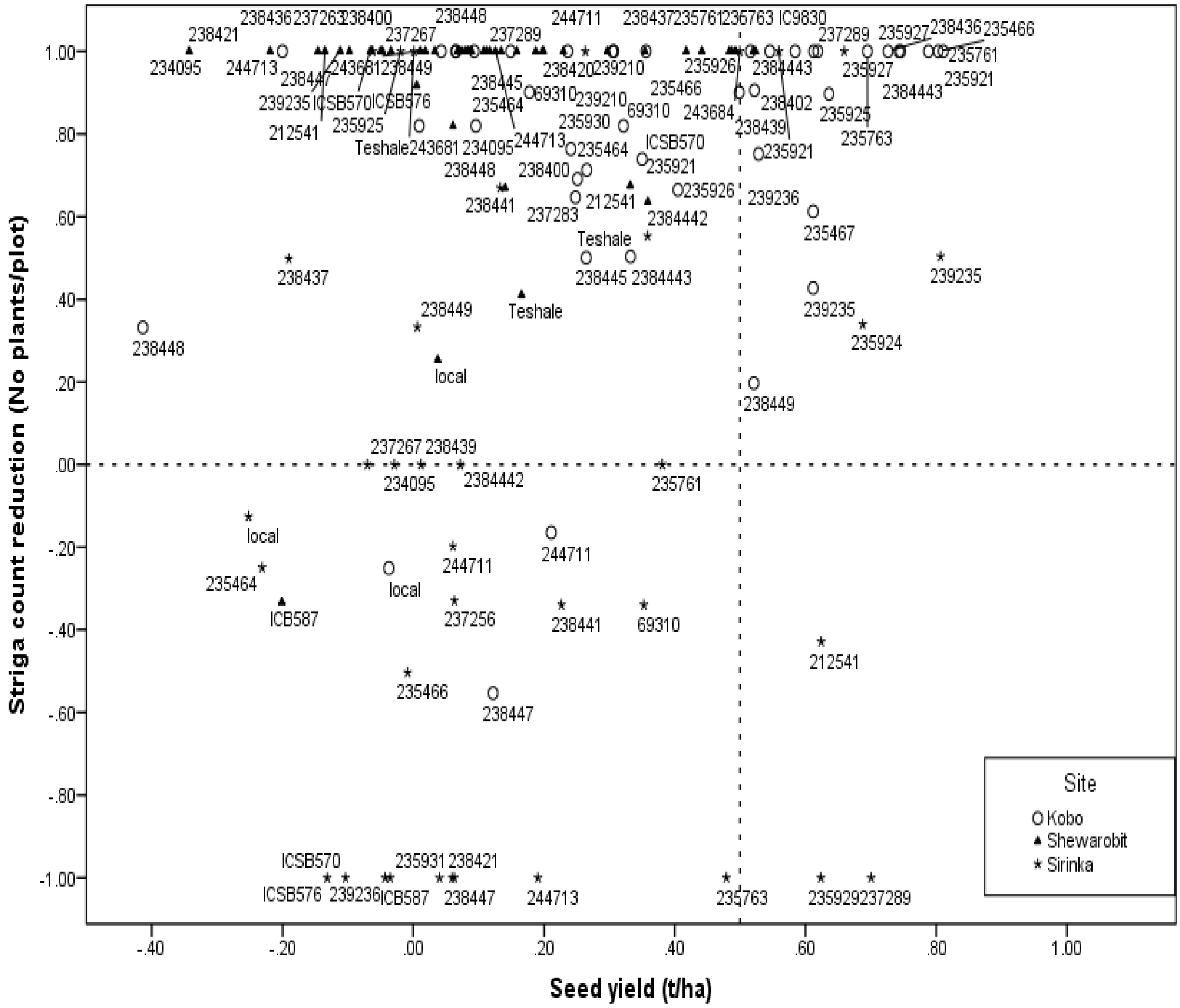


Figure 4.3 Plot of 50 sorghum genotypes showing seed yield and reduction in *Striga* count across sites in Ethiopia after seed treatment with *Fusarium oxysporum* f.sp. *Strigae*

Note: In order to have values for *Fusarium* responsiveness fitted between –1 and +1, the following formula was used: (seed yield with *Fusarium* – seed yield without *Fusarium*)/(seed yield with *Fusarium* + seed yield without *Fusarium*). A similar calculation was done for the *Striga* plants count data (Lendzemo, 2004).

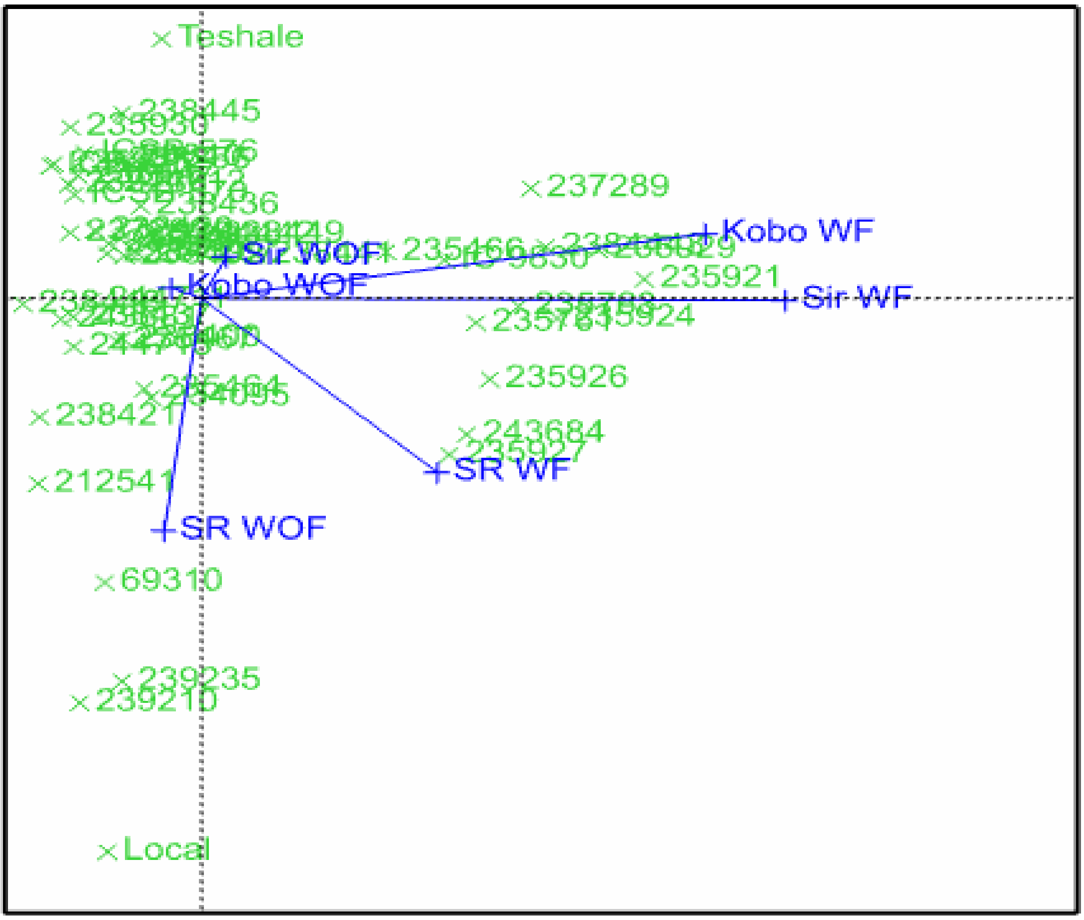
4.4.3 GGE biplot analysis

The GGE biplot (Figure 4.4) displays the seed yield performance of the 50 sorghum genotypes in each of the six environments (3 sites x 2 levels of *Fusarium*). The two principal components of the biplot for sorghum seed yield together explained 86.61% (72.56% and 14.05% by PC1 and PC2, respectively) of the total variation of the GGE. The cosine of the angle between the vectors of the sites with *Fusarium* application was less than 90^o, indicating a positive correlation between *Fusarium* treated sites, while the wide angle (greater than 90^o) between Kobo with *Fusarium* and Shewa Robit without *Fusarium* indicates a negative correlation between the two environments, implying the presence of crossover GE, and further, that the wide distance between the two environments expresses their dissimilarity in discriminating between genotypes as explained by Ding et al. (2007). This type of GE interaction which leads to changes in the ranking of the genotypes is commonly known as a crossover interaction. For instance, Sirinka and Kobo with *Fusarium* application had close proximity, implying that these environments exhibited similar patterns of discriminating between the genotypes, while the Kobo and Sirinka sites without *Fusarium* application were non-discriminating.

With *Fusarium* application, the sorghum genotypes 235929, 2384443, 235921, 235761, 235924, 235926, IC9830, 235927, 237289, 243684, 235763, 238441, and 235466 were the best performing across all sites, with the highest mean seed yields of 4.00 to 9.66 tons per hectare. At the same time, sorghum genotypes local (Shewa Robit), 239210, 239235, 69310, 212541, and 238421 showed a reasonable level of adaptation to the Shewa Robit environment with no *Fusarium* application with mean seed yields of 3.95 of 6.13 tons per hectare. The rest of the genotypes did not show explicit adaptation patterns and hence they might not be compatible with *Fusarium* application to reduce *Striga* infestation. This reveals that the best genotypes manifested remarkable compatibility to the biocontrol agent and expressed specific adaptation to the local environment. Therefore, Fusarium application through sorghum seed dressings could be exploited as a key component of integrated *Striga* management to improve sorghum crop productivity in Ethiopia.

The level of *Striga* infestation, the compatibility of the sorghum genotypes, the soil fertility level, and the unique interaction of the biocontrol agent with other environmental factors could lead to complex genotype by environments interactions across the six different growing environments. Similarly the difficulties of the genotypes by environment interactions in *Striga* management studies on sorghum have been reported (Haussmann et al., 2000).

PC2-14.05%



PC1-72.56%

Figure 4.4 GGE biplot for seed yield of 50 sorghum genotypes tested in six environments: Note three sites [Kobo, Shewa Robit and Sirinka] and two levels of *Fusarium* application [with (WF) and without (WOF)]

4.5 Conclusion

The present study demonstrated the synergistic effect of a combined use of *Fusarium oxysporum* and compatible sorghum genotypes. This was expressed by reduction of *Striga* infestation, together with an improved crop stand that led to increased seed yield, suggesting the potential of this approach in integrated *Striga* management. The field tests confirmed clear differences between the performances of sorghum genotypes when treated or untreated with *Fusarium oxysporum*. Significant variation was detected among treated genotypes. This variation could be attributed to either genotypic differences in compatibility with the biocontrol agent, or the genotypes might be very susceptible to *Striga.* Regarding the first hypothesis, the approach of combining host plant resistance and the use of *Fusarium* is a good option for farmers and should be tested further. Indeed, it was important to choose the best compatible genotypes for further breeding activity to improve their resistance levels. Also, it is important to choose the best compatible genotypes for environments with *Fusarium* treatment for every location because its performance is often affected by the complicated interaction between resistance and agro-ecological conditions.

Among the 50 genotypes tested the top genotypes performed well with *Fusarium* application, and may be suitable for different agro-ecologies. Genotypes, including 235763, 235924, and 235929 gave highly compatible reactions at all study sites so they should be used in future integrated *Striga* resistance breeding programmes. The fact that there were genotypes that showed *Fusarium oxysporum* compatiblity and better agronomic performances specifically for each site reflects the need for site specific breeding with narrow adaptation. Generally, genotypes which were selected for their *Fusarium oxysporum* compatible and better agronomic performances from the previous controlled environment studies such as 235763, 235927, 237289, 235466, IC9830, 235924, 235921, 2384443,235929,243684,235761, and 235926 also performed well in the present study. This suggests that they could be promising candidates for imeediate deployment to farmers in *Striga* prone areas, and for use in the breeding of even better sorghum varieties.

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CHAPTER 5

Assessment of genetic diversity of sorghum resistant to*Striga hermonthica* and compatibile with *Fusarium oxysporum* using phenotypic and SSR markers

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5.1 Abstract

Assessment of genotypic variation using phenotypic and molecular markers is a fundamental step in plant breeding programmes. Sorghum [*Sorghum bicolour* (L.) Moench] is one of the most widely domesticated crops in arid, semi-arid and marginal agro-ecologies owing to its remarkable environmental adaptability. This study aimed to determine the variability present among 14 selected sorghum genotypes exhibiting *Striga hermonthica* resistance, and compatibility with a biological control agent, *Fusarium oxysporum,* using phenotypic and simple sequence repeat (SSR) markers. Genotypes were assessed using nine phenotypic traits under field experiment at Kobo in Ethiopia during 2011/12 growing season and using 20 selected polymorphic SSR DNA primers in South Africa. The analysis of variance for the phenotypic traits revealed highly significant ($p<0.001$) differences among genotypes. Principal component analysis on these traits showed three components that accounted for 73.99% of the total variability exhibited among genotypes. Cluster analysis allocated two major groups, one with a further two subgroups based on morphological traits, showing clear demarcations between the genotypes. The SSR markers revealed a high level of polymorphisms among genotypes, with average alleles per locus of 6.95 and average polymorphism information content at 0.80. The observed genetic diversity was relatively wide with the allele sizes ranging from 203.6-334 bp. The SSR markers allocated genotypes into two distinct clusters similar to phenotypic markers. In one of the clusters, two sub-clusters were distinguished, representing *Fusarium* compatible or *Striga* resistant genotypes. On the basis of the exhibited phenotypic and genotypic variations, genotypes were selected as promising parents for further breeding in the integrated management of *Striga hermonthica*, combining resistance and bocontrol.

Key words: Genetic diversity, polymorphic information content, biocontrol

5.2 Introduction

Genetic diversity is essential in plant breeding programs to develop crop cultivars with improved genetic constitution to serve diverse human needs. Genetic diversity encompasses variations in nucleotides, genes, chromosomes or whole genomes of individuals (Wang et al., 2009). The most common sources of genetic diversity include landraces, modern cultivars, obsolete or primitive cultivars, wild or semi-wild related species (Acquaah, 2012; Shimelis and Laing, 2012). Genetic analyses help to determine the variations present among individuals, populations or groups of genetic resources for breeding and strategic conservation.

Genetic markers including phenotypic, protein (biochemical) or DNA (molecular) markers help to identify characteristics of the phenotype and/or genotype of individuals. Uses of phenotypic characteristics are a common and traditional approach because they form the most direct measure of the phenotype, readily available, relatively cheap to evaluate and requiring simple equipment (Harlan and DeWet, 1972). However, phenotypic markers are subject to environmental influences in the field that may mask the underlying genetic variation among genotypes. DNA based molecular markers are efficient for the analysis of large numbers of genotypes (Melchinger and Gumber, 1998; Reif et al., 2003). The combined use of phenotypic and molecular markers allows for estimation of genetic diversity more reliably and efficiently. Combined, they provide useful information for breeders to select appropriate parents for efficient breeding, and to conserve novel genetic resources.

Various molecular markers are available for genetic analysis such as restriction fragment length polymorphisms (RFLPs) (Smith et al., 1997; Perumal et al., 2007), random amplification of polymorphic DNAs (RAPD) (Agrama and Tuinstra, 2003), amplified fragment length polymorphisms (AFLP) (Perumal et al., 2007), microsatellites or simple sequence repeats (SSRs) (Ganapathy et al., 2012) and single nucleotide polymorphisms (SNPs) (Arai-kichise et al., 2011). SSRs are clusters of short tandem repeated nucleotides bases distributed throughout the genome. SSRs markers have been developed for major crop plants and used in characterization, genetic diversity analysis, chromosome locations of desired genes and marker-assisted breeding (Smith et al., 1997; Ghebru et al., 2002). Various studies have reported combined use phenotypic and molecular markers in genetic analyses of cereals such as ryegrass (Jianyang, 2005), rice (Ogunbayo et al., 2005), maize (Beyene et al., 2005; Wende et al., 2012), and sorghum (Agrama and Tuinstra, 2003; Anas and Tomohiko, 2004; Bucheyeki et al., 2009).

Sorghum [*Sorghum bicolour* (L.) Moench] is a crop of considerable economic importance in many countries of the semi-arid and arid tropical areas (Doggett, 1988). Genetic admixtures, introgression of genes or hybridization events between wild and cultivated species probably lead to genetic diversity in sorghum. Other possible accounts of sorghum genetic diversity may include: thousands of years of selection in response to diverse physical environments and human needs, genetic drift, and natural inter-crossing among the different sorghum races. This has resulted in wide-area adaption and diverse production practises of the crop in varied agro-ecologies (Vavilov, 1951; Stemler et al., 1975; Doggett, 1988). Sorghum shows remarkable genetic diversity with more than 22,000 accessions systematically conserved in the world sorghum collection in India (Kimber, 2000).

In Ethiopia, sorghum is one of the major staple food crops. In addition to its being a staple food crop, sorghum serves as livestock feed, as the basis of traditional beverages, cooking fuel, and construction material. Its wide adaptation to stress environments makes sorghum a crop of choice by millions of farmers in sub-Saharan Africa (CSA, 2011). Sorghum predominantly grows in lowland areas where eratic rainfall, different biotic and abiotic production constraints prevail, which are often the cause of failure of other cereal crops. Natural and artificial selection pressures have played significant roles in sorghum's tolerance of harsh growing environments and its relative tolerance to biotic and abiotic production constraints. Drought and *Striga* [*Striga hermonthica* (Delile) Benth.] are the most common production constraints of sorghum in sub-Saharan Africa (Watson et al., 2007). There is substantial need for sorghum breeding to enhance drought tolerance and *Striga* resistance.

Biological control of *S. hermonthica* using a selected sorghum-compatible strain of *Fusarium oxysporum* f.sp. *strigae,*(*F. oxysporum*) is a control option that can be integrated with other control measures (Elzein et al., 2006; Sauerborn et al., 2007; Rebeka et al., 2013). It is necessary to find the sorghum varieties that have superior *Striga* resistance, and better compatibility with *F. oxysporum* which can be used to suppress *Striga* for the development of integrated *Striga* management. This study aimed to determine the variability present among 14 selected sorghum genotypes exhibiting *Striga hermonthica* resistance, and compatibility with the biological control agent, *F. oxysporum,* using phenotypic and simple sequence repeat (SSR) markers. Results of the study may assist in identifying suitable parents for sorghum breeding in the integrated management of *Striga*.

5.3 Materials and methods

5.3.1 Phenotypic evaluations

Plant material, study site and field planting

The study used 14 sorghum genotypes for both phenotypic and genotypic characterisation (Table 5.1). Ten genotypes were selected with excellent compatibility with *F. oxysporum* for *Striga* management. Landraces were obtained from the Institute of Biodiversity Centre (IBC)/Ethiopia and one from the International Crop Research Institute for the Semi-Arid Tropics (ICRISAT) in India. These genotypes have better agronomic performances and acceptability by famers (Rebeka et al., 2013). The other four genotypes included in the study are known to have a high level of *Striga* resistance, of which two were obtained from ICRISAT and two from sorghum improvement program of Ethiopia (Table 5.1). The test genotypes were homogenous after continued selfing and selection.

The sorghum genotypes were grown at the Kobo Research Sub-Centre in the North Wello Administrative Zone, Ethiopia during 2011/12 growing season. Kobo is situated at E39^{o}37', and N12^{o}09', and represents the dry lowland areas, with a high *Striga* infestation. The experiment was laid out in a randomized complete block design with three replications. The plot size was two rows of 3 m length. Plants were established with 300 mm intra-row and 750 mm inter-row spacing. Fertilizer was applied at the rate of 100 kg ha^{-1} Diammonium phosphate (DAP) and 50 kg ha^{-1} urea as recommended for sorghum in the lowlands of Ethiopia. All the DAP was applied at the time of planting, while urea was applied in a split application. Other agronomic practices were applied uniformly to all the treatments.

Table 5.1Source of sorghum genotypes used in this study

No.	Genotypes	Source	No.	Genotypes	Source
1	235466	IBC-landrace	8	237289	IBC-landrace
2	235761	IBC-landrace	9	2384443	IBC-landrace
3	235763	IBC-landrace	10	IC9830	ICRISAT
4	235921	IBC-landrace	11	Birhan	SARC
5	235924	IBC-landrace	12	Hormat	SARC
6	235926	IBC-landrace	13	N13	ICRISAT
7	235929	IBC-landrace	14	SRN39	ICRISAT

IBC= Institute of Biodiversity Centre /Ethiopia, ICRISAT= International Crops Research Institute for the Semi-Arid Tropics /India, SARC= Sirinka Agricultural Research Centre/Ethiopia

5.3.2 Phenotypic data collection and analysis

Nine important phenotypic traits with production and acceptability issues were collected to phenotype the sorghum genotypes. Data collected included: the number of days to 50% emergence, days to 50% flowering, days to 50% maturity, plant height (in mm) at 50% flowering, panicle length (in mm) during harvest, fresh biomass (gm/plant), panicle weight (gm/plant), seed yield (gm/plant) and the harvest index (the ratio of yield to total biomass).

All the phenotypic data collected from the field evaluation were subjected to analysis of variance to test for significant differences between genotypes using Genstat version 14 (Payne, et al., 2011). Further cluster analysis was conducted using SPSS 15.0 statistical software (SPSS Inc., 2005). The Euclidean distances were used to establish the pattern of phenotypic groupings and to compare with genotypic clustering using the SSR primers. Principal component analysis was also employed to group phenotypic traits based on the variation between genotypes.

5.3.3 SSR analysis

Sampling and DNA extraction

The 14 genotypes listed in Table 5.1 were grown in a greenhouse at University of KwaZulu-Natal, South Africa. Samples were taken from each genotype, and analysed at the Incotec laboratory in South Africa (Incotec SA Pty Ltd., South Africa). Young fresh leaves were sampled and used in bulked amplifications using 10 plants per genotype. Twenty selected and highly polymorphic simple sequence repeat (SSR) markers were used for genotyping. The markers used were selected based on the size of the repetitions and their locations, in order to obtain a representative sampling of the whole genome (Table 5.5). PCR products were fluorescently labelled and separated by capillary electrophoresis on an ABI 3130 automatic sequencer (Applied Biosystems, Johannesburg, South Africa). PCR was done for all of the 20 primers.

5.3.4 SSR data analysis

Data was captured and analysis was performed using GeneMapper 4.1.The polymorphism information content (PIC) values, referring to the value of a marker for detecting polymorphism with in a population, were determined for each marker using the number of alleles detected and the distribution of their frequencies (Abu Assar et al., 2005) as;

$$PIC_i = 1 - \sum_{j=1}^{n} xij^2$$

Where: PIC_i= the polymorphism information content value of the i^{th} marker; X_{ij}= the frequency of the ith allele for the jth marker and totalled over n alleles. The program GGT 2.0 (van Berloo, 2008) was used to calculate the Euclidian distances between samples. The matrix of the genetic distances was used to construct the dendrogram using the unweighted pair group method with arithmetic mean allocated (UPGMA).

5.4 Results and discussion

5.4.1 Phenotypic characterisation

Analysis of variance and mean squares for the phenotypic traits among genotypes and replications are presented in Tables 5.2 and 5.3. Significant differences were detected between genotypes (P<0.01) for seven morphological traits, including days to 50% emergence, maturity and flowering, plant height, panicle length, head weight and seed yield (Table 5.2). No significant differences were found among genotypes on biomass and harvest index. The observed significant variation of the phenotypic traits revealed the probability of a high level of genetic diversity among the genotypes.

Genotype Hormat was the earliest genotype to emerge (5.33 days) whereas Birhan took more days (8.33 days) to emergence. Genotype 237289 had the highest value for number of days to 50% flowering and maturity at 80 and 137 days, respectively. Genotype IC9830 was the earliest to flower at 62 days, and *Striga* resistant genotypes N-13 and SRN-39 had the lowest number of days to 50% maturity at 101 and 105 days, respectively. Based on the number of days taken for flowering and maturity, genotypes could be selected for earliness in areas where moisture stress is prevalent. Ayana et al. (2000) has reported early flowering and short plant height sorghum types as valuable traits for lowland environments with erratic rainfall and short growing periods. Genotype 235921 and 235924 were recorded with greatest plant heights of 1850 and 1823 mm, respectively. However these genotypes markedly took longer days to reach 50% flowering at 74 and 79 days and extended days to 50% maturity of 128 and 131 days, respectively. In alignment with this result, Morgan and Finlayson (2000) reported that late maturing plants are generally taller than early flowering sorghum plants. Selection for these accessions is important in areas where farmers use sorghum straws for firewood, fences, and for animal feed, as in Ethiopia. However, selection for tallness only may not fulfil other yield advantages obtained from these genotypes. Genotype SRN-39 exhibited the highest head weight (438 g) and seed yield (302 g/plant). The harvest indices of the test genotypes were remarkably low, suggesting a general trend of high biomass yield, being an indispensable trait preferred by farmers who have multiple uses for sorghum.

The presence of morphological variation among the sorghum genotypes collected from different parts of Ethiopia was reported by Geleta et al. (2006) and Shewayrga et al., (2006). Similarly, in Tanzania Bucheyeki et al. (2009) observed sorghum genotypic variations on morphological traits including panicle weight, stem diameter, and grain yield as indicators of genetic diversity.

Table 5.2 Mean square values and significant levels of the nine phenotypic characteristics measured among 14 sorghum genotypes tested with three replications

Sources of variation	DF	Traits[1]								
		DEM	DF	DM	PHT	PLN	BM	HW	SY	HI
Replication	2	0.02	6.00	4.79	7215	6000	57202.00	1819.00	18496.00	0.01
Genotype	13	1.34**	95.46**	428.44**	16473**	393**	329309.00NS	2374.00**	14896.00**	0.03NS
Error	26	0.25	6.49	26.53	2648	49	462331.00	508.00	3735.00	0.002

DF= degrees of freedom; **= significantly different at 0.01 probability level; NS=Not significantly different

[1]DEM = days to 50% emergence; DF = days to 50% flowering; DM = days to 50% maturity; PHT = plant height; PLN = panicle length; BM = biomass; HW = panicle weight; SY= seed yield; HI= harvest index

Table 5. 3 Mean, least significant differences, and coefficient of variation of nine phenotypic characters among 14 sorghum genotypes

Genotypes	Traits[1]								
	DEM	DF	DM	PHT	PLN	BMS	HW	SYD	HI
235466	6.33	77.33	123.33	1780.00	287.00	2233.00	404.00	222.90	0.10
235761	6.00	74.00	127.00	1217.00	253.00	1450.00	312.00	213.10	0.15
235763	6.00	77.00	128.00	1363.00	257.00	1967.00	322.00	223.60	0.11
235921	6.00	74.33	127.67	1850.00	310.00	2283.00	341.00	235.80	0.10
235924	6.00	78.67	130.67	1823.00	220.00	2267.00	343.00	136.90	0.06
235926	6.00	80.00	130.33	1523.00	210.00	2050.00	149.00	78.00	0.03
235929	6.33	75.67	128.00	1177.00	203.00	2117.00	235.00	211.50	0.09
237289	6.00	80.33	137.00	1587.00	270.00	2367.00	223.00	101.00	0.13
2384443	6.00	73.67	129.33	1320.00	227.00	1783.00	338.00	210.20	0.12
Birhan	8.33	69.33	98.33	1120.00	260.00	2333.00	405.00	298.70	0.13
Hormat	5.33	70.67	118.67	1417.00	307.00	2650.00	315.00	195.70	0.08
IC9830	6.33	61.67	120.67	1430.00	217.00	1500.00	163.00	110.00	0.08
N-13	6.33	64.00	101.00	1440.00	243.00	2133.00	380.00	269.70	0.13
SRN-39	6.67	70.33	105.00	1613.00	297.00	1950.00	438.00	302.00	0.16
LSD	0.85	4.28	8.64	27.31	3.69	1141.10	119.60	102.55	0.09
CV (%)	8.10	3.50	4.20	11.00	8.70	32.70	22.80	30.50	51.20

[1]DEM = days to 50% emergence; DF = days to 50% flowering; DM = days to 50% maturity; PHT = plant height (mm); PLN = panicle length (mm); BMS = biomass (g $plant^{-1}$); HW = panicle weight (g); SY= seed yield (g $plant^{-1}$); HI= harvest index

5.4.2 Principal component analysis

Based on the principal component analysis, the first three principal components (PCs) which had eigenvalues greater than one were considered (Table 5.4). These three principal components cumulatively explained 73.99% of the total variation. The first PC alone explained 38.87% of the total variation, mainly due to its correlation with days to maturity, seed yield and head weight (Table 5.4, bold faced scripts). Days to maturity contributed to this variation in this PC with a high negative loading. The second PC accounted for 23.99% of the total variation and highly correlated with plant height, panicle length and biomass. The third PC with 11.12% of the variation composed of days to emergence and harvest index.

There was a strong and positive association between the grain yield and the head weight, which fell in to the same PC in this study. This relationship has been also supported by Abe (2010). Similarly, the relationship between plant height and days to 50% flowering has also been observed in other studies (Ayana, 2001; Kebede et al., 2001). Observation on the pattern of morphological traits variation and their relationship among the sorghum genotypes is fundamental in order to facilitate the precise identification of genetic divergence and reliable classification into different groups, which helps sorghum improvement programmes (Ayana, 2001; Grenier et al., 2004; Chozin, 2007).

Table 5.4 Principal component analysis of nine phenotypic characters in 14 sorghum genotypes with the variation explained by the three principal components

Characters	Eigenvectors		
	PC1	PC2	PC3
Days to emergence	0.652	-0.257	**0.535**
Days to flowering	-0.586	0.446	0.069
Days to maturity	**-0.916**	0.134	-0.131
Plant height	-0.262	**0.720**	-0.228
Panicle length	0.390	**0.761**	-0.065
Biomass	0.088	**0.712**	0.349
Head weight	**0.783**	0.453	-0.114
Seed yield	**0.919**	0.136	-0.095
Harvest index	-0.452	0.219	**0.702**
Eigeanvalue	2.71	2.45	1.50
Individual (%)	38.87	23.99	11.12
Cumulative (%)	38.87	62.87	73.99

5.4.3 Cluster analysis

Cluster analysis on the nine phenotypic traits showed clear demarcation between sorghum genotypes (Figure 5.1). The analysis provided two main clusters of the 14 genotypes. Cluster I included six genotypes, of which four landraces acquired from IBC/Ethiopia which were characterized with effective *F. oxysporum* compatibility. The other two genotypes (Birhan and Hormat) are improved varieties having *Striga* resistance. The second cluster consisted of eight genotypes representing IBC and

ICRISAT acquisitions (Figure 5.1). Cluster II was further divided into two sub-clusters. One of the sub cluster comprised of genotypes that exhibited *F. oxysporum* compatibility, the other two genotypes were from ICRISAT (SRN-39 and N-13) which are *Striga* resistant. Genotypes IC9830 (sourced from ICRISAT) and 235761 were characterized as *F. oxysporum* compatible and grouped in the second sub cluster. Overall, the cluster analysis on the phenotypic traits clearly demarcated the genotypes with *F. oxysporum* compatibility and *Striga* resistance which are maintained for crosses for breeding. Souza and Sorrels (1991) reported the significance of morphological traits to characterize and cluster accessions based on their similarity in order to identify and select the possible parents for hybridization.

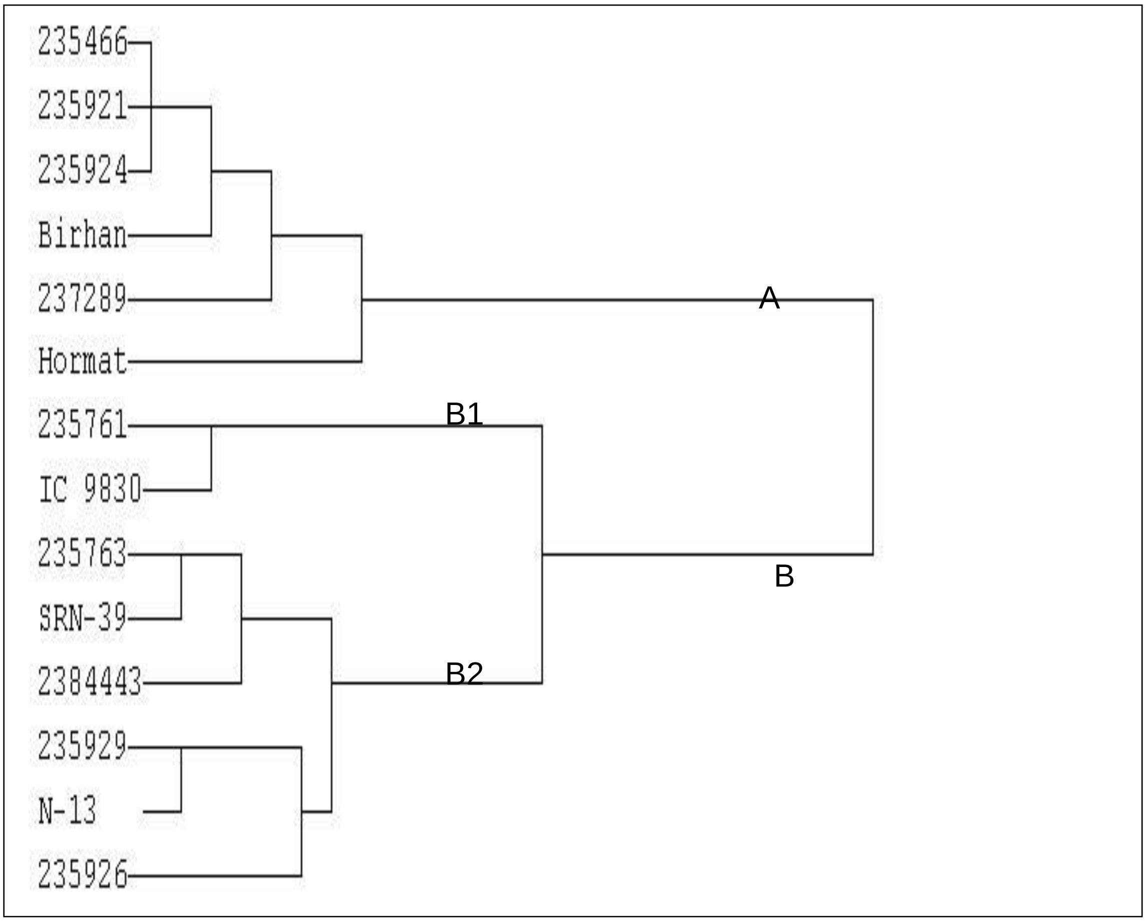

Figure 5.1 Dendrogram showing phenotypic relationships among 14 sorghum genotypes based on nine morphological characteristics using UPGMA clustering

5.4.4 SSR analysis

Polymorphism and allelic diversity of SSR markers

The number and sizes of fragments amplified by each SSR marker, polymorphic information content (PIC) values and heterozygosity of the markers are listed in Table 5.5. The SSR markers generated a total of 139 alleles. Nine of the SSRs generated 2 to 6 alleles, 11 markers generated 7-11 alleles and an average of 6.95 alleles were generated per locus.

The maximum number of alleles detected was 11 using the marker Sb5-206, and the minimum was 2 with Xtxp208. The average number of alleles generated in this study is nearly same to the average number of alleles (7.6) reported by Wang et al. (2009). The large number of alleles generated by the SSRs is a useful indicator of genetic diversity for subsequent breeding (Petit et al., 1998). In this study the size of the amplified fragments ranged on average from 204 to 244 bp, which reflected remarkable differences in the number of repeats between the different alleles.

All the 20 SSR markers used in this study provided high level of polymorphism ranging from 0.71 to 0.87, with an average of 0.80, which allowed for the discrimination of tested genotypes. SSR marker Sb6-57 was the most informative with a PIC of 0.87, whereas Sb4-72 and Xtxp37 were slightly less informative markers both with a good PIC value of 0.71. Markers with higher PIC values have great use in validating the variation between alleles and they are useful in testing genetic variability (Andersen and Lubberstedt, 2003). The PIC value of SSR markers can range from 0 which is monomorphic to 1 which is very highly discriminative, with many alleles in equal frequencies. The markers used in this study were highly informative and able to identify heterozygosity for each locus that ranged from 0.00 to 0.83 with an average of 0.35. Twenty of the SSRs exhibited heterozygosity, indicating the existence of genetic diversity between the tested genotypes, as established by Agrama and Tuinstra (2003).

Table 5.5 Genetic information generated by twenty SSR markers among 14 sorghum genotypes

No	SSR markers	Chromosome	Repeat	No of alleles	Allelic size (bp)	PIC value	He
1	Xtxp41	6	(CT)19	8	278-321	0.84	0.36
2	Xtxp217	4	(GA)23	7	182-203	0.83	0.26
3	Xtxp37	1	(TC)23	6	186-210	0.71	0.81
4	Xtxp208	6	(GGA)8	2	271-285	0.8	0.07
5	Xtxp33	4	(TC)20C(TG)5+ (CT)9CC(TG)7	10	180-260	0.76	0.43
6	Xtxp15	6	(TC)16	5	224-240	0.85	0.29
7	Xtxp10	8	(CT)14	8	153-180	0.86	0.33
8	Xtxp43	6	(CT)28	9	166-206	0.74	0.56
9	Xtxp303	4	(GT)13	4	162-189	0.82	0.21
10	Xtxp8	4	(TG)31	10	132-180	0.81	0.06
11	Sb6-84	8	(AG)14	6	196-235	0.84	0.25
12	Sb4-72	1	(AG)16	4	196-233	0.71	0.83
13	Sb1-10	3	(AG)27	9	256-330	0.79	0.07
14	Sb5-206	2	(AC)13/(AG)20	11	120-174	0.73	0.62
15	Sb6-342	7	(AC)25	8	290-313	0.81	0.05
16	Sb4-121	2	(AC)14	7	228-252	0.79	0.00
17	Sb6-57	11	(AG)18	6	295-334	0.87	0.49
18	Sb5-236	6	(AG)20	9	185-210	0.86	0.57
19	Sb6-34	7	[(AC)/(CG)]15	4	214-223	0.85	0.48
20	Sb1-1	3	(AG)16	6	158-297	0.78	0.19
Average				6.95	204-334	0.80	0.35

PIC= Polymorphic information content; He= Heterozygosity

5.4.5 Genetic distance

The Euclidean distance between each of the 14 bulked sampling units to estimate the genetic diversity is shown in Table 5.6. The Euclidean genetic distance ranged from 2.7 to 6.0 and recorded pair-wise for comparisons. The minimum distance of 2.7 was observed between Genotypes 235929 and 235926, whereas, the maximum distance (6.0) was observed between Genotypes 235929 and 235466. Assessment of the genetic distances assists in the establishment of parental lines and creation of segregating populations in order to exploit the diversity required in crop breeding programs. The Pearson bivariate correlation analysis of the Euclidean distances measured using phenotypic traits and SSR markers resulted significant correlation between the two distances ($r=0.74$; $p<0.05$). This indicated the effectiveness of genotypes grouping using both phenotypic and SSR markers.

Table 5.6 Pair-wise Euclidean genetic distance estimates among 14 sorghum genotypes

	235466	235761	235763	235921	235924	235926	235929	237289	2384443	IC9830	Birhan	Hormat	N13	SRN39
235466														
235761	5.3													
235763	4.8	3.2												
235921	4.6	3.6	4.1											
235924	5.1	3.7	3.1	4.5										
235926	5.9	4.8	5.3	4.7	4.7									
235929	6.0	4.7	5.1	4.6	4.6	2.7								
237289	5.1	4.4	4.8	3.5	4.0	4.4	3.8							
2384443	4.8	3.9	4.1	3.4	3.4	4.7	4.5	3.9						
IC9830	4.7	5.1	4.9	4.4	4.0	4.7	4.9	3.2	3.9					
Birhan	5.0	4.2	4.3	3.6	4.3	5.0	4.8	4.5	4.2	4.4				
Hormat	4.6	3.9	4.2	3.6	4.9	4.9	4.8	3.7	4.2	3.7	3.4			
N13	4.7	5.2	5.1	5.1	4.8	5.1	4.6	4.2	5.2	4.1	5.0	4.7		
SRN39	4.2	4.5	4.8	5.0	4.9	5.8	5.7	4.7	4.8	4.9	4.1	4.4	4.9	

5.4.6 Cluster analysis

The Euclidean dissimilarity matrix was used to cluster genotypes using the UPGMA algorithm. Dendrogram from the cluster analysis revealed two distinct groups of the genotypes (Figure 5.2). The two clusters are referred here with as Group A and Group B. Group B is further defined into two sub-groups namely Group B1 and Group B2 (Figure 2). Group A comprises of accessions 235466, SRN39 and N13. Group B1 comprises of accessions 235761, 235763, 235924, Birhan, Hormat, 235921, 2384443, 237289 and IC9830. Group B2 comprises of genotype 235926 and genotype 235929. The genotypes in Group A were *Striga* resistant and obtained from ICRISAT. Group B predominantly included genotypes sourced from the IBC of Ethiopia, which were screened for their *F. oxysporum* compatibility in *Striga* infested soils. Two *Striga* resistant genotypes sourced from Ethiopian sorghum improvement programme were also included in this group. The clustering of the genotypes may guide for further parental selection in integrated *Striga* management programme.

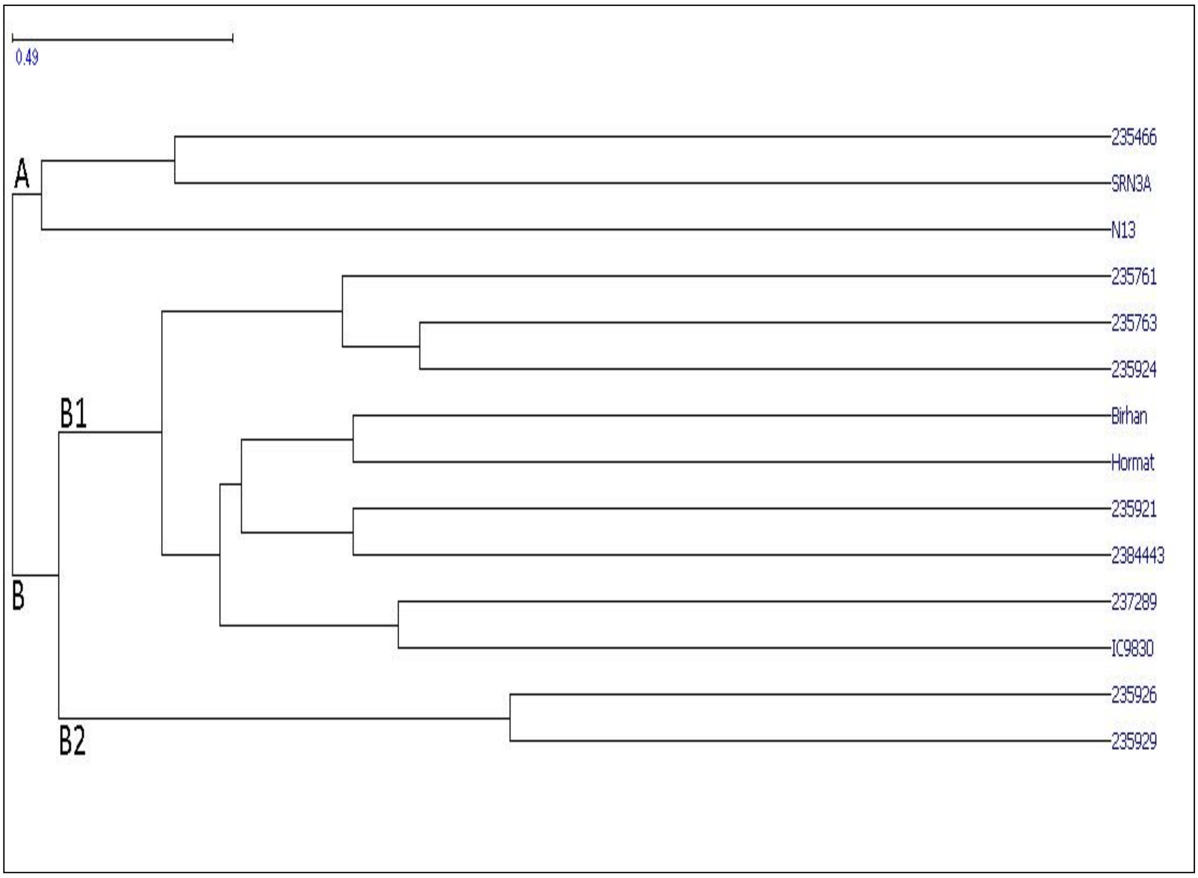

Figure 5.2 Dendrogram depicting genetic relationship among 14 sorghum genotypes selected for *F. oxysporum* compatibility and *Striga* resistance when evaluated using 20 SSR markers.

5.5 Conclusion

A total of 14 sorghum genotypes were evaluated using phenotypic and molecular markers. The phenotypic traits substantially contributed to differentiating the genotypes studied. Moreover, based on the observed variation exhibited using SSR markers, it could be concluded that studying the genetic interrelationship among sorghum genotypes is important to identify the genetic potential of parental lines to increase the efficiency of the sorghum breeding programmes.

The present study concluded the presence of genetic variation using selected morphological and polymorphic SSRs. Genotypes which are *Striga* resistant (N-13, SRN-39) and others which are *F. oxysporum* compatible were grouped clearly.Together they can be used as breeding parents towards cultivar development with integrated *Striga* management. Further strategic breeding can be implemented using the tested genotypes with *Striga* resistance. This trait could be transferred to other genotypes grouped under different clusters. Moreover, genotypes showing *F. oxysporum* compatibility and better phenotypic variation could be targeted for sorghum improvement without altering traits preferred by farmers such as biomass yield and earliness.

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CHAPTER 6

Combining ability of grain yield and *Striga* resistance in sorghum [*Sorghum bicolor* (L.) Moench]

6.1 Abstract

Sorghum [*Sorghum bicolour* (L.) Moench] is an important grain crop as a source of food, feed and bio-energy. However, its yield is significantly low in the sub-Saharan Africa primarily due to one of the major production constraints, *Striga* (*Striga hermonthica)*. The use of *Striga* resistant cultivars and Integrated *Striga* Management (ISM) are considered as novel approaches to minimize damage inflicted by *Striga*. The objective of this study was to identify promising parents and/or hybrids with high combining ability for grain yield, yield components, and *Striga* resistance for breeding under ISM. Forty sorghum hybrids were developed through the line by tester mating design involving 10 lines selected for their compatibility with *Fusarium oxysporum,* a bio-control agent, and high agronomic performances and four *Striga* resistant tester parents. The F_1s and their parents were field evaluated with complementary *in-vitro* tests. Field evaluations were conducted at two locations: Kobo and Shewa Robit in Ethiopia known for their severe *Striga* infestation, using the row by column lattice experimental design. Important data were collected and analysed on both sorghum and *Striga* parameters. Significant ($p<0.05$) general combining ability (GCA) effects were observed among testers and lines at both sites on days to 50% flowering and maturity, plant height, biomass, number of *Striga* plants and *Striga* plant height. Furthermore, significant ($p<0.05$) specific combining ability (SCA) effects were detected on days to 50% flowering, biomass, grain yield and number of *Striga* plants. At Kobo, crosses 235763 x N-13 and Shewa Robit IC9830 x SRN-39 had significantly negative SCA effects on the numbers of *Striga* plants. From the complementary *in-vitro* experiment, highly significant variation ($p<0.01$) was exhibited due to line x tester interaction for maximum *Striga* germination distance. The study identified parents with high GCA effects including SRN-39 and Birhan (as paternal) and 235761, 2384443, IC9830, 235466, 237289,235763, and 235929 (as maternal) useful for breeding for ISM in sorghum.

Key words: general combining ability, sorghum, specific combining ability, *Striga*

6.2 Introduction

Sorghum [*Sorghum bicolour* (L.) Moench] is the fifth most important cereal crop worldwide. The world average sorghum yield is estimated at 1314 kg/ha. The average sorghum yields of developed and developing countries are 3056 kg/ha and 1127 kg/ha, respectively. The developing countries account for 90% of sorghum production area despite the low productivity (FAOSTAT, 2005). In Ethiopia, during 2010, sorghum was the fourth most important cereal crop after tef, wheat, and maize. In the country, sorghum encompasses about 16.38% of the total of 84.69% area allotted to cereal crop production (CSA, 2011). The low yield level in the developing countries is attributed to both biotic (pests, diseases and the parasitic weed, *Striga*) and abiotic (drought and low soil fertility) stress factors (Obilana, 2004).

Striga hermonthica (Del.) Benth, the root parasitic weed endemic in Africa, is the most pernicious biotic factor that threatens cereal crops (sorghum, maize, millet, rice) production in sub-Saharan Africa (Dashiell et al., 2000; Ejeta, 2007). About 21 million hectares of cereal production area in Africa are estimated to be infested by *Striga*, causing an annual grain loss of about 4.1 million tons (Ejeta, 2007). Losses due to *Striga* could vary from 5 to 100% depending on the level of infestation, susceptibility of the crop, climatic conditions, and nature of the soil (Salle et al., 1987; Lagoke et al., 1991; Haussmann et al., 2000a). In Ethiopia, approximately 600,000 hectares of sorghum land is severely infested with *Striga* and remains a significant factor in grain yield losses of over 640,000 tons (Tesso et al., 2007). This is compelling farmers to shift from growing sorghum to other less susceptible crops, or abandoning their farm land (Yohanness et al., 1999). *Striga* is threatening the food security imperative of many resource poor farmers in sub-Saharan Africa. Developing effective technological options for the control of *Striga* has been one of the top sorghum research priorities globally (Ejeta, 2007).

Several methods of controlling *Striga* have been recommended. These include host plant resistance (Lagoke et al., 2000; Adeosun et al., 2001; Gwary et al., 2001; Haussmann et al., 2001); cultural control practices such as hand weeding, crop rotation, trap-cropping, intercropping, use of nitrogen fertilizer (Kuchinda et al., 2003; Ast et al., 2005; Reda and Verkleij, 2007); chemical control (Ejeta et al., 1996;

Ndung'u, 2009) and biological control (Ciotola et al., 2000; Fen et al., 2007; Rebeka, 2007). However, the use of the various control options singly has proved to be ineffective to control *Striga* and consequently sorghum yield losses remain high.

Recently, the combined application of two or more control measures has been promoted for effective *Striga* management. The use of biocontrol agent such as virulent isolate of *Fusarium oxysporum* f.sp. *strigae* as a component of integrated *Striga* management was identified to have several advantages (Ciotola et al., 2000; Fen et al., 2007). For instance, Elzein and Kroschel(2004) and Gupta and Lagoke (2000) reported that the use of mycoherbicide and host plant resistance to control of *Striga* led to simultaneous reduction of *Striga* seed bank, prevention of new *Striga* seed setting and grain yield improvement. The application of integrated *Striga* management package combining a mycoherbicide based on *F. oxysporum* isolate and host plant resistance has been demonstrated on farmers' fields as effective *Striga* control approach (Marley et al., 2004; Schaub et al., 2006).

Combining ability studies are useful in plant breeding programs to determine the nature of gene action and consequently to identify suitable parents for cultivar development. The general combining ability (GCA) is defined as the average performance of a line in hybrid combinations and specific combining ability (SCA) referred as the deviation in a particular cross from performance predicted on the basis of general combining ability (Sprague and Tatum, 1942; Schlegel, 2010). The GCA effect is due to additive gene action and that of SCA is largely dependent on genes with dominance and/or epistatic effects. Comstock and Robinson (1948) introduced the line by tester matting scheme to study the combining ability of lines. In the line by tester design a set of female parents are crossed with a genetically different set of male parents in all possible combinations. Combining ability studies in sorghum using the line by tester design indicated the influence of GCA effect for grain yield and its components and earliness (Kenga et al., 2004; Prabhakar and Raut, 2010). Previous findings suggested that *Striga* resistance in sorghum is controlled by a relatively few genes with additive genetic effects (Shinde and Kulkani, 1982). Haussmann et al. (2000b) reported on the difficulty of examining the nature of genes conferring *Striga* resistance. The complex *Striga* resistance should be assessed in the field and pot experiments to identify traits for direct and indirect selection for *Striga* resistance (Omanya et al., 2004). In sorghum one of the resistance triggering mechanisms is low stimulation of *Striga* seed germination which can be easily assessed using agar-gel assay described by Hess et al. (1992).

Recent findings indicated the effectiveness of integrated use of *Fusarium oxysporum* compatible and *Striga* resistant sorghum genotypes to control *Striga* in Ethiopia (Rebeka et al., 2013). To realize the full potential of this approach it is important to recombine traits of *Fusarium* compatible and *Striga* resistant sorghum lines. This would allow continued selection of targeted progenies with combined resistance and *Fusarium* compatibility and for subsequent seed treatment of suitable hybrid(s) for direct use. Thus effective *Striga* control would be possible through synergistic effect of biocontrol and host resistance. Therefore, the objective of this study was to identify *F. oxysporum* compatible parents and hybrids with high combining ability for grain yield, yield components, and *Striga* resistance for integrated *Striga* management (ISM).

6.3 Materials and methods

6.3.1 Plant material and crosses

Forty F_1 hybrids were developed from crosses of 10 selected sorghum lines as females with four *Striga* resistant sorghum genotypes as males or testers. Crosses were done using the line by tester mating design whereby every female parent was crossed with every male parent. The 10 female parents were identified based on confirmed compatibility with *Fusarium oxysporum* as a *Striga* biocontrol agent as well as their promising agronomic performances. The description of sorghum parents used in the crosses is shown in Table 6.1.

Parents were planted in four rows of 5 meters long plots at three different planting dates at 15 days intervals to synchronize flowering for crosses. Pollen collected from each of the male parents was used to pollinate hand emasculated and bagged female panicles. For each of the crosses approximately 10 female plants were pollinated to obtain sufficient quantity of hybrid seeds.

Table 6.1 Description of sorghum genotypes used in the crosses with source, pedigree, parentage, and level of *Striga* resistance

No.	Genotype	Source*	Pedigree	Parentage**	Level of *Striga* resistance***
1	Birhan	SARC	PSL85061	M	R
2	Hormat	SARC	ICSV 1112 BF	M	R
3	N-13	ICRISAT	N-13	M	R
4	SRN39	ICRISAT	SRN39	M	R
5	235466	IBC	Landrace	F	S
6	235761	IBC	Landrace	F	S
7	235763	IBC	Landrace	F	S
8	235921	IBC	Landrace	F	S
9	235924	IBC	Landrace	F	S
10	235926	IBC	Landrace	F	S
11	235929	IBC	Landrace	F	S
12	237289	IBC	Landrace	F	S
13	2384443	IBC	Landrace	F	S
14	IC9830	ICRISAT	IC9830	F	S

*****IBC : Institute of Biodiversity Center (Ethiopia); ICRISAT : International Crop Research Institute for the Semi-Arid Tropics (India); SARC : Sirinka Agricultural Research Center (Ethiopia).

**M=male; F=Female; ***R=Resistant; S=Susceptible

6.3.2 Field experiments

6.3.2.1 Description of study sites

The 40 F_1 hybrids together with the 14 parents were evaluated at two severely *Striga* infested sorghum growing sites namely, Kobo and Shewa Robit in 2012. The two sites are located in the East African rift valley lowland part of north eastern Ethiopia at an altitude ranging between 1350 and 1650 meters above sea level. Kobo is situated at the geographical location of E39^{o}37' and N12^{o}09', and Shewa Robit at E 29^{o}53' and N09^{o}98'. Temperatures in the study sites typically range from 17 to 35 ^{o}C, with total annual rainfall of <1000 mm and its distribution is often erratic (McGuire, 2008).

6.3.2.2 Experimental design and field management

Crosses and parent genotypes were planted in the row-by-column α-lattice design using two replications. Each entry was sown in three rows of three metres long with inter-row spacing of 75 cm. Seedlings were thinned to 30 cm spacing between plants at the 3rd week after sowing. Diammonium phosphate and urea fertilizers were applied at the recommended rate for the study areas. Weeding was done as much as necessary by leaving *Striga* plants intact.

6.3.2.3 Data collection

Data were collected from the middle row on panicle weight (g), grain yield (kg ha^{-1}), 1000-seed weight (g) and above ground biomass (kg ha^{-1}). A randomly selected five plants were used to measure plant height (cm) and panicle length (cm). Data on days to 50% emergence, flowering, and maturity of all entries were recorded on whole plot bases.

Further, data were recorded on number of *Striga* plants, *Striga* plant height, numbers of *Striga* branches from the entire plot to determine the level of *Striga* resistance of sorghum genotypes. The count data on the number of *Striga* plants and number of branches on each *Striga* plant were square root transformed before conducting analysis of variance for these traits. *Striga* vigour and severity were assessed following Haussmann et al. (2000a). Briefly, scoring on *Striga* vigour per plot was carried out using a 0-9 scale as described in Table 6.2. The *Striga* severity on each entry was obtained by multiplying the number of *Striga* plants and *Striga* vigour score.

Table 6.2 Description of the scoring scale for assessing *Striga* vigor (Haussmann et al., 2000a)

Score	*Striga* height (cm)	Number of *Striga* branches
0		No emerged *Striga* plants
1	≤5	None
2	6-20	None
3	6-20	≥1
4	21-30	≤5
5	21-30	>5
6	31-40	≤10
7	31-40	>10
8	>40	≤10
9	>40	>10

6.3.3 *In-vitro* experiment

Striga seeds collected during the 2011 cropping season from the two experimental sites were used for agar gel assay. The 40 F_1 hybrids and 14 parents indicated in Table 6.1 were used in this experiment. Seed viability tests, sterilization and conditioning of *Striga* seeds were done following the method described by Hess et al. (1992). Accordingly, viability of *Striga* seeds was tested by plating 50-100 seeds on a petri dish lined with filter paper and covered with alumunium foil. Tetrazolium solution (pH 6-8) was added until it covered all the seeds and then placed in autoclave at 40^{o}C in the dark for 48 hours. After this, the solution was drained by pouring the mixture into a funnel lined with filter paper. Finally, the seeds were placed on new filter paper in a clean petri dish. Thereafter 1% NaOCl solution was added for microscopic observation where red stained endosperm seeds indicated viable seed.

After confirming the *Striga* seeds viability, the *Striga* seed conditioning was done following the procedure explained by Mohamed et al. (1998). Briefly, 5 drops of Tween 20 were poured into a flask containing *Striga* seeds to remove debris and to break the *Striga* seeds surface tension. Then the seeds were rinsed using distilled sterile water and allowed to settle in a flask containing 15ml of aqueous benomyl solution. The flasks were then placed in a 28^{o}C incubator for two weeks for conditioning.

The preconditioned *Striga* seeds were pipetted in a sterile 25 cm diameter petri dish. The seeds were placed at a 3 cm distance from the edge of the petri dish. Finally, water agar (0.7%) was poured over the seeds. Roots of seven days old germinating sorghum seeds were placed in the solidifying agar with the root tip pointing at the centre of the petri dish. The petri dishes were incubated in the dark at 28^{o}C for seven days.

The experiment was arranged in a completely randomized design with three replications. Each genotype was represented by three petridishes in each replication. *Striga* seed germination was observed through the bottom of the petridish using a dissecting microscope fitted with graduated eyepieces. Maximum germination distance (MGD), that is, the distance between the sorghum seedling root and the most distant germinated *Striga* seed, was measured twice, i.e., after five and seven days of incubation (Hess et al., 1992). The distance is an indication of the level of genotypes resistance through giving an indication for the amount of *Striga* germination stimulant production. According to Hess et al., (1992), sorghum genotypes with a MGD of less than 1cm are group as a resistant while those with more than 1cm are group as a susceptible genotype to *Striga*.

6.3.4 Data analysis

Data analysis was performed using the general linear model procedure of the Statistical Analysis Systems statistical package (SAS 9.1, 2003). The general linear model procedure was performed by fitting the fixed effects of location, genotype and their interactions. Sources of variation due to the main effect of genotype and genotype x site interaction were partitioned into variations due to crosses and parents. Similarly, the source of variation due to crosses was partitioned into variations due to the main effects of lines, testers, lines x testers, and their interactions with sites. The effects of general combining ability (GCA) and the specific combining ability (SCA) as well as contribution of lines, testers and lines x testers to the total variation were estimated for each site by the Macro program of SAS developed for analysis of line x tester mating scheme (Bartolome and Gregorio, 2003). The significance effects of GCA and SCA were tested by a two-tailed t-test procedure.

6.4 Results and discussion

6.4.1 Field experiments

The combined analysis of variance (Table 6.3) revealed significant differences among crosses, lines and testers for yield and yield related traits. Line x tester interaction was significant only for days to emergence, plant height and panicle length. Interactions of location x crosses were significant ($p < 0.01$) for days to emergence, days to maturity, biomass, panicle weight and grain yield. Similarly, location x line interaction was significant for most of the traits except for days to flowering and thousand seed weight. Location x tester interaction had significant effect except on plant height, panicle length and thousand seed weight. The line x tester interaction across locations was significant ($p < 0.05$) for days to emergence, biomass, panicle weight and thousand seed weight. The relative performance of sorghum genotypes across sites was inconsistent for most of the traits considered. The significant genotype x site interactions indicates the need for targeted selection of crosses or genotypes to the target site and desired traits.

Significant variations existed among lines, testers and crosses and their interactions with site on days taken to 50% emergence, flowering and maturity. This indicates inherent genetic differences among test genotypes and/or the environment. Selection of early maturing genotypes has been considered as one of the possible options to mitigate the drastic effect of *Striga.* Early flowering genotypes are relatively less affected to *Striga* damage. However, these genotypes did not always yield more than the late maturing genotypes. Farmers prefer early maturing genotypes where moisture shortage and variability is the common phenomenon. Variations in grain fill duration also can be associated with the various and principal yield components (Kriegshauser et al., 2006).

Table 6.3 Combined analysis of variance for grain yield and yield related parameters of sorghum genotypes

Sources of variation	DF	Mean squares[1]								
		DEM	DFL	DM	PHT (cm)	BM (kg/ha)	PNL (cm)	PNW (g/plot)	GY (kg/ha)	TSWT (g)
Sites	1	15.042***	626.963***	19741.782***	50.267	1899195.766***	35.974**	2021668.656***	27927722.80***	4957.459***
Rep(sites)	2	0.079	20.296	1.708	3944.241***	26710.364***	19.019**	137953.378***	1499227.06***	6.169
Crosses	39	1.101***	53.944***	92.494***	3135.979***	113582.850***	20.497***	30516.160***	543263.31***	40.314**
Lines	9	2.63***	183.628***	148.361***	9820.459***	342223.389***	44.770***	33649.760**	433470.85 **	101.381***
Testers	3	0.973*	101.573***	615.883***	6747.049***	318145.040***	96.118***	194708.754***	4382886.56***	76.857**
Lines*Testers	27	0.709**	14.286	28.226	527.786**	26695.08	7.949**	11913.277	176894.97	15.949
Sites* Crosses	39	0.982***	9.509	76.383**	310.602	75552.290***	5.838	22377.124**	323205.08**	21.738
Sites *Lines	9	2.020***	7.562	58.944*	545.829*	124804.640***	7.400*	23792.554**	280803.80*	12.367
Sites *Testers	3	1.356**	43.489**	673.717***	176.583	141065.31**	13.285	110169.703***	2082757.88***	125.745
Sites *Lines* Testers	27	0.685**	5.953	20.272	257.584	535219.70**	4.622	14314.798**	177721.87	12.242**
Error	78	0.320	9.239	24.758	249.510	189814.70	3.695	8502.942	126201.99	17.006
R-Square (%)		83	82	91	88	94	81	86	86	83

1. * : significant at 5%; ** : significant at 1%; *** : highly significant at 0. 1% probability level.

DEM, days to emergence; DFL, days to flowering; DM, days to maturity; PHT, plant height; BM, biomass; PNL, panicle length; PNW, panicle weight; GY, grain yield; TSWT, thousand seed weight.

Table 6.4 shows the combined analysis of variance for *Striga* related parameters considered in the study. Significant differences were found in numbers and severity of *Striga* among crosses. *Striga* related traits varied significantly among lines. While testers varied significantly in their effect on the numbers of *Striga* plants and branches. Location x line interaction was significant for number and severity of *Striga*. Lines expressed significantly heterogeneous levels of tolerance to *Striga*. This could be attributed to the relatively broader genetic base of lines compared to the known *Striga* resistant tester genotypes.

Table 6.4 Combined analyses of variances for *Striga* related parameters

Sources of variation	DF	Mean squares[1]				
		SN	SPHT	SNB	SV	SSV
Sites	1	104.728***	19153.500***	75.789***	459.672***	50493.551***
Rep(sites)	2	1.872	1231.713**	6.997**	36.427**	143.352
Crosses	39	1.798*	166.459	1.058	4.929	3927.432*
Lines	9	3.657**	320.292*	1.982*	9.961*	3747.822*
Testers	3	3.411*	199.617	2.151*	8.241	2133.801
Lines*Testers	27	0.957	104.889	0.642	2.811	1399.368
Sites* Crosses	39	1.375	152.351	0.557	4.043	1881.504
Sites *Lines	9	2.510*	211.192	0.577	4.558	3571.447*
Sites *Testers	3	0.958	12.817	0.186	0.117	1820.683
Sites *Lines* Testers	27	1.078	138.108	0.587	4.112	1424.289
Error	78	1.205	148.484	0.803	4.381	1607.920
R-Square		0.69	0.71	0.68	0.70	0.61

[1] *: significant at 5%; ** : significant at 1%; *** : highly significant at 0.1% probability level
SN, number of *Striga*; SPHT, *Striga* plant height; SNB, number of *Striga* branches; SV, *Striga* vigourasity; SSV, *Striga* severity

6.4.2 General and specific combining abilities

The combined analysis of variance revealed the existence of variation among the crosses across locations. Subsequently, the general and specific combining abilities of the tested sorghum genotypes were estimated for each site. At Kobo, significant ($p<0.05$) GCA effect were observed for testers on days to maturity, panicle weight, and yield (Table 6.5). Among the testers, only SRN-39 and Birhan had significant ($p<0.05$) GCA effect for days to flowering and panicle length. SRN-39 and N-13 also showed significant ($p<0.05$) GCA effects for plant height. All lines had significant ($p<0.05$) GCA effects for plant height except line IC9830. However, IC9830 and line 235761 had significant ($p<0.05$) negative GCA effects, where negative effects are desirable, for the number of *Striga* plants, *Striga* plant height and number of *Striga* branches. Though not consistently shown, some lines (235924, 235926, and 235466) exhibited significant GCA effects on other parameters such as plant height, biomass, and grain yield.

Table 6.6 shows the GCA effects at Shewa Robit site. The GCA effect on plant height was significantly different ($p <0.05$) among testers and lines, except lines 235763 and 237289. Concerning *Striga* resistance, among testers only Birhan had significant negative ($p<0.05$) GCA effects in the number of *Striga* plants. Among lines significant negative GCA effects were expressed for *Striga* traits by line 235761 ($p <0.01$), and in plant height and number of branches of *Striga* by line 235466 ($p <0.05$). Other lines (235926, 235929, 237289 and IC9830) had significant negative GCA effect for *Striga* plant height ($p < 0.05$).

The highly significant GCA component of the genotypic variance indicated that the performance of the progeny could be adequately predicted on the basis of additive gene action and these genes are preferred for genotypes improvement through selection (Stoskopf et al., 1993). For instance, the significant negative GCA effects recorded by 235761, IC9830, and Birhan, indicated that *Striga* resistance can be inherited to the progeny because resistant genotypes imparted *Striga* resistance by reducing the number of *Striga* counts in the progeny. Therefore, breeders can expect genetic gains through selection from the segregating generations derived from the abovementioned parents showing significant GCA towards the desired direction for each trait.

Significant specific combining ability effects were noted only for limited traits including days to flowering, biomass, grain yield and number of *Striga* plants (Tables 6.7 and Table 6.8). This indicates the relatively less importance of non-additive genetic effects (dominance and/or epistasis) in the expression of such traits. Regardless of this general trend, specifically at Kobo, cross 235763 X SRN-39 had significant ($p<0.05$) positive SCA effect for the panicle weight, thousand seed weight, and yield. Also, this cross had greater negative SCA effect (-11.83) for the number of *Striga* plants. Likewise, crosses 235466 X SRN-39 and IC9830 X SRN-39 revealed significant negative and positive SCA effects for days to emergence and biomass, respectively. Cross 235924 X Birhan showed negative SCA effects on panicle weight and grain yield, which is undesirable to exploit heterosis (Table 6.7). On the other hand Table 6.8 presents that crosses of line 237289 with all testers except SRN-39 showed negative SCA effect for the number of *Striga* plants at Shewa Robit. Cross IC9830 X SRN-39 had significant SCA effects for days to flowering, plant height, and biomass and panicle length which is desirable. Crosses showed better specific combining ability effects at Shewa Robit than at Kobo (235763 X N-13, IC9830 X SRN-39 and 237289 crosses with N-13, Birhan and Hormat) for the sorghum biomass, plant height and panicle length improvement and for the number of *Striga* count reduction are the as the best combiners for *Striga* related parameters at this site. Sometimes two poor general combiners may have good specific combination due to epistatic gene action. For instance, crosses derived from 237289 with N-13, Birhan and Hormat have showed negative significant SCA effects for the number of *Striga* count but the parents do not exhibit good GCA effect for this specific trait. In this case, such crosses would not yield transgressive segregants in the ensuing selection generations. Therefore, they could not appreciably proceed for further generational selection in self-fertilizing crops (Akbar et al., 2009).

Overall, the above results indicated the importance of additive genetic effect for trait expression and selection response for the targeted *Striga* resistance and grain yield and its components in the test sorghum population. Thus selection of good general combiners and continued recurrent selection from recombined parents is highly important to enhance breeding progress. Similar to the present findings, other reports (Kenga et al., 2004; Tadesse et al., 2008; Makanda et al., 2009) suggested the presence of additive gene action controlling plant height and panicle length in sorghum. Further, in line with this study, significant site or environmental differences and genotype by environment interaction were found to contribute to higher GCA effect (Chapman et al., 2000; Kenga et al., 2004). During the evaluation of sorghum genotypes for *Striga* resistance across locations, Haussmann et al. (2001) accentuated the significance of genotype x environment interaction affecting *Striga* resistance and grain yield.

Table 6.5 General combining ability estimates on agronomic parameters among sorghum parents and associated *Striga* number, plant height and branches at Kobo during 2012

Sorghum parents	Sorghum									*Striga*		
	DEM	DFL	DM	PHT (cm)	BM (kg/ha)	PNL (cm)	PNW (g)	GY (kg/ha)	TSWT (g)	SN	SPHT	SNB
SRN-39	0.03	-2.78**	-6.95**	-10.63*	-994.44	-1.55*	71.54*	380.74**	0.23	-0.58	-1.05	-0.44
N-13	-0.03	-0.58	8.15**	12.83*	816.66	-0.10	-75.97*	-337.26*	0.45	0.32	1.50	0.81
Birhan	-0.08	1.98*	3.10*	-5.73	-783.33	1.25*	-73.26*	-327.08*	-0.45	-1.83	-1.35	-0.34
Hormat	0.08	1.38	-4.30*	3.53	961.11	0.40	77.69*	283.59*	-0.23	2.08	0.90	-0.04
235763	-0.08	-0.15	-1.13	-29.73**	847.78	1.28	48.76	100.97	0.51	5.98*	3.05	2.44*
237289	-0.08	-4.40**	-3.63	20.78**	-502.22	0.28	17.86	142.80	4.51**	-1.03	9.93*	1.81
235466	0.18**	0.35	5.25*	15.03*	1247.78	2.03*	78.64	355.13*	-1.09	-1.78	-3.45	-1.56
IC9830	0.18**	-3.28**	-1.50	11.65	-852.22	-0.85	-11.53	-40.31	0.21	-4.65*	-14.33*	-2.31*
235924	0.05	4.85**	1.25	22.03**	3092.22**	1.15	55.92	-8.37	2.06	-1.40	1.93	0.31
235921	-0.08	1.73	2.75	26.15**	1370.00	2.78**	52.21	233.41	0.41	-3.03	-3.83	-0.56
2384443	0.05	0.23	-8.50**	-39.73**	-2841.1**	-3.23**	-83.97*	-191.26	-5.49**	0.48	0.18	-0.94
235929	-0.08	-3.28**	-3.00	-21.35**	-2018.9*	-1.85*	-59.53	-99.26	1.71	-1.65	-0.33	-0.31
235761	-0.08	5.60**	5.50*	-30.60**	-1963.3*	-1.23	-152.03**	-637.53**	-4.24**	-9.98**	-15.18**	-2.56*
235926	-0.08	-1.65	3.00	25.78**	1620.00*	-0.35	53.69	144.41	1.41	-2.90	-8.33	-1.44

[1] *: significant at 5% and **: significant at 1% probability level

DEM, days to emergence; DFL, days to flowering; DM, days to maturity; PHT, plant height; BM, biomass; PNL, panicle length; PNW, panicle weight; GY, grain yield; TSWT, thousand seed weight ; SN, number of *Striga* ; SPHT, *Striga* plant height; SNB, number of *Striga* branches

Table 6.6 General combining ability estimates on agronomic parameters among sorghum parents and associated *Striga* number, plant height and branches at Shewa Robit during 2012

Sorghum parents	Sorghum									*Striga*		
	DEM	DFL	DM	PHT (cm)	BM (kg/ha)	PNL (cm)	PNW (g)	GY (kg/ha)	TSWT (g)	SN	SPHT	SNB
SRN-39	-0.19	-0.19	0.25	-10.91**	-546.67*	-2.01**	10.59	64.83	-3.07**	-1.21*	1.83	0.19
N-13	0.16	-0.74	-0.10	15.65**	257.78	0.49	-26.41	-155.66*	2.22*	-0.39	-0.59	-0.18
Birhan	0.16	-0.44	-0.40	-6.40**	111.11	1.13	-3.65	-0.92	1.69	-0.59	-2.32	-0.48
Hormat	-0.14	1.36	0.25	1.66*	177.78	0.39	19.46	91.74	-0.85	-0.24	1.08	0.47
235763	-0.21	0.21	-1.00	-25.88**	662.22	1.51	-2.65	-9.57	-0.01	0.56	6.57**	0.94*
237289	-0.96**	-3.66**	-2.88*	29.22**	-393.33	0.48	24.60	112.43	5.09**	4.06**	11.01**	2.88**
235466	-0.09	1.46	1.25	-1.36	206.67	0.53	17.99	114.66	-3.31*	-0.56	-0.62	-0.56
IC9830	0.41	-2.79*	-1.00	2.99**	1315.56**	-0.85	77.94**	243.01*	-0.56	-0.19	-0.37	0.19
235924	1.04**	4.34**	3.38*	8.55**	1406.67**	-0.97	31.80	90.54	1.39	-0.56	-1.87	-0.31
235921	-0.21	1.09	1.25	25.97**	40.00	1.16	4.03	-15.23	-1.16	-0.69	-3.12*	-0.68
2384443	0.79*	-0.41	-0.38	-26.41**	-971.11*	-2.65**	-15.99	-83.40	-2.16	-0.69	-3.12*	-0.68
235929	-0.46	-1.79	-0.38	-1.21	-604.44	-0.62	-10.49	-33.57	1.09	-0.69	-3.12*	-0.68
235761	0.41	2.84*	1.88	-37.41**	1095.56**	-0.35	19.36	26.21	-0.66	-0.56	-2.24	-0.43
235926	-0.71*	-1.29	-2.13	25.52**	-126.67	1.76*	9.29	40.93	0.25	-0.69	-3.12*	-0.68

[1] *: significant at 5% and **: significant at 1% probability level
DEM, days to emergence; DFL, days to flowering; DM, days to maturity; PHT, plant height; BM, biomass; PNL, panicle length; PNW, panicle weight; GY, grain yield; TSWT, thousand seed weight ; SN, number of *Striga* ; SPHT, *Striga* plant height; SNB, number of *Striga* branches

Table 6.7 Specific combining ability (SCA) estimates for agronomic parameters of sorghum crosses and parameters measured from *Striga* at Kobo in 2012 crop season

Crosses	DEM	DFL	DM	PHT (cm)	BM kg/ha	PNL (cm)	PNW (g)	GY (kg/ha)	TSWT (g)	SN	SPHT	SNB
235763 X SRN-39	-0.03	-0.60	-2.18	3.13	-1461.11	1.18	-17.89	-11.52	3.87	-5.93	3.05	-0.69
235763 X N-13	0.03	1.20	7.23	16.18	661.11	-1.28	-197.89*	-814.63*	-4.95*	-11.83*	-13.50	0.56
235763 X Birhan	0.08	0.15	-6.23	-5.78	-983.33	-0.13	67.51	315.86	3.35	10.33*	4.35	-0.79
235763 X Hormat	-0.08	-0.75	1.18	-13.53	1783.33	0.23	148.27	510.30	-2.27	7.43	6.10	0.91
237289 X SRN-39	-0.03	1.15	-1.68	6.13	-800.00	0.68	-93.49	-316.69	-0.73	-0.43	-11.33	-1.56
237289 X N-13	0.03	2.45	5.23	-23.83	-766.67	-0.78	-28.34	-175.58	-2.35	1.68	-1.88	1.69
237289 X Birhan	0.08	-1.60	-6.23	14.73	877.78	1.88	117.31	510.91	1.35	2.33	13.98	2.34
237289 X Hormat	-0.08	-2.00	2.68	2.98	688.89	-1.78	4.52	-18.64	1.73	-3.58	-0.78	-2.46
235466 X SRN-39	-0.28*	-0.10	-1.05	2.38	-2327.78	-0.58	-73.33	-237.02	-1.93	4.83	12.05	2.81
235466 X N-13	0.28*	0.70	-3.15	10.93	3416.67*	1.48	111.68	316.53	3.05	0.43	-2.00	-1.44
235466 X Birhan	-0.18	4.15*	1.90	-18.53	1772.22	-2.38	53.47	166.58	-0.25	-2.93	-17.65	-1.79
235466 X Hormat	0.18	-4.75*	2.30	5.23	-2861.11	1.48	-91.82	-246.09	-0.87	-2.33	7.60	0.41
IC9830 X SRN-39	-0.28*	5.53*	10.20*	23.75	3105.56*	2.80	-17.20	-204.24	-1.43	1.20	-2.08	0.56
IC9830 X N-13	-0.23*	-2.18	-7.40	-7.70	-705.56	-0.65	58.15	451.09	2.55	-1.70	-2.13	-1.19
IC9830 X Birhan	-0.18	-0.73	0.15	3.85	672.22	-0.50	41.99	-86.20	-5.15*	-0.05	-6.78	-1.04
IC9830 X Hormat	0.68**	-2.63	-2.95	-19.90	-3072.22*	-1.65	-82.95	-160.64	4.03	0.55	10.98	1.66
235924 X SRN-39	0.35**	-6.10*	-3.05	-10.13	-1616.67	-0.70	121.45	658.92*	4.32	1.95	6.68	0.94
235924 X N-13	-0.10	1.20	0.35	3.93	683.33	-0.65	124.70	437.59	2.90	-0.45	-5.88	-1.31
235924 X Birhan	-0.05	0.15	8.90*	0.48	-605.56	0.50	-171.10*	-640.81*	-3.20	2.20	9.48	1.84
235924 X Hormat	-0.20	4.75*	-6.20	5.73	1538.89	0.85	-75.05	-455.70	-4.02	-3.70	-10.28	-1.46

Table 6.7 Continued

Crosses	DEM	DFL	DM	PHT (cm)	BM kg/ha	PNL (cm)	PNW (g)	GY (kg/ha)	TSWT (g)	SN	SPHT	SNB
235921 X SRN-39	-0.03	0.03	4.45	6.75	2994.44*	0.18	34.71	12.26	-0.03	0.08	-10.08	-1.19
235921 X N-13	0.03	-5.18	-9.15*	-6.70	-1150.00	0.23	93.42	424.03	1.15	-2.33	9.88	0.56
235921 X Birhan	0.08	0.28	7.40	5.85	-550.00	0.88	-70.84	-270.14	-0.55	-0.68	-7.28	-1.29
235921 X Hormat	-0.08	4.88*	-2.70	-5.90	-1294.44	-1.28	-57.29	-166.14	-0.57	2.93	7.48	1.91
2384443 X SRN-39	0.35**	0.03	3.20	0.13	1205.56	0.18	-1.82	-78.63	-2.53	-1.43	8.43	1.19
2384443 X N-13	-0.10	1.33	0.10	0.18	172.22	0.73	104.39	430.03	0.25	6.68	11.38	0.44
2384443 X Birhan	-0.05	-2.23	-7.85	3.73	-338.89	-1.63	-56.72	-204.14	2.35	-4.18	-8.78	-0.91
2384443 X Hormat	-0.20	0.88	4.55	-4.03	-1038.89	0.73	-45.86	-147.26	-0.07	-1.08	-11.03	-0.71
235929 X SRN-39	-0.03	5.03*	3.20	-14.75	-727.78	-1.20	-113.75	-514.63	-2.93	-0.30	-1.08	-1.44
235929 X N-13	0.03	-1.18	7.60	-2.20	-1316.67	-0.65	-127.99	-541.97	-3.95	-2.20	-2.63	-0.69
235929 X Birhan	0.08	-3.23	-6.35	9.35	172.22	2.50	67.09	423.19	4.35	-1.05	9.23	1.96
235929 X Hormat	-0.08	-0.63	-4.45	7.60	1872.22	-0.65	174.65*	633.41	2.53	3.55	-5.53	0.16
235761 X SRN-39	-0.03	-3.85	-6.80	-8.00	327.78	-1.33	107.64	429.64	0.02	0.08	-0.08	0.19
235761 X N-13	0.03	0.45	-0.40	23.55	-38.89	1.73	-60.69	-203.91	0.60	10.68*	5.88	0.44
235761 X Birhan	0.08	1.40	6.15	-11.40	-327.78	-1.63	-72.15	-324.97	-2.70	-8.18	-3.78	-0.91
235761 X Hormat	-0.08	2.00	1.05	-4.15	38.89	1.23	25.20	99.24	2.08	-2.58	-2.03	0.28
235926 X SRN-39	-0.03	-1.10	-6.30	-9.38	-700.00	-1.20	53.67	261.92	1.37	-0.05	-5.58	-0.81
235926 X N-13	0.03	1.20	-0.40	-14.33	-955.56	-0.15	-77.42	-323.19	0.75	-0.95	0.88	0.93
235926 X Birhan	0.08	1.65	2.15	-2.28	-688.89	0.50	23.42	109.74	0.45	2.20	7.23	0.59
235926 X Hormat	-0.08	-1.75	4.55	25.98	2344.44	0.85	0.33	-48.48	-2.57	-1.20	-2.53	-0.71
S.E	0.109	2.123	3.974	13.226	1436.654	1.527	83.4603	317.0576	2.384	4.5156	9.761	1.860

[1] *: significant at 5% and **: significant at 1% probability level

DEM, days to emergence; DFL, days to flowering; DM, days to maturity; PHT, plant height; BM, biomass; PNL, panicle length; PNW, panicle weight; GY, grain yield; TSWT, thousand seed weight ; SN, number of *Striga* ; SPHT, *Striga* plant height; SNB, number of *Striga* branches

Table 6.8 Specific combining ability (SCA) estimates for agronomic parameters of sorghum genotypes and parameters measured from *Striga* at Shewa Robit in 2012 crop season

Crosses	DEM	DFL	DM	PHT (cm)	BM kg/ha	PNL (cm)	PNW (g)	GY (kg/ha)	TSWT (g)	SN	SPHT	SNB
235763 X SRN-39	-0.69	-1.06	-1.75	3.36	-475.56	1.79	17.08	55.28	0.87	-1.96	2.48	-0.82
235763 X N-13	-0.54	0.99	2.10	19.80	1564.44*	-1.72	-9.67	-18.46	2.59	1.63	5.66	0.56
235763 X Birhan	0.96	0.18	-0.10	-8.05	-1000.00	0.45	-11.83	-40.08	-1.09	-0.66	-7.37	-1.14
235763 X Hormat	0.26	-0.11	-0.25	-15.11	-88.89	-0.52	4.42	3.26	-2.36	0.99	-0.77	1.40
237289 X SRN-39	0.06	-0.69	1.13	-1.34	224.44	-0.19	64.43	283.50	1.57	11.04**	1.04	0.99
237289 X N-13	0.21	1.36	-1.03	-18.20	175.56	-1.69	9.98	114.43	0.69	-3.86**	-3.03	-0.38
237289 X Birhan	-0.29	-1.94	-0.23	-2.95	-77.78	2.47	-6.43	-49.63	-1.79	-3.66**	-6.81	-2.08
237289 X Hormat	0.01	1.26	0.13	22.49	-322.22	-0.59	-67.98	-348.30*	-0.46	-3.51**	8.79*	1.47
235466 X SRN-39	-0.81	0.69	0.50	2.24	602.22	-1.64	-22.91	-48.28	0.57	-0.83	5.67	0.18
235466 X N-13	-0.16	-0.76	-0.65	-14.33	-1224.44	0.16	-60.31	-226.90	-0.72	0.26	-1.91	0.06
235466 X Birhan	0.84	-1.06	-1.35	-0.08	-322.22	-1.48	-20.37	-92.97	-4.19	0.46	-0.18	0.36
235466 X Hormat	0.14	1.14	1.50	12.17	944.44	2.96	103.58*	368.14*	4.35	0.11	-3.58	-0.59
IC9830 X SRN-39	-0.81	-5.44*	2.75	24.49*	1635.56*	4.24*	59.42	160.94	1.02	-1.71	-4.58	-1.07
IC9830 X N-13	-0.66	-1.01	0.60	-19.38	-1213.33	-2.27	-24.28	-63.90	-6.07*	-0.11	-2.16	-0.69
IC9830 X Birhan	0.34	-2.31	-1.60	2.78	-488.89	-2.01	-43.44	-193.97	3.86	0.59	2.57	0.60
IC9830 X Hormat	1.14*	-2.11	-1.75	-7.89	66.67	0.04	8.31	96.92	1.19	1.24	4.17	1.16
235924 X SRN-39	-0.94	-2.19	1.38	3.44	-420.00	0.96	70.13	264.50	3.87	-0.84	1.92	0.93
235924 X N-13	0.21	0.36	-1.78	4.58	731.11	1.06	37.93	99.66	-4.22	0.26	-0.66	-0.19
235924 X Birhan	0.21	0.06	-2.98	-1.08	700.00	-1.58	35.22	136.26	2.51	0.46	1.07	0.10
235924 X Hormat	0.51	1.76	3.38	-6.94	-1011.11	-0.44	-143.28**	-500.41*	-2.16	0.11	-2.33	-0.84

Table 6.8 Continued

Crosses	DEM	DFL	DM	PHT (cm)	BM kg/ha	PNL (cm)	PNW (g)	GY (kg/ha)	TSWT (g)	SN	SPHT	SNB
235921 X SRN-39	0.81	-0.94	-2.00	-3.79	-164.44	-2.87	-46.65	-170.83	0.42	-1.21	-1.83	-0.19
235921 X N-13	-0.04	0.11	1.35	-6.95	8.89	0.24	32.96	128.10	-0.47	0.39	0.59	0.18
235921 X Birhan	-0.54	-0.69	0.15	5.90	22.22	0.59	-10.56	-23.30	-0.15	0.59	2.31	0.48
235921 X Hormat	-0.24	1.51	0.50	4.84	133.33	2.04	24.25	66.03	0.19	0.24	-1.08	-0.47
2384443 X SRN-39	-0.19	-0.44	-1.88	-11.62	-42.22	-2.77	-90.33	-382.44*	-3.59	-1.21	-1.83	-0.19
2384443 X N-13	0.96	0.61	0.48	-3.88	-1024.44	0.94	90.82	332.49	3.94	0.39	0.59	0.18
2384443 X Birhan	-0.54	2.31	2.78	3.38	766.67	2.19	37.81	179.09	2.66	0.59	2.32	0.48
2384443 X Hormat	-0.24	-2.48	-1.38	12.12	300.00	-0.37	-38.29	-129.13	-3.01	0.24	-1.08	-0.47
235929 X SRN-39	1.06*	1.44	0.63	-5.82	35.56	1.31	-25.58	-134.28	-1.84	-1.21	-1.83	-0.19
235929 X N-13	-0.29	-0.01	0.48	14.03	253.33	2.51	-25.23	-184.46	3.49	0.39	0.59	0.18
235929 X Birhan	-0.29	-0.81	0.78	1.98	-44.44	-0.83	44.71	262.82	0.21	0.59	2.32	0.48
235929 X Hormat	-0.49	-0.61	-1.88	-10.19	-244.44	-2.99	6.11	55.92	-1.86	0.24	-1.08	-0.47
235761 X SRN-39	1.19*	-1.19	-1.13	1.99	-864.44	-0.37	-41.28	-155.17	-2.49	-0.84	0.79	0.56
235761 X N-13	0.84	-2.64	-1.28	23.33*	197.78	0.44	-12.03	-10.90	1.04	0.26	-0.28	-0.07
235761 X Birhan	-1.16*	2.06	0.53	-1.23	388.89	-0.01	-9.69	-55.41	-2.85	0.46	1.44	0.23
235761 X Hormat	-0.86	1.76	1.88	-24.09*	277.78	-0.07	63.01	221.48	4.29	0.11	-1.96	-0.71
235926 X SRN-39	0.31	-1.06	0.38	-12.94	-531.11	-0.47	15.69	126.78	-0.39	-1.21	-1.83	-0.19
235926 X N-13	-0.54	0.99	-0.28	1.00	531.11	0.33	-40.16	-170.07	-0.27	0.39	0.59	0.18
235926 X Birhan	0.46	2.19	2.03	-0.65	55.56	0.19	-15.42	-122.80	0.86	0.59	2.32	0.48
235926 X Hormat	-0.24	-2.11	-2.13	12.59	-55.56	-0.07	39.88	166.09	-0.21	0.24	-1.08	-0.47
S.E	0.542	1.899	2.262	11.274	716.041	1.591	47.518	171.619	2.294	1.121	3.087	0.783

[1] *: significant at 5%

DEM, days to emergence; DFL, days to flowering; DM, days to maturity; PHT, plant height; BM, biomass; PNL, panicle length; PNW, panicle weight; GY, grain yield;

TSWT, thousand seed weight ; SN, number of *Striga* ; SPHT, *Striga* plant height; SNB, number of *Striga* branches

6.4.3 Proportional contribution

At Kobo the contribution of lines to the total variance was greater for all traits considered in this study except to days to 50% emergence, maturity, panicle weight and yield. The contribution of testers was greater than that of lines for days to maturity (Table 6.9). The line x tester variance contribution was the highest for days to emergence, panicle weight and yield. On the other hand, at Shewa Robit, there was maximum contribution of crosses resulting from line x tester to total variances for days to emergence, panicle length, panicle weight, thousand seed weight, yield and number of *Striga* plants (Table 6.9). The remaining variation was highly contributed by lines for days to flowering, maturity, plant height, biomass and *Striga* related traits (*Striga* plant height and number of *Striga* branches).

Table 6.9 Proportion of the total variation (%) contributed by lines, testers, and line x testers on the expression of some traits in sorghum and *Striga* at two study sites

Characters	Contribution (%)					
	Lines		Testers		Lines x Testers	
	Kobo	Shewa Robit	Kobo	Shewa Robit	Kobo	Shewa Robit
Sorghum						
DEM	22.54	47.37	7.04	3.35	70.42	49.29
DEF	48.96	62.76	16.65	7.30	34.39	29.93
DM	22.09	57.93	45.37	1.29	32.54	40.78
PHT (cm)	75.26	68.32	9.37	13.84	15.37	17.84
BM (kg/ha)	51.86	56.78	12.42	8.45	35.72	34.77
PNL (cm)	54.95	28.62	18.33	24.99	26.72	46.39
PNW (g)	27.30	24.86	29.56	8.42	43.14	66.72
GY (kg/ha)	21.81	18.26	35.42	16.20	42.77	65.54
TSWT (g)	53.54	30.18	0.86	28.84	45.60	40.98
Striga						
SN	47.97	27.61	5.28	7.09	46.75	65.31
SPHT	49.95	63.59	1.16	7.68	48.89	28.73
SNB	58.83	64.64	5.26	7.22	35.91	28.14

DEM, days to emergence; DFL, days to flowering; DM, days to maturity; PHT, plant height; BM, biomass; PNL, panicle length; PNW, panicle weight; GY, grain yield; TSWT, thousand seed weight ; SN, number of *Striga* ; SPHT, *Striga* plant height; SNB, number of *Striga* branches

6.4.4 *In-vitro* experiment

Parents, crosses and parents versus crosses and the lines x testers showed highly significant difference (p<0.01) for the *Striga* maximum germination distance recorded twice, i.e. five and seven days after incubation (Table 6.10). Only crosses that resulted in significant combining abilities are isolated and summarised in Table 6.11. Among the 40 tested crosses seven had significant specific combining ability effects, most of these consisting of good general combiner parents. This implies that the observed variation for the MGD was predominantly determined by additive genes together with non- additive genes. Two crosses, 235921 x SRN-39 and 235929 x SRN-39 resulted in the lowest germination distance followed by crosses 235761 x Birhan and 235763 x Birhan. Tester lines SRN-39 and Birhan are known sources of resistance to *Striga*. In crosses involving SRN-39 genetic gains for *Striga* resistance can be realised through continued and simultaneous selection of this trait with improved yield and its components. However, in the other crosses involving tester Birhan, the parents were not good general combiners. The above findings are in agreement with different genetic studies that reported from a single gene to a few genes are involved for *Striga* germination stimulant production on sorghum (Greiner et al., 2001; Mutengwa et. al., 2005; Mohamed et. al., 2010). Haussmann et al. (2000b) and Ejeta et al. (1993) have also reported that the maximum germination distance observed in different *in-vitro* studies are positively correlated with *Striga* resistance under *in-vivo* study but variation was exhibited among different sorghum genetic materials in the amount of stimulant they produce and test locations.

Table 6.10 Analysis of variances for *Striga* maximum germination distances during *in-vitro* tests

Sources of variation	DF	MGD[1] mean squares	
		After 5 days	After 7 days
Replications	2	4.447	5.089
Parents	13	76.317***	105.612***
Parents Vs Crosses	1	258.539***	802.791***
Crosses	39	58.252***	73.298***
Lines	9	63.243*	69.957
Testers	3	302.902**	370.503**
Lines X testers	27	29.405***	41.390***
Error	52	8.040	8.440
CV (%)		28.81	23.20

1: Maximum germination distances

*: significant at 5%; **: significant at 1%; ***: highly significant at 0.01% probability level

Table 6.11 Mean seed germination distance between seven selected sorghum crosses and *Striga* showing ranks, SCA estimates and corresponding GCA effect and their parents on maximum germination distance measured through *in-vitro* tests

Crosses	Mean (cm)	Rank	SCA	GCA of parents	
				P1	P2
235761 X N-13	20.00	7	-4.475*	0.93	4.21**
235761 X Birhan	6.67	3	-5.308**	0.93	-1.16
235763 X Birhan	7.33	4	-6.375**	-1.24	-1.16
235921 X SRN-39	1.67	1	-5.542**	-2.62**	-4.11**
235929 X SRN-39	6.17	2	-4.592*	5.26**	-4.11**
235929 X Hormat	12.67	5	-4.300*	5.26**	1.06
IC 9830 X SRN-39	14.00	6	-3.588*	-2.49*	-4.11**

*: significant at 5%; **: significant at 1%;

6.5 Conclusion

The present study used both field experiments and complementary laboratory tests and found promising sorghum parents and crosses with *Striga* resistance and better agronomic performances. The study identified testers, SRN-39 and Birhan and lines 235761, 235763, IC9830, 235466, 235929 useful for *Striga* resistance breeding in sorghum. Under field experiments, genotypes showed significant interactions with the growing environments. As such their agronomic performances were different across the two locations. Also there were variations on the GCA effects of some lines across locations for *Striga* parameters. Two lines (235761 and IC9830) at Kobo and two (235466 and 235929) at Shewa Robit were isolated as good general combiners for all parameters measured for *Striga*. Significant GCA and SCA effects were observed for different lines across the two testing sites, predominantly with significant GCA effects on agronomic parameters of sorghum. This implies the preponderance of additive genetic effects influencing these traits though non-additive genetic effects also have some degree of contribution. The prevalence of additive genetic effects for maximum germination distance in the current *in-vitro* study was observed for SRN-39, IC9830, 235921 and 235929. Overall, the study identified novel parents and crosses useful for *Striga* resistance breeding or ISM or conservation strategies in sorghum. Further stability analysis and farmers participatory selections at more representative growing environments are needed for release and large scale production.

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CHAPTER 7

Participatory assessment of farmers' and breeders' trait preferences of sorghum genotypes under integrated *Striga* management

7.1 Abstract

Sorghum is the main food crop for millions of subsistence and food-insecure people of sub-Saharan Africa (SSA). In the region sorghum yields are often low due to several constraints, including the devastating damage caused by the parasitic weed, *Striga hermonthica*. Farmers' preferences are critical for successful adoption of improved sorghum genotypes and their production packages, for instance, an integrated *Striga* management (ISM) approach. ISM involves the integration of host resistance and a sorghum seed dressing with a biocontrol agent, *Fusarium oxysporum* f.sp.strigae. The objective of this study was to undertake a farmers' participatory assessment inorder to identify their preferred traits in sorghum genotypes, when grown under ISM, simultaneously with the breeders' evaluation. Forty sorghum hybrids were developed through crosses of 10 *F. oxysporum* compatible and four *Striga* resistant sorghum parents. The seeds of the hybrids and the 14 parents, including one local check variety, were coated with the *Fusarium,* and evaluated at two locations (Kobo and Shewa Robit) known for their high levels of *Striga* infestation in the major sorghum production areas of Ethiopia in 2012. Farmers were requested to participate to identify their preferred traits, and to assess the genotypes using their own selection criteria at crop maturity, and at harvest. The agronomic and *Striga* parameters relevant for breeding were also collected by the breeders. Earliness, *Striga* resistance, yield, seed appearance, seed colour, seed size, and threshability were the most important farmers'-preferred traits in sorghum genotypes. Genotypes were scored based on the farmers' preferred traits and selections made based on mean ranks. Further comparative analyses between farmers' and breeders evaluations revealed highly significant correlations ($p<0.01$) between farmers' preferred and breeders preferred traits except between *Striga* resistance and *Striga* damage, and pest resistance and insect damage. Repeatability of scoring genotypes among farmers was consistent (>0.80) for all traits except *Striga* and pest resistance. The prioritised traits are important for further breeding and release of sorghum genotypes with resistance to *S. hermonthica* and *F. oxysporum* compatibility, which meet the needs and preferences of the farmers.

Key words: *Fusarium oxysporum*, farmers-preferred traits, participatory assessment, *Striga hermonthica*, sorghum

7.2 Introduction

Sorghum [*Sorghum bicolour* (L.) Moench] is the third most important staple food crop in Ethiopia, which largely grown by resource poor farmers. In the country highly diversified sorghum landraces are grown over a wide range of agro-ecologies ranging from 400 to 3000 meters above sea level (Teshome et al., 2007). In Ethiopia smallholder farmers predominantly select and grow landrace sorghum varieties that meet the farmers' requirements for quality traits and adaptation for the crop environments (Mekbib, 2006). In the past, new varieties have been bred and released using different introduced germplasms with to their improved yield, and resistance to pests, diseases, and *Striga*. However, due to farmers' variety preferences, the released sorghum varieties have not seen adopted by many sorghum farmers as expected (Adugna, 2007).

In sub-Saharan Africa there is a declining trend of sorghum productivity, largely because of biotic production constraints such as infestation by the parasitic weed, *Striga hermonthica* (Parker, 1991; Ejeta, 2007; Yemane et al., 2009). *Striga* threatens the livelihoods of millions of smallholder cereal crop farmers throughout semi-arid Africa. It has been estimated that 100 x 10^6 ha of the African savannah zone are infested with *Striga* (Ejeta, 2007). The weed is endemic to sub-Saharan Africa and causes annual crop losses ranging from 30 to 90%, and sometimes leading to complete yield loss (Watson, 2007). In Ethiopia, on average sorghum yield losses of 65% were estimated for moderate to heavy infestations (Tesso et al., 2007). A multitude of *Striga* control options have been studied and recommended including the use of resistant varieties, cultural, chemical, and biological control methods (Hearne, 2008). However, no single option on its own has been effective in sorghum production by resource poor farmers, and the level of adoption of these technologies is very low. Therefore, the use of properly designed integrated *Striga* management (ISM) approach has recently been recommended as a way to provide a long-lasting solution to the *Striga* problem (Kuchinda et al., 2003; Schulz et al., 2003, Aliyu et al., 2004; Tesso et al., 2007; Rebeka et al., 2013).

A pathogenic strain of *Fusarium oxysporum* f.sp.strigae can be an effective component of integrated *Striga* management (Abbasher et al., 1998; Ciotola et al., 2000; Marley et al., 2004; Rebeka et al., 2013). The use of resistant cultivars is also another robust and effective approach to control *Striga,* integrated with other control options. In Ethiopia for the last 15 years, *Striga* resistant varieties have been tested on *Striga* infested experimental plots. Recently a few *Striga* resistant sorghum varieties were released for production in *Striga* prone areas. However, these released varieties have not been well-adopted by the farmers as expected because they lacks attributes that the farmers demand. The combined use of host resistance with the application of *F. oxysporum* as a seed treatment can lead to drastic reduction and elimination of *Striga* plants (Marley et al., 2004; Julien et al., 2009; Rebeka et al., 2013). It would be appropriate to assess this ISM approach involving host plant resistance developed from landraces and for compatibility with *F. oxysporum* with farmers' evaluation. This will be followed by continued pedigree selection breeding method to increase desirable gene combinations for *Striga*.

Knowledge of farmers-preferences is critical for the successful adoption of improved sorghum genotypes, and their production technologies such as the ISM. With respect to sorghum, farmers have complex and diverse selection criteria including high grain yield and resistance to *Striga*, among others. Farmers' participation in setting up research priorities and technology evaluation is crucial to scientists in order to design, test and recommend appropriate new production technologies. This can be achieved through participatory research and evaluation that allows incorporation of farmers' indigenous technical knowledge, identification of farmers' criteria and priorities, and the definition of the research agenda. Application of this information will accelerate the uptake and diffusion of novel technologies. The objective of this study was to undertake farmers' participatory assessment and to identify farmers' preferred traits in sorghum genotype under integrated *Striga* management, simultaneously with the breeders' evaluation. Results of this study may assist in strategic breeding of sorghum with *Striga* resistance and farmers-preferred traits.

7.3 Materials and Methods

7.3.1 Study sites

Field experiments were conducted at two locations, Shewa Robit and Kobo, situated in the North-Eastern lowland areas of Ethiopia where sorghum is predominantly grown. The Shewa Robit site was contracted from a sorghum farmer because of its high level of *Striga* infestation. The Kobo site is in the North Wello administrative zone. This site has a long established *Striga* sick plot which is administered by the Sirinka Agricultural Research Station of the Amhara Agricultural Research Institute. Shewa Robit is located at E 29^{o} 53' and N09^{o}98' and Kobo at E39^{o}37' and N12^{o}09'. Temperatures in the study sites typically range from 17 to 35^{o}C, and the amount of annual rainfall is variable and often low (<1000 mm) and the distribution is often erratic.

7.3.2 Experimental set up

In this study 40 experimental hybrids, 14 parents and one local check cultivar were grown and evaluated. The experiment was laid out using the row-by-column α-lattice design with two replications. Formulated powder of *F. oxysporum* spores was used to treat seeds of the 55 genotypes before planting at the rate of 2.5 g, per plot. Each genotype was planted in three rows of 3m long at 75 and 30 cm inter-row and intra-row spacing, respectively. Fertilizers were applied in the form of diammonium phosphate (DAP) and urea at the recommended rate for the study areas. The plots were weeded as frequently as needed. However *Striga* plants were not removed.

7.3.3 Data collection

7.3.3.1 Farmers' evaluation

A total of 37 farmers (19 from Shewa Robit and 18 from Kobo sites) were participated in the evaluation of test genotypes at maturity and at harvest because these are the two critical stages when farmers usually select the seed source genotypes. At both sites men and women farmers were included. The farmers were selected using prior information from the district agricultural offices, based on their indigenous knowledge and experience on sorghum production. A group discussion was held to discuss and record farmers' selection critera at both crop stages. Each genotype was scored for these traits.

The group discussion identified and listed four attributes needed at the maturity stage, namely: early maturity, high grain yield, resistance to *Striga*, and resistance to pests. At harvest quality traits (seed- size and seed colour) and threshability were identified based on their visual assessment to be important and applied to score each genotype. At both stages, each farmer was asked to rank the traits in order of importance from 1 to 4 (1 being most important and 4 least important trait).

Farmers evaluated each genotype using a checklist of traits. Farmers were also asked to give an overall evaluation of each genotype and to explain their reasons. The farmers were randomly allocated to four groups of at least 5 farmers to rank the 55 sorghum genotypes at full maturity and harvest stages. Each group ranked sorghum genotypes independently through consensus among members of the same group. Farmers' ratings were then translated into numerical values ranging on a scale of 1 to 5 (1 = poor; 2 = fair; 3 = average; 4 = good; 5 = excellent). This process was conducted at crop maturity and at harvest.

7.3.3.2 Breeders' evaluation

A group of four sorghum and one cereal breeders participated in an assessment of same 55 sorghum genotypes. Their ratings of agronomic traits and *Striga* resistance were collected for comparison with farmers' evaluations. The criteria included days to 50% maturity, yield per plot (g/plot), *Striga* damage (1-9) scale, number of *Striga* plants, and level of insect damage.

7.3.4 Data analysis

Data on farmers' evaluations were ranked and scored and agronomic traits assessed were subjected for analysis using the Statistical Package for Social Sciences computer software (SPSS 20.0 Inc., 2011).Trait preference ranking across farmers and sites were summarized using descriptive statistics. Rank indices were calculated using trait preference ranking of individual farmers to provide a weighted average of all rankings for a particular trait according to the following formula adopted from Ceccarelli (2012):

> Rank index = [4 for rank 1 + 3 (rank 2) + 2 (rank 3) + 1 (rank 4)] of an individual trait/ [4 for rank 1+ 3 (rank 2) + 2 (rank 3) + 1 (rank 4)] over all traits.

The non-parametric Mann-Whitney U Test statistics for independent samples was used to test the statistical significance difference of preference rankings of farmers across study sites. Comparison between average scores of sorghum genotypes by farmers and agronomic data were evaluated using the Spearman's rank correlation test procedure. Spearman's correlation analysis is based on the ranks of the data rather than the actual values. Moreover, a reliability test (Cronbach reliability statistics) was conducted to test whether there was consistency between farmers' assessments of genotypes (Carmines and Zeller, 1979).

7.4 Results and discussion

7.4.1 Farmers' selection criteria

The four most desirable traits for selection of sorghum genotypes were identified by farmers each at physiological maturity (Table 7.1) and at harvest (Table 7.2). The traits were ranked by each farmer in order of importance from 1 to 4 (1 being the most important and 4 the least important trait). The rank index, a derived score, indicated the overall importance of a given trait. At physiological maturity, earliness and *Striga* resistance were ranked as the most important traits by farmers at Kobo, followed by high grain yield and pest resistance. Likewise, farmers at Shewa Robit ranked grain yield, earliness and *Striga* resistance in order of importance (Table 7.1). Farmers have excellent varieties mainly landraces which require long growing periods. However, such varieties perform poorly in droughts that regularly occur in these areas. For farmers drought tolerance *per se* was the most difficult trait to describe and to rate for. Instead they used proxy traits such as grain-filling capacity and earliness. Therefore, they were looking for varieties with a short growing period to escape the suffering from late season harsh droughts. *Striga* were also found to be the second important sorghum production constraint in the north and north eastern parts of the country where these study areas are found. Wortmann et al. (2006) also found *Striga* resistance to be one of the most important farmers' preferred traits for selecting sorghum varieties.

The Mann-Whitney U test revealed that farmers at Kobo ranked *Striga* resistance significantly higher than the farmers at Shewa Robit ($P < 0.05$). Conversely, grain yield was ranked significantly higher at Shewa Robit than at Kobo ($P < 0.001$). The varying rankings of the traits might be explained by differences in the relative importance of constraints in these specific sites. Resistance to diseases and pests were the lowest ranked traits at both sites, and this was reflected by the lack of disease or pest outbreaks during the study season.

In general, both the Kobo and Shewa Robit study sites are located in high drought risk and *Striga* infested areas. Both are the most important sorghum production constraints in the sites. Sorghum was being grown by smallholder farmers, predominantly under rainfed conditions, using minimal external inputs. Recurrent drought in these areas precluded the use of sorghum landraces which require a growing period of above six months or longer. Consequently, the farmers' want to access early maturing and *Striga* resistant genotypes as coping strategies against these two production constraints. Similarly other studies in Ethiopia have indicated that farmers prefer sorghum genotypes that display high grain yield, a wide adaption to variable growing environments, and drought stress tolerance (McGuire, 2008; Sinafkish et al., 2010). Thus, assessment of farmers' trait preferences assists to devise informed breeding strategy to accommodate farmers' desire in the newly developed sorghum genotypes.

Table 7.1 Farmers-preferred traits and number of farmers with corresponding ranks and rank indices when rating 55 sorghum genotypes evaluated at Kobo and Shewa Robit sites at physiological maturity

Preference traits	Kobo					Shewa Robit					
	1st	2nd	3rd	4th	Rank index	1st	2nd	3rd	4th	Rank index	P level[a]
Earliness	7	7	2	1	0.32	4	8	6	1	0.28	0.175ns
Striga resistance	10	2	2	3	0.31	2	5	7	5	0.22	0.021*
Yield	0	4	8	5	0.19	11	4	3	1	0.33	<0.001**
Pest resistance	0	4	5	8	0.18	2	2	3	12	0.17	0.573ns

[a] P level = Probability of significance using Mann-Whitney U test.

* and ** denote significant differences among preferences across two sites at the 5% and 1% probability levels, respectively; ns=non-significant.

During harvesting and grain processing, farmers' prefered traits in sorghum genotypes were associated with indirect measurement of sorghum grain food quality such as seed appearance, size, colour and threshability of the head at both sites.

The preference for larger grain size was more important than grain colour at Kobo while farmers at Shewa Robit expressed greater preferences for grain colour (Table 7.2). Farmers had a particular preference for white colored grain for food making. However, white grain sorghum types are more susceptible to bird attack in the field than to red or brown grain types, which are often associated with higher tannin content and are less preferred by birds.

Varied preferences by smallholder farmers for a range of traits in choosing sorghum cultivars are well recognized. Similar studies by Wortmann et al. (2006), McGuire (2008) and Sinafkish et al. (2010) identified farmers' preferred phenotypic characteristics to include plant height, panicle types, grain size, and grain color, environmental adaptability and yield stability. In Ethiopia, the main focus of the formal sorghum breeding program has been the development of early maturing cultivars with high grain yield. These are considered to be the main varietal performance indicators. However, these traits have relative significance and might not meet farmers' selection criteria. Assessment of farmers' preferred traits at an early stage of the breeding program and incorporating of the desired attributes in to the breeding programme would encourage better adoption of new sorghum genotypes by smallholder farmers.

Table 7.2 Farmers-preferred traits and the number of farmers with corresponding ranks and rank indices screening 55 sorghum genotypes, evaluated at Kobo and Shewa Robit sites at the harvesting stage

Preference traits	**Kobo**				Rank index	**Shewa Robit**				Rank index	P level[a]
	1st	2nd	3rd	4th		1st	2nd	3rd	4th		
Threshability	6	4	0	5	0.27	5	8	2	0	0.32	0.595[ns]
Seed appearance	7	6	1	1	0.33	9	5	1	0	0.35	0.486[ns]
Seed size	2	4	7	2	0.24	0	1	3	11	0.13	0.305[ns]
Seed color	0	1	7	7	0.16	1	1	9	4	0.19	0.001**

[a]P level = Probability of significance using Mann-Whitney U test.

** denote significant differences among preferences across two site at the 1% probability level; ns=non-significant.

7.4.2 Rating of sorghum genotypes by farmer groups

The sorghum genotypes were ranked using scores given by farmers for each trait. The average scores for earliness, *Striga* resistance, grain yield, and pest resistance of the 55 sorghum genotypes across study sites are displayed in Figure 7.1 Farmers rated their preferred-genotypes with an overall consideration of such traits. In general the farmers rated and preferred hybrids derived from crosses of genotype SRN-39 at both locations. These hybrids include: 235466 X SRN-39, 235763 X SRN-39, 235921 X SRN-39, 235926 X SRN-39, 235929 X SRN-39, and IC9830 X SRN-39. These genotypes were selected as the best performers for most of the traits considered at maturity. Farmers rated hybrids such as 235466 X Birhan and 235466 X Hormat as the best performers for *Striga* resistance at crop maturity. Hybrids, 235763 X Birhan, 2384443 X Birhan, and 2384443 X SRN-39 were scored highly for their high yield potential at Kobo. Only cross 235929 X SRN-39 was consistently recorded as an excellent performer at both locations for most of the traits farmers considered at maturity.

Furthermore, farmers at Kobo and Shewa Robit rated a given genotype differently except for a few overlaps. Table 7.3 provides the list of sorghum genotypes ranked top for each of the preferred selection traits per farmers' evaluations at Kobo and Shewa Robit. The Mann-Whitney U test showed significant differences for the mean scores of *Striga* and pest resistance ($P < 0.05$) between Kobo and Shewa Robit. However, the mean scores for earliness and yield were not significantly different between sites.

At harvest, most genotypes ranked more than 4 for their threshability in both locations. Genotypes 235921 X Hormat, 235926 X SRN-39, 235929, 237289, 2384443, 2384443 X SRN-39, Hormat, IC9830, and SRN-39 had consistent ranks as good performers for all traits, when considered at harvest, both at Kobo and Shewa Robit.

The frequency distributions of sorghum genotypes with mean score intervals are summarized in Table 7.4. Farmers at Kobo were more stringent in the rating of genotypes for *Striga* resistance than at Shewa Robit. *Striga* infestation and post-flowering drought are most important constraints and thus they were very relevant selection criteria during the study season. Genotypes with mean scores of 4 and above were considered highly by farmers at both Kobo and Shewa Robit, who rated the same genotypes differently, except few overlaps.

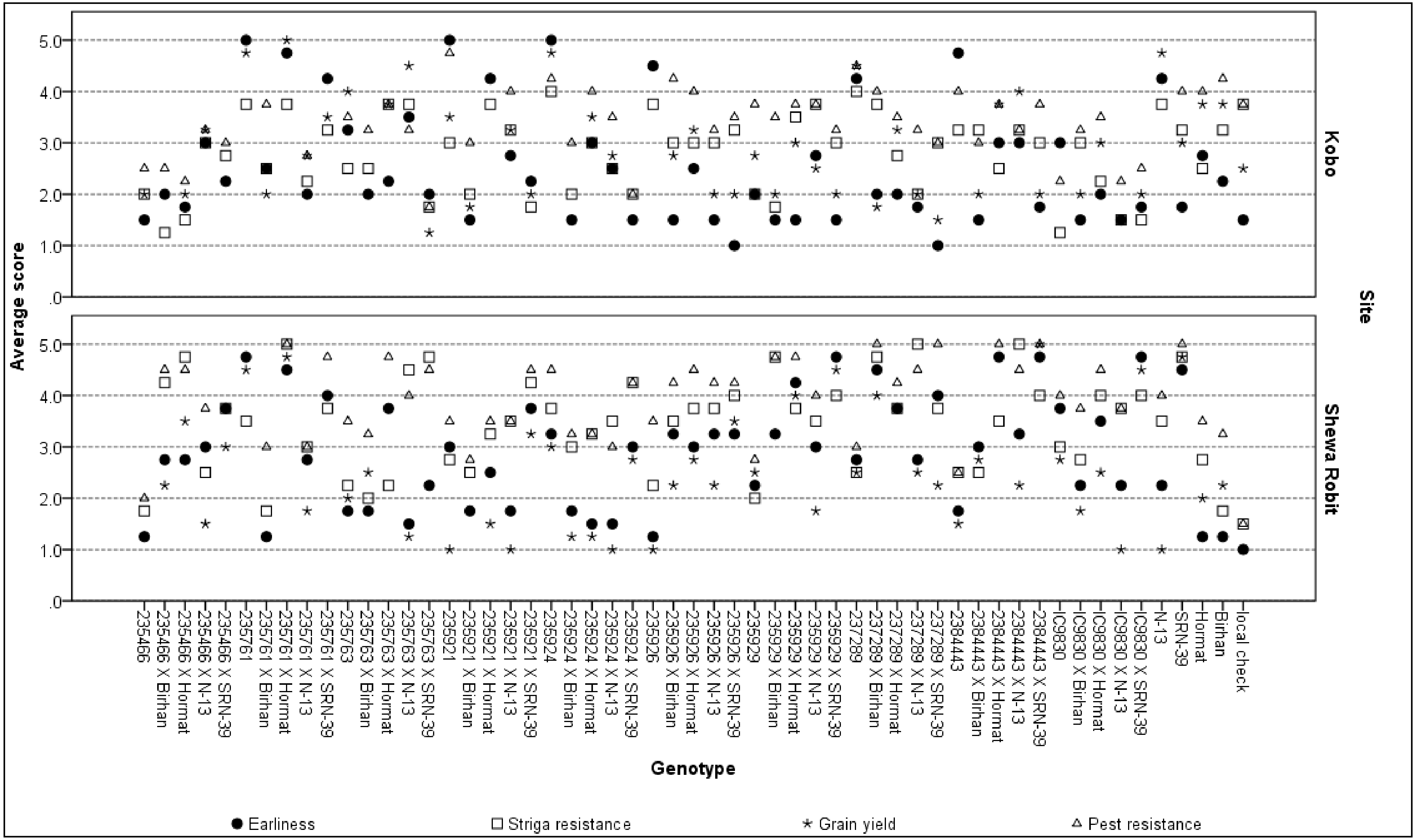

Figure 7.1. Mean score of four traits in 55 sorghum genotypes, evaluated by farmers as selection criteria at crop maturity

Table 7.3 Farmers'-preferred traits and the list of top ranked sorghum genotypes selected by farmers at Kobo and Shewa Robit

Trait	Site		
	Kobo	Shewa Robit	Kobo and Shewa Robit
Earliness	IC9830 237289 X Hormat IC9830 X Hormat 235466 X SRN-39 235763 X SRN-39 235921 X SRN-39 235926 X SRN-39 235929 X SRN-39 237289 X SRN-39 IC9830 X SRN-39	235761 SRN-39 237289 X Birhan 235929 X Hormat 235761 X Hormat 2384443 X Hormat 235929 X SRN-39 IC9830 X SRN-39 2384443 X SRN-39	IC9830 X SRN-39 235929 X SRN-39
Striga resistance	235921 X SRN-39 235929 X SRN-39	SRN-39 235929 X Birhan 237289 X Birhan 235466 X Hormat 235761 X Hormat 2384443 X N-13 237289 X N-13 235763 X N-13 235763 X SRN-39	
Yield	2384443 X Birhan 237289 X Hormat IC9830 X Hormat 235466 X SRN-39 235921 X SRN-39 235926 X SRN-39 235763 X SRN-39 235929 X SRN-39 IC9830 X SRN-39	235761 SRN-39 235761 X Hormat 2384443 X Hormat IC9830 X SRN-39 235929 X SRN-39 2384443 X SRN-39	235929 X SRN-39 IC9830 X SRN-39
Pest resistance	IC9830 235466 X Birhan 237289 X Hormat IC9830 X Hormat 235926 X N-13 IC9830 X SRN-39 235466 X SRN-39 235921 X SRN-39 235926 X SRN-39 235929 X SRN-39 237289 X SRN-39	235761 SRN-39 235929 X Birhan 237289 X Birhan 235761 X Hormat 235763 X Hormat 235929 X Hormat 2384443 X Hormat IC9830 X SRN-39 235761 X SRN-39 235929 X SRN-39 237289 X SRN-39 2384443 X SRN-39	235929 X SRN-39 237289 X SRN-39

Table 7.4 Number (No.) and percentage (%) of sorghum genotypes rated by farmers for each trait at Kobo and Shewa Robit

Trait	Site	Mean score							
		>4-5		>3-4		>2-3		1-2	
		No.	%	No.	%	No.	%	No.	%
Earliness	Kobo	10	18.2	2	3.6	15	27.3	28	50.9
	Shewa Robit	9	16.4	14	25.5	17	30.9	15	27.3
Striga resistance	Kobo	2	3.6	19	34.5	20	36.4	14	25.5
	Shewa Robit	9	16.4	26	47.3	14	25.5	6	10.9
Yield	Kobo	9	16.4	13	23.6	11	20.0	22	40.0
	Shewa Robit	7	12.7	9	16.4	18	32.7	21	38.2
Pest resistance	Kobo	11	20.0	29	52.7	13	23.6	2	3.6
	Shewa Robit	13	23.6	32	58.2	8	14.5	2	3.6

The Cronbach reliability statistic (Carmines and Zeller, 1979) is often used to summarize consistency among groups. This value indicates a good level of consistency if the Cronbach's alpha is 0.8, although an alpha of 0.6 may also be acceptable (Carmines and Zeller, 1979). Different groups composed of men and women farmers scored genotypes consistently (>0.80) for all traits considered at both stages, except for *Striga* resistance (0.591) and pest resistance (0.347) (Table 7.5). The observed disparity of perception among farmers for these two traits (*Striga* and pest resistance) occurred either because of the complexity of these traits or it may confirm the diversity of opinions between farmers groups in the farming systems for these traits. This consistent scoring of genotypes during the two growing stages (maturity and harvest) showed that selection at both stage captured farmers' preferred traits.

Table 7.5 Cronbach Reliability statistics test for the farmers group consistent scoring using important sorghum traits

Traits scored by farmers	Reliability Statistics	95% confidence interval	
		Lower boundary	Upper boundary
Earliness	0.816	0.753	0.867
Striga resistance	0.591	0.450	0.703
Yield	0.774	0.696	0.836
Pest resistance	0.347	0.122	0.526
Threshability	0.943	0.921	0.959
Seed appearance	0.949	0.930	0.964
Seed colour	0.925	0.897	0.946
Seed size	0.891	0.851	0.922

7.4.3 Comparison between farmers' ratings and agronomic data

The Spearman rank correlations coefficients between farmers' ranked traits versus breeders' observations are presented in Table 7.6. In both study areas there was highly significant negative ($p<0.01$) correlation between the farmers' score for earliness and breeders' data for days to 50% maturity of tested genotypes. This negative correlation confirms that farmers scoring early maturing genotypes highly. The correlation between the farmers' score for *Striga* resistance and the number of *Striga* counts by the breeders were also highly significant ($p<0.01$) but not with the *Striga* damage score recorded by the breeder. The non-significant correlation with *Striga* damage is not unusual because of the complex symptoms of *Striga* causes on the crop (usually very similar to disease symptom). The symptoms of *Striga* damage are not easily recognised by farmers. In Shewa Robit, there was also a significantly high correlation ($p<0.01$) between farmers' score for yield potential and measured grain yield. Despite the precarious food insecurity at Kobo, farmers prefer sorghum varieties which fulfil all their preferences and not only a high yield. However, breeders tend to focus on selecting high yielding varieties for food security. As such there was no significant correlation between the farmers' and breeders choice of varieties. At both sites, there was no significant correlation between the score given to pest resistance by the farmers' and the insect damage score measured by the breeder.

It is not surprising to observe the lack of correlation between the yield score by the farmers at Kobo and breeders' measured yield data because farmers' selection decisions are often influenced by their need for multiple traits in stressed growing conditions which are unlike the breeder's well-managed experimental data. Various authors have reported similar disparity between farmers' assessment for crop performance and breeders observation (Weltzien et al., 2005; Brocke et al., 2010). Wale and Yallew (2007) noted that improved variety development often lack the fitness attributes that farmers' prefer. Under harsh growing conditions, diseases and pest infestations hinder the adoption rate of the breeders improved varieties because they are less well adapted than landrace varieties. Thus, a balance between farmers-preferred traits and solutions to production constraints should be the breeders' goal in order to enhance the uptake of new varieties by farmers.

Table 7.6 The Spearman rank correlation coefficients between farmers' ranking based on their preferred traits versus breeders' observation

Farmers' traits score	Study sites	Traits measured by breeders				
		Days to 50% maturity	*Striga* damage score	Number of *Striga*	Grain yield per plot	Insect damage score
Earliness	Kobo	-0.816**	--	--	--	--
	S.Robit	-0.396**	--	--	--	--
Striga resistance	Kobo	--	-0.030*	0.364**	--	--
	S.Robit	--	-0.166^{ns}	0.406**	--	--
Yield potential	Kobo	--	--	--	0.192^{ns}	--
	S.Robit	--	--	--	0.365**	--
Pest resistance	Kobo	--	--	--	--	0.129^{ns}
	S.Robit	--	--	--	--	-0.159^{ns}
Total	Kobo	-0.663**	-0.152^{ns}	0.293**	0.105^{ns}	0.064^{ns}
	S.Robit	-0.421**	-0.308^{ns}	0.184**	0.192*	-0.225^{ns}

** = Correlation is significant at the 0.01 level;*= Significant correlation at 0.05 level, ns=non-significant

7.5 Conclusion

Sorghum is an important food security crop growing in heterogeneous and marginal environments in Ethiopia. Despite 25 years of sorghum breeding in the country, none of the many released 'superior' cultivars have been widely adopted by the farmers. This study detected that farmers preferred varieties that were able to withstand a variety of stresses. Further, these varieties should possess desirable traits to meet their diverse needs over and above grain yield *per se,* which are often the primary goal of sorghum breeding programmes.

The study focused on early stage evaluation of 55 sorghum genotypes (40 F_1 hybrids and 15 pure lines) with farmers' participation in two contrasting environments. Sorghum farmers' know their growing environments and their social needs. Farmers' selection criteria for sorghum genotypes are often more multivariate than breeder's who usually aim to improve a few selected traits. Earliness, *Striga* resistance and high yield were the preferred traits of farmers at the maturity stage. Sorghum seed appearance, seed size, colour and threshability were the most important desired traits at the harvesting stage.

Farmers were also able to select genotypes in relation to their environmental conditions. There was more or less consistent scoring for the desired genotypes between farmers groups. Overall, involvement of farmers during the early selection phases of sorghum breeding through participatory assessment is essential for successful adoption of improved sorghum genotypes and new technology, including integrated *Striga* management (ISM).

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CHAPTER 8

An overview of the research findings

8.1 Introduction and objectives of the study

Sorghum is one of the major food security crops supporting some 500 million people in sub-Saharan Africa who survive under harsh environmental conditions. The yields of the crop are significantly reduced in much of Africa due to the devastating damage caused by the parasitic weed, *Striga*. Thus so far a range of *Striga* control options have been identified and recommended. However, when each control method is used separately, then it is not effective, and is often affected by environmental conditions. Therefore, several control options need to be integrated for successful and sustainable *Striga* control. Biological control of *Striga* using selected strains of *Fusarium oxysporum* f.sp.strigi can be integrated along with resistant sorghum genotypes for Integrated *Striga* Management (ISM). *Striga* causes pronounced crop damage before its emergence from the soil. Consequently, treatment of seeds of resistant sorghum genotypes with *F. oxysporum* could provide early control and reduce the *Striga* seed bank in the soil. This chapter highlights the study objectives with subsequent summary of major findings of each objective. Finally, the implications of the findings are presented for sorghum breeding to *Striga* resistance and improved agronomic attributes according to the needs of the growers.

The objectives of this study were initially established as to:

- determine farmers' sorghum production opportunities, threats, indigenous knowledge and perceptions, emphasising breeding priorities and *Striga* infestation, and the coping mechanisms of farmers in the north eastern and north western Ethiopia,
- evaluate sorghum genotypes for compatibility to *F. oxysporum* inoculation under *Striga* infested soil in controlled environments,
- determine the field responses of the sorghum genotypes for *F. oxysporum* compatibility as a basis for ISM,
- determine the variability present among selected sorghum genotypes exhibiting *S. hermonthica* resistance, and compatibility with the biological

control agent, *F. oxysporum*, using phenotypic and simple sequence repeat (SSR) markers,

- identify *F. oxysporum* compatible parents and hybrids with high combining ability for grain yield, yield components, and *Striga* resistance for ISM, and
- undertake farmers' participatory assessment and identify preferred traits of sorghum genotypes under integrated *Striga* management simultaneously with the breeders' evaluation.

8.2 Research findings in brief

8.2.1 A diagnostic appraisal of the sorghum farming system and breeding priorities in *Striga* infested agro-ecologies of Ethiopia.

- A participatory rural appraisal (PRA) research was conducted involving 315 farmers at nine districts of three administrative zones within two provinces in Ethiopia.
- Important sorghum production constraints were identified and prioritized, of which drought and *Striga* damage were the most significant factors limiting sorghum production and productivity.
- Farmers-preferred sorghum landraces were identified and characterized based on farmers' indigenous knowledge. Farmers' preferences and reasons towards continued use of landraces instead of improved superior sorghum varieties were documented. These included reasonable grain yield, tall plant height and straw yield, the ability to grow in harsh environments, and pest and disease resistance.
- Farmers predominantly practice hand weeding as the major *Striga* control option but they realised that this method was not effective for *Striga* management which needing a better control option.

8.2.2 Evaluation of sorghum genotypes compatibility with *Fusarium oxysporum* under *Striga* infestation.

- Fifty sorghum genotypes were evaluated for their *Fusarium* compatibility under controlled environments in a greenhouse and laboratory through artificial *Striga* infestation condition.

- Controlled greenhouse study revealed that sorghum seed treatment with *F. oxysporum* reduced *Striga* counts by up to 92% and increased yield by 144% than untreated control.
- Sorghum genotypes grew faster and matured 14 days earlier than comparative controls when seeds were treated wih *Fusarium.* The root rhizosphere of *Fusarium* compatible sorghum genotypes enhanced growth and development of *F. oxysporum* confirmed from the complementary laboratory analyses. This was associated with increased number of colony forming units counted from soil and sorghum plant root samples.
- Twelve sorghum genotypes (235763, 235927, 237289, 235466, IC9830, 235924, 235921, 2384443, 235929, 243684, 235761, and 235926) with better *F. oxysporum* compatibility, supporting no or less number of *Striga* plants, with relatively higher seed yield and suitable agronomic traits, were identified as promising breeding parents for integrated *Striga* management.

8.2.3 Field evaluation of sorghum genotypes through *Fusarium oxysporum* application against *Striga hermontica.*

- Field experiments were conducted in three different agro-ecologies of Ethiopia, namely, Kobo, Sirinka and Shewa Robit, which are known for their severe *Striga* infestation. The responses of fifty sorghum genotypes to F. *oxysporum* inoculation were assessed.
- Field experiments confirmed the prior results conducted under controlled conditions and showed clear growth differences among sorghum genotypes when seeds were treated with *F. oxysporum*.
- Sorghum genotypes such as 235763, 235927, 237289,235466, IC9830, 235924, 235921, 2384443, 235929, 243684, 235761, and 235926 were selected for their compatibility with *F. oxysporum* and for better agronomic performances, confirming prior controlled studies. These entries showed similar and positive responses in the field experiments across all study sites.
- Sorghum genotypes including 235763, 235924, and 235929 had excellent *F. oxysporum* compatibility and agronomic performances at all study sites, and were selected for ISM.

- Sorghum genotypes including 235921, 235927, 237289, 239235, 243684, and IC9830 at Kobo and Sirinka sites; 235761 and 2384443 at Kobo and Shewarobit sites; 235466, 235467, 235925, 238402, 238436, 238439, 238441, 238449, 239236 at Kobo site only and 212541 and 235926 at Sirinka site showed better *F. oxysporum* compatibility and agronomic performances, suggesting site specific genotype selection.

8.2.4 Assessment of genetic diversity of sorghum resistant to*Striga hermonthica* and compatibile with *Fusarium oxysporum* using phenotypic and SSR markers.

- Significant phenotypic variability was observed among 14 selected sorghum genotypes for traits including days to 50% emergence, maturity and flowering, plant height, panicle length, head weight and, seed yield.
- Genotypes were clustered into two distinct phenotypic groups, one cluster comprised six genotypes with good *F. oxysporum* compatibility, and two genotypes had *Striga* resistance. The other cluster consisted of six genotypes which were *Fusarium* compatible and two genotypes with *Striga* resistance, both acquired from ICRISAT (SRN-39 and N-13).
- The SSR markers showed a high level of polymorphisms among genotypes, with the mean number of alleles per locus being 6.95 and the mean polymorphic information content being 0.80.
- The SSR markers also allocated genotypes into two distinct clusters, which were comparatively similar to the grouping, developed using phenotypic markers.

8.2.5 Combining ability for grain yield and *Striga* resistance in sorghum [*Sorghum bicolor* (L.) Moench].

- Forty F_1 sorghum genotypes were developed using a line by tester mating scheme, where 10 Fusarium compatible female parents were crossed with four *Striga* resistant genotypes as males. The 40 hybrids and 14 parents were evaluated for their combining ability at two areas known to be *Striga* infested, namely Kobo and Shewa Robit, in Ethiopia.
- Sorghum hybrids derived from crosses of *F. oxysporum* compatible and *Striga* resistant genotypes indicated the preponderance of general combining ability

(GCA) effects on most agronomic parameters. This implies that most traits were controlled by additive genetic effects although non-additive genetic effects had a low level of contribution.

- The study identified tester lines SRN-39 and Birhan and lines 235761, 235763, IC9830, 235466, 235929 as the best general combiners useful for *Striga* resistance breeding in sorghum.
- There were variations on the GCA effects in some lines across locations for *Striga* parameters. Two lines (235761 and IC9830) at Kobo and two (235466 and 235929) at Shewa Robit were identified as good general combiners for all parameters measured for *Striga* resistance.
- Significant ($p<0.05$) specific combining ability (SCA) effects were detected for the traits days to 50% flowering, biomass, grain yield and number of *Striga* plants.
- At Kobo, cross 235763 x N-13 and Shewa Robit IC9830 x SRN-39 had significantly negative SCA effects on the numbers of *Striga* plants. These crosses were selected for use in the subsequent breeding program.
- From a complementary laboratory *in-vitro* agar assay experiment, the prevalence of additive genetic effects for maximum germination distance was confirmed for genotypes SRN-39, IC9830, 235921 and 235929.

8.2.6 Participatory assessment of farmers' preferences of sorghum genotypes under integrated *Striga* management.

- Forty sorghum hybrids and 14 parents were coated by the *F. oxysporum* and grown and evaluated by growers and breeders at two locations.
- Earliness, *Striga* resistance, grain yield, grain quality and threshability were the most important farmers'-preferred traits for sorghum genotypes.
- Farmers used the aforementioned traits to score and select the following crosses; 235466 X SRN-39, 235763 X SRN-39, 235921 X SRN-39, 235926 X SRN-39, 235929 X SRN-39, and IC9830 X SRN-39.
- Congruent analyses between farmers' and breeders evaluations revealed highly significant correlations ($p<0.01$) between farmers'-preferred and breeders assessed traits, except for *Striga* resistance and *Striga* damage ,and pest resistance and insect damage.

- The selected crosses will be subjected for continued selections to identify and release high and stable yielding and *Striga* resistant pure line sorghum cultivars.

8.3 Implications of the study for breeding and integrated *Striga* management

- The participatory rural appraisal study showed that farmers preferred to grow landraces than improved introduced varieties necessitating targeted breeding using both germplasm sets. There is a need to exploit the genetic potential of existing landraces and exotic genetic resources in sorghum improvement programme in Ethiopia, but also to meet the farmers' requirements.
- In the past, adoption rate of improved varieties by smallholder farmers has been low in Ethiopia. This was due to a mismatch between sorghum breeding goals aimed primarily at improved yield and farmers' preferred attributes such as straw yield, drought tolerance and *Striga* resistance. This shows the need to have a consolidated breeding objective incorporating farmers-preferred traits, and involving farmers in the entire breeding process such as through participatory evaluation and selection.
- Farmers practice hand weeding to control *Striga* like other weed species because of a lack of information, and limited access to other technologies. There is a need for the various available technologies should be made accessible to growers, and enhancement for the linkage between research and agricultural extension is essential.
- *F. oxysporum* seed treatemnt showed high efficacy in reducing *Striga* damage and improving sorghum yield under both controlled and field conditions. Therefore, integration of this biocontrol agent with resistant sorghum genotypes could play a crucial role to reduce the *Striga* seed bank and to control this noxious weed on a routine basis.
- Sorghum genotypes compatible with *F. oxysporum* were identified among the landrace collections of Ethiopia, providing the possibility of exploiting the genetic resources for breeding towards integrated *Striga* management. Adequate genetic variability was observed through phenotypic and molecular characterizations.

- Both additive and non-additive gene effects were detected as controlling sorghum agronomic traits and *Striga* resistance related parameters which is useful for further selection and hybridization programs developed to exploit heterosis.

8.4 Possible challenges in the deployment of ISM in Sorghum

- **Mass production of the bio-control agent (*F. oxysporum*)**: this would be one of the possible challenges to implement and scale up the technology. Among these challenges the most important ones include: (1) the unavailability of local capacity to mass produce the Biocontrol agent through a starter culture, (2) the stringent controls by the local regulatory authority on the importation mass of reproductions or any formulations of the bio-herbicide from external sources. The isolate used in this study was originally isolated from severely diseased *Striga* samples in Ethiopia and the mass formulation of the fungus was done at Plant Health Products (pty) Ltd, South Africa which has presently the required technology and production capacity in Africa. Unlike chemical herbicides, the fungal spores (bio-herbicides) are living organisms, thus the reason for strict regulatory measures for introduction. These are the factors needing considerations for successful scaling up and wide application of the bio-herbicide including other international markets.
- **Delivery system**: the seed coating delivery system is an efficient method for uniform application of *Fusarium*. However, through the existing extension and seed supply system, providing access for sorghum seeds after treating with the bio-agent could be one of the challenges. Farmers should be educated about the technology through formal local extension services moreover the seed system should be integrated with this novel approach.
- **Incorporation of the ISM in to the current sorghum breeding programs: ISM** shows promise to boost sorghum productivity through synergistic use of *F. oxysporum* and host resistance. The success of the ISM, however, is contingent upon its incorporation and acceptance into the present *Striga* resistance breeding of sorghum in Ethiopia.