MAKERERE UNIVERSITY

STUDIES ON GENETIC DIVERSITY, GENOTYPE BY ENVIRONMENT INTERACTION, COMBINING ABILITY AND FARMERS' PERCEPTION ON SWEET SORGHUM (*Sorghum bicolor* (L.) Moench)

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DECLARATION

The work presented in this thesis is my own research and to my knowledge it has not been presented for the award of degree or diploma in any other University.

Signed……. Date…….

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This thesis has been submitted for examination with our approval as the university supervisors.

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PREFACE

This thesis is based on the following published and draft research articles

1. Olweny. C. ,Ong'ala, J. Dida, M.M.,Okori, P. (2013).Farmers' perception on sweet sorghum (*Sorghum bicolo*r [L] Moench) and potential of its utilization in Kenya. World Journal of Agricultural Sciences Vol. 1(2), pp. 065-075

2. Olweny. C, Jamoza. J, Kimani. W, Yao. N, Njuguna. J, Githae. D, Kiawa. B, Kosambo.L, Chikuta. S, Dida, M.M, Okori.P. (2014). Assessment of genetic diversity for improvement of sweet sorghum (*Sorghum bicolor* (L.) Moench) genotypes for sugar and allied products Molecular Plant Breeding, Vol.5, No. 6 29-35 (doi: 10.5376/mpb.2014.05.0006)

3. Olweny.C, Dida, M.M., Abayo.G, Okori, P. (2015). Genotype × Environment interaction on sugar and biomass production in sweet sorghum (*sorghum bicolor (*L). Moench) in western Kenya. Draft article.

4. Olweny.C, Dida,M.M., Abayo.G, Okori, P. (2015).Combining ability of parents and hybrids for sugar and its attributing traits in sweet sorghum [*Sorghum bicolor* (L.) Moench]. Draft article.

ABSTRACT

Sweet sorghum (*Sorghum bicolor* (L.) Moench) is a cultivated sorghum recognized as potential alternative source of bio-fuel due to its high fermentable sugar content in the stalk. Sweet sorghum has elicited the interest of breeders due to its capacity to provide renewable energy products such as biofuel, industrial commodities, food and animal feed. The constraints for its large scale cultivation are the limited availability of genotypes suited to different agro-climatic conditions. There is limited information about the combining ability, gene action and genetic effects on stem sugar and biomass traits which is required in formulating appropriate strategies for developing super performing sweet sorghum varieties. Sweet sorghum gene pool creation has also not received much attention mainly because it is not considered to be among important crops in Kenya, and the pedigree information is incomplete. Furthermore, there is also lack of information about the perceptions of resource-limited, small-scale farmers on the potential of sweet sorghum and feasibility of its utilization in Kenya. Therefore, the objectives of this study were to: (i) determine farmers perceptions on sweet sorghum and potential of its utilization in Kenya (ii) investigate the influence of genotype by environment interaction on sugar and biomass production of sweet sorghum (iii) assess genetic diversity and relationship among a collection of sweet sorghum germplasm by simple sequence repeats (SSR) markers and (iv) determine the combining ability in respect of stem sugar and biomass traits in sweet sorghum.

A survey was undertaken in Western and Coastal regions of Kenya to determine farmers' perception on sweet sorghum and feasibility of its utilization. Descriptive and inferential statistical tests were used to analyze the data. It was observed that 72.9% of the respondents were male, 95.7% married and 46% had formal education. Most of the farmers (73.0%) cultivated below 2 acres of farmland. About 40% of the respondents were aware of existence of sweet

sorghum varieties while 50% of them were aware of sweet sorghum processing technologies. The study revealed that farmers appreciate the potential of sweet sorghum and existence of capacity for its exploitation.

To assess of genetic diversity and relationship among a collection of sweet sorghum germplasm using simple sequence repeats (SSR) markers, eighty six sweet sorghum cultivars from Argentina, Brazil, Kenya (ICRISAT and Moi University), United States of America and Zambia were genotyped with 11 SSR markers that generated 86 alleles with an average of 8 alleles per locus. Polymorphism information content (PIC) value was 0.53 indicating a moderate diversity with a range of 0.09–0.89. The variability among the populations was low at 3 % but amounted to 22% and 75 % within individual genotypes and among individuals respectively. Clustering analysis based on the genetic similarity (GS) grouped the 86 sweet sorghum genotypes into 2 distinct clusters. The study also revealed the genetic relationship of cultivars with unknown parentage to those with known parentage. Information generated from this study can be exploited to select parents for hybrid development to maximize sugar content and total biomass and for development of segregating populations to map genes controlling sugar content in sweet sorghum.

To investigate the influence of genotype by environment interaction on sugar and biomass production of sweet sorghum, field experiments were conducted to evaluate sweet sorghum genotypes in Western Kenya during the 2011, 2012 and 2013 rainy season of April to August at Alupe, Kibos, Homa Bay and Spectre International farm. The materials used in the study consisted of sixteen sweet sorghum genotypes and two sorghum genotypes sourced from ICRISAT and KARI. The treatments were laid out in a Randomized Complete Block Design (RCBD) and replicated three times. Data were collected on sorghum traits in accordance with the procedure outlined in the ICRISAT sorghum descriptor. The study revealed that genotype by

environment interaction had significant influence on most of the traits. This indicates that selection for plant height, girth, brix juice, juice volume and stalks weigh cannot be carried out across the four environments, suggesting that selection for these traits have to be carried separately in each of the four environments. High performance demonstrated by genotypes IESV 93046 and IS2331 for stem brix and stem biomass shows their potential for exploitation for ethanol production.

On the combining ability study, an investigation was carried out to assess the combining ability and nature of gene action in respect of sugar yield and its attributing traits in 25 new hybrids of sweet sorghum developed by crossing five (5) high sugar lines with five (5) low sugar lines in a North Carolina II mating design and grown in alpha lattice with two replications during long rains of April to July 2014 in western Kenya. The variance among the lines in respect of their general combining ability (GCA) was highly significant for Brix and plant height at 90 days. Specific combining ability (SCA) variance was relatively higher in magnitude for grain weight and plant height indicating predominance of non-additive gene action in the genetic control of these traits. GADAM, MALON and PAISANO among the females and IESV93036, IS2331 and NTJ 2 among males were identified as good general combiners indicating their ability in transmitting additive genes in the desirable direction to their progenies. The best hybrids for total biomass and total sugar content were GADAMxIESV93036, GADAMxIS2331 and MALONxIS2331 and after adequate testing in many locations across the target production environments, these hybrids can be recommended for commercial exploitation for ethanol production.

Overall, the study showed that development of sweet sorghum cultivars and hybrids is feasible and genotypes identified as potential cultivars can be exploited for ethanol production.

DEDICATION

This work is dedicated to my late father, Hannington Olweny Orori for the myriad of ways in which, throughout my life, he actively supported me in my determination to find and realize my potential, and to make this contribution to our world.

LIST OF ABBREVIATIONS AND SYMBOLS

% Percentage

< Less than

> Greater than

AMMI	Additive main effects and multiplicative interaction
AMOVA	Analysis of molecular variance
ANOVA	Analysis of variance
CTAB	Mixed alkyl-trimethyl-ammonium bromide
C. V	Coefficient of variation
ddH_2O	double-distilled water
dNTPs	deoxynuleoside 5' –triphosphate
DNA	Deoxyribonucleic acid
ICRISAT	International crop research institute for semi arid tropics
F – STAT	Computer Programme to analyze F- statistics
GGE	Genotype plus genotype by environment interaction
GLM	General linear model
KARI	Kenya Agricultural Research Institute
KESREF	Kenya Sugar Research Foundation
LSD	Least significant difference
PAGE	Polyacrylamide gel electrophoresis
PCA	Principal coordinate analysis
PCR	Polymerase chain reaction
QTL	Quantitative trait loci
RAPD	Random amplified polymorphic DNA
REML	Restricted maximum likelihood
RFLP	Restriction Fragment Length Polymorphism

SAS	General software package for statistical analysis
SSR	Simple sequence repeat
UPGMA	Unweighted pair group method with arithmetic averages
g	gram
kb	kilo bases
min	minute
ng	nanogram = 10^{-9} gram
RNA	ribonucleic acid
RT	room temperature
sec	second
SGB	sample gel buffer
TAE	Tris –acetate EDTA (buffer)
U	unit of enzyme
UV	ultra violet
V	volts
^{0}C	degree Celsius
ug	microgram = 10^{-6} gram
ul	micro litre = 10^{-6} litre
cM	Centimorgan

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CHAPTER ONE

1.0 GENERAL INTRODUCTION

1.1. Background

Sorghum (*Sorghum bicolor* (L.) Moench) is one of the crops that are traditionally produced in marginal areas, where low soil moisture and high ambient temperatures are the main limiting abiotic factors (Gebeyehu *et al*., 2004).Sweet sorghum is a C_4 plant (Sipos *et al*., 2009) characterized by a high photosynthetic activity. Sweet sorghum is resistant to drought and flooding (Tesso *et al*., 2005). Sugars in sweet sorghum are made up of 85 % sucrose (Woods, 2000). Sweet sorghum sugar yields range between 1.6 and 13.2 tons ha^{-1}, with significant variations observed between different years and regions (Zhao *et al*., 2009; Tamang, 2010). The juice sugar content is depended on the plant stage of development. At the early development stage, fructose is more abundant and sucrose is dominant after heading (Sipos *et al*., 2009). At maturity, the sweet sorghum juice sugar content can range from 10 to 25 brix (Reddy *et al*., 2007) At the global level, more than 35% of the sorghum produced is directly used for human consumption with the rest used for animal feed, production of alcohol and industrial products (Awika and Rooney, 2004). The high demand for water coupled with climate change is limiting expansion of sugarcane production zones in many countries of sub-Saharan Africa. Sweet sorghum, a close relative of sugarcane could serve as an alternative source of these products because it accumulates large amounts of sugar in their stems (Hunter and Anderson 1997). Sweet sorghum has potential for cane yield of 40 t/ha. Near physiological maturity, sweet sorghums have 10 to 25% sugar in stalk juice, with sucrose being the predominant disaccharide (Hunter and Anderson 1997). Sweet sorghums are distinct due to higher sugar content in the stalks (Brix 10–18%) from flowering to maturity than that of grain sorghum (Brix 9–11%) during the same period

(Srinivasa Rao *et al*. 2009).The sugar in the stalk juice of sorghum can be industrially utilized to produce crystal sugar, and ethanol for biofuel, jaggery and syrup. Stillage, the leftover stalks after juice extraction, can be used for power generation, animal feeds as well as organic manure.

Sorghums have lower nutritional and moisture requirements for growth than sugarcane, a traditional source of sugar and allied products. Sorghum's water requirement during a growing period of about 4 months is about 1000 m^3 per month, about one-third that of sugarcane's requirement of 36000 m^3 per crop that requires 12-16 months (Soltani and Almodares, 1994).While sugarcane is propagated from stem cuttings, sweet sorghum is sown with seed, i.e., just 4.5 kg is enough for a hectare of land, compared to 4,500-6,000 kg of sugarcane cuttings. The cost of cultivation of sweet sorghum is estimated to be one-third that of sugar cane. Sweet sorghum could therefore used as an alternative crop to sugarcane in drier lands for the production of sugar and allied products. Yet the exploitation of this valuable crop is limited by few accessions and no breeding programmes to continuously supply high yielding sweet sorghum varieties. The limited supply and increasing demand for sugar therefore indicates an urgent need to seek alternative sources of sucrose for industrial and domestic uses. Sweet sorghum can bridge that gap.

The increase in the sugar content of sorghum stalk will also enhance its palatability and the forage quality (Blummel *et al.* 2009). Therefore, sweetness along with juiciness and biomass are the important targets in sweet sorghum breeding. Its potential, as a sugar source for bio-ethanol production at industrial scale has not been fully exploited owing to the availability of raw material for a limited period in any given year resulting in the poor performance of commercial ethanol distilleries. It is therefore, necessary to increase the window of sweet stalk availability, which can either be achieved by breeding cultivars that mature at varying periods or by identifying cultivars

that sustain high sugar levels over a longer period of time even after reaching maturity (Reddy *et al*. 2005; Srinivasa Rao *et al*. 2009). A few cultivars like Brandez, Wray and ME 84-1 have been identified to possess sustained sugar levels for a longer period (between 25 and 40 days) for industrial utilization in Brazil (Schaffert, EMBRAPA, personal communication). Studies aimed at determining hexoses at physiological maturity (Smith *et al.* 1987) established that sucrose is major component of sugars followed by glucose and fructose in sweet sorghum juice.

The success of a breeding programme depends on well characterized breeding populations, yet very little information is available on the genetic heterogeneity of sweet sorghum lines collections in Kenya. Recent advances in sorghum genomics provide new frontiers for the application of information and better tools for breeding and scientific studies. Studies have shown molecular differences between sweet sorghum and grain sorghum (Calvino *et al.* 2009). Information generated from diversity study can be used to select parents for hybrid development to maximize the sugar content and total biomass, and development of segregating populations to map genes controlling sugar content in sweet sorghum.

Success of any crop-breeding program is based on the knowledge of and availability of genetic variability for efficient selection. Genetic similarity (or genetic distance) estimates among genotypes are helpful in selecting parental combinations for creating segregating populations so as to maintain genetic diversity in a breeding program (Becelaere *et al*. 2005) and the classification of germplasm into heterotic groups for hybrid crop breeding (Menz *et al.* 2004). The search for and establishment of heterotic groups can be based on geographical origin, agronomical traits, pedigree data or on molecular marker data (Melchinger 1999). Before the advent of molecular genetics tools, genetic diversity was estimated from pedigree or agronomic and morphological characteristics. However, the estimates based on pedigree information are

generally inflated and often found unrealistic (Fufa *et al*. 2005; Almanza-Pinzon *et al*. 2003; Cox *et al.* 1986). The morphologically based genetic diversity estimates suffer from the drawback that morphological characteristics are limited in number and are influenced by the environment (van Beuningen and Busch 1997). Therefore, neither pedigree-based nor morphologically- based estimates may reflect the actual genetic difference of the studied populations. On the other hand, molecular markers are not influenced by environment, reflect genetic similarity (and differences), and do not require previous pedigree information (Bohn *et al*. 1999) which is valuable for crops where pedigree information is lacking. Various types of molecular markers are available for genome analysis in the grass family. Simple sequence repeats (SSRs) in particular have been reported to be very useful to analyze the structure of germplasm collections as these are abundant, codominant, multiallelic, highly polymorphic and chromosome-specific (Ahmad 2002; Huang *et al.* 2002; Parker *et al.* 2002).

The origin and pedigree information for many of the introductions and derived cultivars are not available or poorly documented. Hence, a study on genetic relationship among sweet sorghum cultivars (with and without parentage information) will help determine their genetic relationships. Molecular markers have been extensively used in genetic diversity studies in many plants, including wheat (Triticum aestivum L.; Fufa *et al.* 2005; Mahmood *et al*. 2004), pearl millet (Pennisetum glaucum L.; Budak et al. 2003), sorghum (Casa *et al.* 2005; Smith *et al.* 2000) and triticale (Kuleung *et al*. 2006).

The sweet sorghums have not been a major focus of commercial breeding programmes; hybrids have been developed between grain and sweet sorghums, usually for fodder or dual purpose use (grain and fodder). Thus, increasing stalk sugar yields is becoming an important objective in sweet sorghum breeding (Murray *et al*. 2009).

In sweet sorghum, genetic improvement of sugars has not been intensively studied compared to that of sugarcane. Papini-Terzi *et al*. (2009) reported that pathways for sucrose content are overlapping with drought and other abiotic stress response pathways thus making sucrose yield a complex trait. Genetic enhancement of the crop for increased sugar yield is very critical to make sweet sorghum more profitable to the farmers and the industry, while sustaining grain yield, juice volume, plant height, plant girth and other important components. The choice of an efficient breeding programme depends largely on knowledge of the type of gene action involved in the expression of the character. The knowledge on nature of gene action for sugar yield and its component traits like brix% and juice content in the breeding material can provide useful information for selecting proper breeding procedure for future genetic enhancement. Inheritance of stalk biomass, brix percentage and stalk weight in sugar stalk was subject to both additive gene effect and non-additive gene effect, but mainly controlled by non-additive genes Zhou *et al*. (2005). However, the literature regarding inheritance of these traits and their genetic interactions in sweet sorghum is scanty. Keeping this in view, an attempt has been made to understand the gene action controlling sugar yield and its component traits using different lines of sweet sorghum with varied brix% and juice content.

Several national research programs in the semi-arid regions have shown an increased interest in hybrids (Axtell *et al.,* 1999). The immediate task that faced those breeding programs is to gain information on the combining ability of the various varieties and populations developed and improved over the years. Information on combining ability is needed to identify potentially superior parents and hybrids, and would help to define the pattern of gene effects in the expression of quantitative traits (Goyal and Kumar, 1991).

1.2. Statement of the problem

Farmers' subjective assessments of agricultural technologies influence adoption behavior (Nowak, 1992). Understanding the farmers is an initial step towards the search for an effective and sustainable way to make agricultural research more relevant to them (Kudadjie *et al.,* 2004). Many researchers have reported on the negative consequences of not including farmers in setting up research and policy agenda (Derera *et al*., 2006). National and international research centers have reported significant yield increases in many crops but farmers remain unaware and have low perception of the skills to take full advantage of these technologies (Ekpere, 1995).

The size of high potential agricultural land in Kenya is dwindling as population increases. This leaves large expanses of low potential and semi arid lands that cannot support the production of key food crops such as wheat, rice and maize. Demand for renewable energy sources and biofuel, which would minimize pollution, is expected to rise rapidly in the coming years (Belum *et al*., 2007). Sweet sorghum by virtue of its C_4 photosynthetic system and rapid dry matter accumulation serves as an excellent bioenergy crop. The constraints for its large scale cultivation are the limited availability of genotypes suited to different agro-climatic conditions with all built-in resistances for biotic and abiotic stresses, photoperiod sensitivity and non –availability of quantity of feedstock suited to off-season crushing in sugar industries(Ortiz *et al*., 2006).

Knowledge of combining ability of the parents, especially for hybrid cultivar development is important for optimizing the breeding strategy. Combining ability of sorghum lines for brix has been scarcely reported in the literature. However, there are reports of significant general combining ability (GCA) and specific combining ability (SCA) effects for the associated traits, (Falconer and Mackay 1996) but their level of importance was dependent on the germplasm that was evaluated. Kenga et al. (2004) reported that SCA effects were predominant over GCA for

grain yield and days to anthesis, while Haussmann et al. (1999) reported that GCA effects were more important than SCA effects. Falconer and Mackay (1996) reported that combining ability and heritability information is pertinent to the set of genotypes and the environment where it has been tested.

Sweet sorghum gene pool creation has not received much attention mainly because it is not considered to be among important crops in Kenya, and the pedigree information is incomplete.

1.3. Justification

Appropriate cultivar development requires a holistic approach that includes all stakeholders to facilitate adaptation and subsequent adoption. The information on availability of genetic variability for traits to be considered during breeding of the ideal cultivar is also important and an attempt to quantify this has been made in southern Africa (Makanda *et al*., 2009). Stakeholders' perception that may impact on sweet sorghum cultivar production and adoption has not been reported yet this is important in any breeding program. Many researchers have reported on the negative consequences of not including farmers in setting up research and policy agenda (Ceccarelli and Grando, 2007). Understanding the farmers' preferences is an initial step towards the search for an effective and sustainable way to make agricultural research more relevant to them (Kudadjie *et al.*, 2004). Adoption of improved varieties might fail if farmers' preferences are not seriously considered, as was observed in maize and wheat (Derera *et al.* 2006). Most importantly, the comparative advantage of sweet sorghum over the competing crops and technologies has not been clearly identified. Reports from Malawi revealed that including farmers' views and having them participate in the selection process improved selection and adoption rate of grain sorghum (Nkongolo *et al*. 2008).

Participatory methodologies and their application are successful in obtaining vital information and can be effective in boosting crop productivity and adoption of new crop varieties (Kudadjie *et al.*, 2004; Derera *et al.*, 2006). This has made breeders to shift from the traditional approaches of scientist-centered research agenda to the inclusion of the farmers in problem identification and research agenda formulation (Dixon *et al.*, 2001).

To date, breeding objectives in countries where traditional cropping systems are dominating have not been appropriately oriented towards the perceptions of farmers, specifically their needs and preferences for the difficult growing conditions of their regions (Witcombe *et al.*, 2006; Mekbib, 2006). To overcome this predicament, participatory plant breeding methods have been proposed to bring about a more decentralized breeding approach and the integration of farmers, and their complex selection criteria into a plant improvement program from the early stages (Thapa *et al.*, 2009). Other authors hope to achieve better adoption rates for improved varieties by quantifying farmers' selection criteria and adjusting the breeders' criteria (Defoer *et al*., 1997). Mekbib (2006), on the other hand, proposed combining farmer breeding with formal breeding in an integrated scheme specifically designed for the centers of crop origin and diversity.

In order to meet the industry demand for raw materials especially after crushing of sugarcane crop, there is a need to develop sweet sorghum cultivars that produce high stalk yield per unit time, input, and energy and land area in different agro-climatic areas of the country. These cultivars should also be photo-and thermo-insensitive with desired levels of resistance/tolerance to various stresses such as drought and should be of different maturities to widen the harvest window which would ensure a continuous supply of feed stock to the industry. Sweet sorghum research needs attention especially for enhancing their genetic potential for high sugar content. Through exploitation of sweet sorghum for sugar and other allied products such as jaggery and as

source for ethanol production, poverty reduction can be addressed by linking farmers with new biofuel markets.

Sweet sorghum is best suited for ethanol production because of its higher fermentable sugar content in the stalk compared to sugarcane Reddy *et al*. (2008). The feasibility of converting stalk sugars to ethanol, syrup and jaggery on or near farms, and the adaptability of sorghum to a wide range of environments prompted researchers to evaluate the potential of sweet sorghum as an alternative crop for ethanol production (Daniel *et al*. 1991).

1.4. Objectives

1.4.1. Overall objectives

To contribute to the development of high potential varieties and hybrids of sweet sorghum for various sugar and allied products

1.4.2. Specific objectives

1. To determine farmers perceptions on sweet sorghum and potential of its utilization in Kenya
2. To investigate the influence of genotype by environment interaction on sugar and biomass production of sweet sorghum
3. To assess of genetic diversity and relationship among a collection of sweet sorghum germplasm by SSR markers
4. To determine the combining ability in respect of stem sugar and biomass traits in sweet sorghum.

1.4.3. Research hypotheses

The research tested the following hypotheses:

1. Farmers are not aware of sweet sorghum cultivars and their potential benefits
2. Some sweet sorghum varieties are not stable across different environments
3. Sweet sorghum varieties have low diversity for stem sugar traits among the germplasm collection
4. Stem sugar and biomass traits in sweet sorghum are controlled by genes that do not act predominantly in additive manner.

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CHAPTER TWO

2.0 LITERATURE REVIEW

2.1. Sweet Sorghum

Sweet sorghum (*Sorghum bicolor* [L] Moench) is a single-stemmed cereal grass with a plant height of more than 2 m. In many cases, it is taller than grain sorghum. The crop has become more attractive to breeders due to its capacity to provide renewable energy products, industrial commodities, and food and animal feed (De Jesus and Punzalan, 2007). Because sweet sorghum is a multipurpose crop, it has potential to aid development in semi-arid regions. Sweet sorghum stalk are rich in sugars, which can be primarily used for biofuel production. It can produce up to 7000 L of ethanol/ha (Seetharam, 2005) or can be processed into jaggery and syrup (FAO, 2002).The grains are used for human consumption and stalks used for animal feed (De Jesus and Punzalan, 2007). Sweet sorghum has major advantages compared with its competitive crops such as maize, sugar beet, and sugar cane because it can produce high yield under extreme environmental condition (Reddy *et al*.2008). It can tolerate drought, water logging, and saline-alkali soils due to a number of physiological and morphological characteristics (Reddy *et al*.2008).

2.2. Origin and domestication of sorghum

The genus *Sorghum* comprises a high genetic diversity and therefore, there is potential for crop improvement (Abu Assar *et al*, 2005). The wild distribution and high genetic diversity of sorghum in Africa indicate that this crop must have originated in Africa (Uptmoor *et al*., 2003). To date, Ethiopia and surrounding countries are considered the geographical area of origin and these countries are well recognized in world sorghum improvement programs (Dillon *et al*.,

2007). There are four wild S. *bicolor* subspecies *verticilliflorum* and five cultivated *S. bicolor* subspecies *bicolor* races in sorghum differentiated by head type, grain size, yield potential, and adaptation, among other traits (Acquaah, 2007). The cultivated races are bicolor, guinea, kafir, caudatum and durra (Acquaah, 2007). Plant breeders focus now is on improving sorghum to serve as food, feed, fuel and fibre (Laopaiboon *et al*., 2007 and Vermerris *et al.,* 2007). This involves improving characters such as yield performance and stability, resistance to pests and pathogens, and grain and stem qualities.

2.3. Participatory Rural Appraisal in sweet sorghum cultivar development

Participatory methodologies and their application have been discussed in detail by many authors (FAO, 1990; Burkey, 1993; Anyaegbunam, 1998; Matata *et al.,* 2001), and have been reported to be successful in obtaining vital information and shown to be effective in boosting crop productivity and adoption of new crop varieties (Kudadjie *et al.,* 2004; Derera *et al.,* 2006). Understanding the farmers is an initial step towards the search for an effective and sustainable way to make agricultural research more relevant to them (Kudadjie *et al.,* 2004). Many researchers have reported on the negative consequences of not including farmers in setting up research and policy agenda (Derera *et al*., 2006). Participatory rural appraisals (PRAs) approaches is used in breeding to solicit farmers' views on various agricultural resource management options necessary to ensure household food security and improvement in their welfare. This has led to breeders shifting from the traditional approaches of scientist-centered research agenda to the inclusion of the farmers in problem identification and research agenda formulation (Dixon *et al.,* 2001). Situational studies are very important as a first step in new cultivar development. They generate information about the farmer and their socio-economic conditions that influence on cultivar adoption (Derera *et al.,* 2006). In situations where the farmers and other stakeholders are

not familiar with the technology, as is the case with dual-purpose sorghum cultivars in the lowland areas of Zimbabwe, interacting and discussing with the farmers also helps to create awareness (Makanda *et al.*,2009). This information can be gathered using participatory research techniques used to gather information prior to, during and after technology deployment (Matata *et al*., 2001). The situational studies can also help to explain the anticipated adoption pattern, which aid future breeding projects for the farmers (Matata *et al.*, 2001).

Studies focused on participatory variety selection (PVS) and participatory plant breeding (PPB) show that breeders' selection criteria and their way of assessing cultivar performance – mainly quantitative and statistically based – often differs widely from the methods traditionally implemented by farmers (Mekbib, 2006). Even among farmers and farmer groups themselves, these criteria can vary considerably depending on gender, environmental concerns and economic status (Defoer *et al.*, 1997; Weltzien *et al*., 1998).

2.4. Genetics of stem sugar and biomass in sweet sorghum

Sugar accumulation in sweet sorghum is under the control of recessive genes acting in an additive manner with broad sense heritability (H^2) estimates of 0.65 to 0.81 in different populations (Guiying *et al*., 2000) and GCA and SCA effects are important for stem brix in sorghum (Makanda *et al*., 2009). Guiying *et al.* (2000) reported that crosses between sweet and non-sweet sorghums provided transgressive segregants in the F2 for stem sugar content but crosses between low sugar types resulted in negative heterosis for stem sugar accumulation, giving evidence for gene additivity for low sugar.However, plant biomass is known to be controlled by four dwarfing genes, *Dw1, Dw2, Dw3 and Dw4,* with tall being incompletely dominant to short (Rooney, 2000).

2.5. Genotype by Environment interaction

Over decades, the knowledge of the association between crop performance and environment has been vital in plant breeding and genetic studies. With regard to the comparison of plant material in a set of multi-environment yield trials, crop performance is a function of the genotype; a specific cultivar, the environment which relates to the set of climatic, soil, biotic (pests and diseases) and management conditions where a given cultivar is being grown and finally the interaction between the genotype and environment which becomes important when certain genotypes significantly change ranks in different environments (Weikai & Manjit, 2002). The genotype main effects which constitute differences in mean yield between genotypes provide the only relevant information when genotype (G) by environment (E) interaction (G X E) effects are absent or ignored. However, differences between genotypes may vary widely among environments in the presence of high G x E interaction effects (Rodriguez *et al.*, 2007). The assessment of the magnitude and potential of G x E interaction is important in crop improvement because its effects can be exploited in raising yields for target regions (Rodriguez *et al.*, 2007). It also aids researchers to identify genotypes that exhibit stable performance across diverse environments (Weikai & Manjit, 2002) and consequently, conservation of limited resources that would be used in breeding and maintaining different genotypes for diverse target regions. There are many reports on G × E and stability studies in sorghum (Kenga *et al.,* 2004). Studying G × E for yield using 12 sorghum genotypes of diverse origin across 25 environments, Alagarswamy and Chandra (1998) found that 12% of the variation was due to genotypes, 61% due to environment while G × E accounted for 27%. Chapman *et al.* (2000) reported that most of the G × E in sorghum was a result of the genotype by location by year and suggested that breeders should deal with the genotype by location type over a fixed number of seasons. Allard and Bradshaw

(1964) have discussed the significance of the genotype environment interaction on the basis of the relative magnitude of different variances estimated from multi-location and year test. When genotype × environment variance is very large, the selection for average performance over the entire area from which the locations were drawn may not be considered. If a criterion is found for establishing sub areas or regions, the interaction variance can be reduced (Allard and Bradshaw 1964). Finlay and Wilkinson (1963) reported the stability parameters of a genotype as its phenotypic regression coefficient (bi), which is a measure of the response of ith variety to changing environments. A genotype with unit bi value and higher mean yield (Xi) is said to be stable variety for a range of environments. As the mean yield decreases, the genotypes with high or low slopes are regarded as being specifically adapted to favourable and unfavourable environments, respectively. Improvement in Finlay and Wilkinson (1963) model were made by Eberhart and Russell (1966) by adding another stability parameter *viz.*, deviation from regression. According to them ideal variety is one which has a high mean yield *(*μ*),* unit regression coefficient (b=l), and the least deviation from the regression (S2di=0). Kaltsikes and Larter (1970) while studying the genotype × environment interaction in durum wheat compared the approaches of Finlay and Wilkinson (1963) and Eberhart and Russell (1966).They concluded that for selection of varieties for stable performance, any of the above mentioned three methods can be used. Eberhart and Russell (1966) model is equally efficient to other two models for deciding stability of genotypes. (Luthra and Singh, 1974) recommended that the breeding material should be evaluated both for sensitivity to the environment and for relative mean performance.

Kambal and Mahmoud (1978) estimated variety × environment interactions for grain yield from a study involving 16 sorghum varieties grown at three locations over 3 year period. Whereas, variety × year interaction was small and not significant, the variety x location × year interactions

was highly significant. They suggested that the years of testing could be reduced by increasing the number of test locations. It is therefore important to conduct multi-location testing, quantify G × E and conduct stability analyses to select superior materials in sorghum.

2.6. Use of molecular markers in genetic diversity

Genetic similarity (or genetic distance) estimates among genotypes are helpful in selecting parental combinations for creating segregating populations so as to maintain genetic diversity in a breeding program (Becelaere et al. 2005). Currently there are no criteria (morphological traits or molecular markers) to differentiate sweet sorghums from grain sorghums (Murray *et al*., 2009), and most of the accessions lack the proper information to help distinguish between sweet and grain sorghum. Therefore when requesting sweet sorghum germplasm, one is limited to a few characters that are common in sweet sorghum like tall plants that are leafy (high biomass), and where available the brix degree, which also is subjective as there is no definite value for distinguishing grain sorghums from sweet ones (Murray *et al*., 2009). The majority of the United States of America released sweet sorghum cultivars have a narrow genetic base that can be traced to six African landraces (Murray *et al.*, 2009).

Molecular markers are nucleotide sequences corresponding to a physical position in the genome, and their polymorphisms between accessions allow the pattern of inheritance to be easily traced (Schulman, 2006).The use of SSR markers as a tool to assess relatedness in and between cultivated and wild sorghum have been successfully used (Ritter *et al.*, 2007). Powell *et al*. (1996) reported that SSRs were able to discriminate among closely related individuals, and have advantage over other markers in their ability to trace pedigrees in plants.

2.7. Combining ability

An understanding of gene action involved in stem sugar accumulation and the associated traits may help in developing a viable breeding strategy. Schlehuber (1945) reported that genes with partial dominance action controlled sucrose content in sorghum hybrids. Baocheng *et al*. (1986) reported that genes with additive and dominance effects influenced stem sugar accumulation. Guiying et al. (2000) reported that recessive genes exhibiting additive effects controlled stem sugar accumulation in sorghum. Following a QTL analysis, Natoli *et al.* (2002) reported no significant segregation for genes with major effects on stem sugar percentage. However, studies by Ritter *et al*. (2008) suggested involvement of major genes in addition to genes with minor effects for stem brix. Moderate to high heritability (h^2) estimates, ranging between 40% and 96% (Guiying *et al.,* 2000), and predominance of genes with additive effects suggested that brix could be improved through selection.

Knowledge of combining ability of the parents, especially for hybrid cultivar development is important for optimizing the breeding strategy. Combining ability of sorghum lines for brix has been scarcely reported in the literature (Makanda 2009). However, there are reports of significant GCA and SCA effects for the associated traits, but their level of importance was dependent on the germplasm that was evaluated. Kenga *et al.* (2004) reported that SCA effects were predominant over GCA for grain yield and days to anthesis, while Haussmann *et al.* (1999) reported that GCA effects were more important than SCA effects. Nevertheless, results obtained elsewhere do not necessarily give an indication of the behaviour of the genes in a different environment (Kenga *et al*. (2004). Falconer and Mackay (1996) reported that combining ability and heritability information is pertinent to the set of genotypes and the environment where it has been tested. The GCA of each parent should be examined when the objective is the development of superior

genotypes, while the SCA effects provide information about the performance of hybrids (Cruz and Regazzi, 1994). The differences in GCA are mainly due to the additive genetic effects and higher order additive interactions, while the differences in SCA are attributed to the non-additive dominance and other types of epitasis (Falconer, 1989). The breeder can make use of this information to find the best strategy to select desirable parents or determine which breeding procedure will efficiently improve the performance of the traits of interest (Dudley and Moll, 1969).

2.8. References

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CHAPTER THREE

3.0 MATERIALS AND METHODS

3.1 Survey

A survey was conducted in Homabay, Ndhiwa, and South Nyanza Sugar zone, Kakamega, Mumias, Busia, Nyando and Kwale from October to November, 2012.These are predominantly sugarcane growing areas with established infrastructure for sugar processing. From each region farmers were identified purposively based on whether they are growing sorghum and have potential of growing sweet sorghum. Once the farmers were identified they were assembled in one place briefed on the project and thereafter the questionnaire administered. A total of 70 farmers were interviewed. Data was collected on sorghum production, cultivars grown, the preferred cultivar traits, farmers' awareness and perceptions on use of sweet sorghum as a bio-energy crop, and farmers' preparedness to grow sweet sorghum cultivars, potential of sweet sorghum for sugar and allied products and challenges and opportunities for bio-ethanol production in Kenya. The data collected were analyzed using both the descriptive and inferential statistical tools such as frequency counts and percentages to indicate the proportion of responses to certain variables. Chi-Square tests and Pearson Product Moment Correlation were used to test for significant relationship between awareness and perception of farmers on utilization of sweet sorghum at 0.05 level of significance.

3.2 Germplasm collection, test materials, experimental design and sucrose analysis

For diversity study, a set of 86 cultivars of sweet sorghum genotypes were selected as follows: Four from Argentina, seven from Brazil, 29 from Kenya (ICRISAT and Moi University), four

from United States of America and 42 from Zambia. For genotype x environment study a total of sixteen varieties and two checks (Table 3.1) from ICRISAT (IESV 92038/2-SH, NTJ2, IESV 92008 DL, IESV 93042-SH, IS 2331, IESV 91-018 LT, IESV 91104 DL, IESV 93046,Kenya Agricultural Research Institute (KARI) (KARI Mtama 2, GADAM,) Argentina (Malon, Paisano, Argensor 151 DP, Argensor 165 BIO) and United States of America(NK 5989-29005, NK 7829-29006, NK 8416-19075, NK 8830-29007) were evaluated in Randomized Complete Block Design with three replications in three seasons. Each entry was raised in four rows of 3 m length with a spacing of 70 cm × 20 cm. Sowing was done manually by placing 3 seeds in holes spaced 20cm apart. Data were obtained from plants harvested from the two inner rows of each plot. Care was taken to reduce border effects due to unequal competition of cultivars by the appropriate use of sorghum buffer rows. Nitrogen fertilizer was added at a rate of 100 kg N/ha. All the package of practices were followed to raise a good and healthy crop.

Table 3.1: Characteristics of sweet sorghum varieties used in the study

Variety	**Important features**	**Brix %**	**Purpose**
IESV 93046	High stalk and juice yield	14.0	Stem sugar
IESV 91104 DL	High stalk and juice yield	14.0	Stem sugar
IESV 92008 DL	High stalk and juice yield	13.2	Stem sugar
IS 2331	High stalk and juice yield	16.0	Stem sugar
NTJ 2	Very good leaf disease and stem borer resistance	11.2	Stem sugar
IESV 91018 LT	Vigorous and tall	7.7	Stem sugar
IESV 92038/2-SH	Very good leaf disease and stem borer resistance	11.0	Stem sugar
IESV 93042-SH	Very good leaf disease and stem borer resistance	11.6	Stem sugar
GADAM	High grain yield	8.5	Grain
KARI MTAMA 2	High grain yield and resistant to birds	10.4	Grain
MALON	Very good foliar disease and bird resistance	8.7	Grain and forage
PAISANO	Excellent foliar disease resistance and vigorous	8.4	Grain and forage
ARGENSOR 151 DP	Excellent foliar disease and bird	9.3	Stem sugar and

	resistance		grain
ARGENSOR 165 BIO	Excellent foliar disease and bird resistance	11.8	Stem sugar and grain
NK 5989-29005	High yield potential with excellent leaf disease package	9.1	Stem sugar and fodder
NK 7829-29006	High yield potential with excellent leaf disease package	7.7	Grain and fodder
NK 8416-19075	High yield potential with excellent leaf disease package	7.8	Grain and fodder
NK 8830-29007	High yield potential with excellent leaf disease package	7.4	Grain and fodder

For combining ability study, ten sorghum lines were divided into two groups based on their sugar content (High sugar sweet sorghum and low sugar sorghum).The five high sugar lines were designated as males and crossed with the five low sugar lines designated as females according to North Carolina Design II mating scheme to generate 25 hybrids. The males were from the International Crops Research Institute for the Semi-Arid Tropics (ICRISAT) India and the female were constituted from introduced lines from Argentina, United States of America and Kenya. 25 hybrids, ten parents and one check were laid out in alpha lattice replicated twice and evaluated in three environments.

Sucrose Analysis

Carbohydrate quantification was done only in the main stem. The six plants per plot were pooled to obtain an average sample extract. Pol analysis was done using polarimetric method. Six gm of basic lead acetate was added to 300 ml of juice in clarification process. The juice was filtered through a Whatman filter paper No. 91. The pol reading was then fitted in the formula below to obtain Pol at 20^0C; POL20 = PT {1+ 0.000185(T-20) - 0.000003 (T-20)2}. Brix analysis was done using refracrometric method ICUMSA (1978).

3.3 DNA extraction

Total genomic DNA was extracted from young leaves (12 days old) of five plants of each line planted in the green house at Biosciences eastern and central Africa (BeCA) ILRI hub in Nairobi, Kenya. Five (5) leaves of plants per genotype were harvested and pooled into microtubes. DNA was extracted from leave samples using a modified CTAB protocol (Mace et al. 2003). Two steel beads were put in each well of strip tubes (Greentree Scientific, USA) that were processed in a Geno Grinder 2000. The samples were placed in microtubes and then, 450 µL preheated (65ºC) Extraction Buffer (EB) (3% (w/v) CTAB, 1.4 M NaCl, 0.2% (v/v) β-Mercapto-ethanol and 20 mM EDTA) was added and ground using the Geno-grinder. The quantity and quality of the DNA were checked using a Nano-drop spectrophotometer.

3.4 PCR and SSR assay

Eleven SSR primers from a reference microsatellite kit used to assess genetic diversity of *Sorghum bicolor* (Billot *et al*., 2012) were selected based on their clear polymorphic patterns and on their position in the sorghum genome, covering ten linkage groups or chromosomes. Upon dilution of DNA samples to 20 ng µL-1, a 10 µL PCR mix consisting of 20 ng of DNA, 10 X reaction buffer, 2 mM $MgCl_2$, 0.2 mM dNTPs, 2 pmols of forward and reverse primers, 0.5 U *Taq* polymerase was prepared for each genotype .Temperature cycling was carried out using the GeneAmp PCR systems 9700 (PE-Applied Biosystems), using a program with an initial denaturation at 94°C for 60 s followed by 35 cycles at 94°C for 60 s, 55°C for 60 s, and 72°C for 60 s with a final hold at 72°C for 20 min. After the PCR, a few accessions in each primer were randomly selected and their PCR products (3 ul) run on agarose (2 %) gel electrophoresis stained with gel red (2.5 ul) at a voltage of 100 V for 30 minutes. Genotyping was carried out by capillary electrophoresis using the ABI PRISM 3730 (Applied Biosystems). DNA fragments were denatured and size-fractioned using capillary electrophoresis. The SSRs used in the study

represented di-, tri-, tetra- and penta- nucleotide repeat units. The peaks were sized and the alleles identified using Gene-Mapper software and the internal GS500 (-250) LIZ size standard.

3.5 Study sites

Four study sites were used; Kibos, CYMMIT farm; altitude 1190 meters above the sea level (masl), average daily temperature is 24 ^{0} C, rainfall per annum is 1441mm and the soils are planosol. Alupe; altitude is 1165 masl, average daily temperature is 22.2 ^{0} C, rainfall per annum is 1550 mm and the soils are acrisols. Spectre International Farm-Kisumu**,** The soil type is chromic vertisol described as poorly drained, very deep, very dark grey to black, very firm, cracking clay. The average daily temperature is 23.1^{o}C. The annual average rainfall is 1353mm. The altitude is 1164 masl. Homabay; soil types are black cotton, cracking and swelling montmorillonite. The altitude is 1190 masl. The mean daily temperatures are 25.8 ^{o}C. The annual rainfall ranges from 900 – 1200mm. The materials were evaluated for three seasons. Data collected included days to 50 per cent flowering, , plant height (cm), stem thickness (cm), cane weight (g), juice weight (g), juice volume (ml), Brix at heading stage, soft dough stage, black layer stage and harvesting, and ear head weight (g), 100-seed weight (g) and grain yield per plant (g).

3.6 Data analyses

For farmer perception study data collected were analyzed using both the descriptive and inferential statistical tools of frequency counts and percentages to indicate the proportion of responses to certain variables. Chi-Square tests and Pearson Product Moment Correlation were also used to test for significant relationship between awareness and perception of farmers on utilization of sweet sorghum.

For genetic diversity study, alleles were called and identified using Gene-Mapper version 3.7. Data was subjected to Allelobin software to check the quality of the SSR markers. Data generated

from Allelobin was analyzed using Power-Marker version 3.25 to calculate the Polymorphic Information Content (PIC), heterozygosity, number of alleles for each marker, percentage of polymorphic loci estimates, and genetic diversity among the genotypes and their genetic distances. Allele and genotype frequencies were scored using haplotype diversity values with PowerMarker version 3.25.Darwin (Schenider *et al*., 2000). Version 5.0 software was used to calculate the principle coordinate analysis (PoCA) and clustering among the genotypes. To determine the genetic relationships and differentiation; 86 sweet sorghum accessions were clustered based on the matrix of genetic similarities using the Un-weighted Pair Group Method using the Arithmetic Averages (UPGMA) algorithm. Dissimilarity Index was calculated from allelic data by simple matching. The distances were computed for microsatellite data (11 loci) and trees constructed using the neighbor-joining method with DARwin Version 5.0 software. The genetic distance between genotypes was subjected to sequential agglomerative hierarchical nested (SAHN) with un-weighted, pair-group analysis (UPGMA) using Dice's indices as provided in DARwin 5.0. Major clusters were generated from Nei (1987) genetic distance matrices. Analysis of molecular variance (AMOVA) (Excoffier *et al.* 1992) was used to partition SSR variation among groups. Significance levels for variance component estimates were computed by a non-parametric permutation procedure using 100 permutations. AMOVA and Fst indices were calculated using the GenAlEx program, version 6.5 (Peakall and Smouse, 2012).

For genotype x environment study, the data obtained on all the characters over four environments and three seasons was subjected to GenStat 14th edition to perform the analysis of variance (ANOVA). Randomized completely block experimental design was used in the analysis of variance. The analysis used the Linear Model for randomized completely block design.

$Y_{ij} = \mu + r_i + g_j + e_{ij}$ Where: Y_{ij} = Observed effect for ith replication and jth genotypes

μ = grand mean of the experiment r_i= effect due to the i[th] replication
g_j= effect due to jth genotype e_{ij}= effects due to the residual or random error of the experiment
Other analysis done included AMMI and IPCA.

For combining ability study data, were analyzed using REML procedure in GenStat statistical package (Payne *et al.*, 2007) following a fixed effects model: $Y_{ijkl} = \mu + s_i + r_j(s_i) + m_k + f_l + mf_{kl} + s_i \times m_{ik} + s_i \times f_{ij} + s_i \times mf_{kl} + e_{ijkl}$ where: Y_{ijkl} = observed hybrid performance; μ = overall population mean; s_i = effect of the ith site; $r_j(s_i)$ = effects of the jth replication in the ith site; m_k= effect of the kth male parent; f_l= effect of the lth female parent; mf_{kl} = interaction effect of the kth male and the lth female parents; and e_{ijkl} is the experimental error. The hybrid variation was partitioned into male and female parent main effects giving two independent estimates of GCA effects, while the male and female interaction estimates the SCA effects (Kearsey and Pooni, 1996). The GCA values for the parents and SCA effects for crosses and their standard errors were also estimated (Kearsey and Pooni, 1996).

To estimate heritability, the analogous broad-sense and narrow-sense coefficients of genetic determination were estimated as follows:

Broad-sense heritability;

$$H^2 = \frac{\delta^2\mathrm{GCA_f} + \delta^2\mathrm{GCA_m} + \delta^2\mathrm{SCA_{fm}}}{\delta^2\mathrm{GCA_f} + \delta^2\mathrm{GCA_m} + \delta^2\mathrm{SCA_{fm}} + \delta^2\mathrm{e}}$$

Narrow-sense coefficient genetic determination (heritability);

$$\mathrm{NS-CGD} \approx \mathrm{h}^2 = \frac{\delta^2\mathrm{GCA_f} + \delta^2\mathrm{GCA_m}}{\delta^2\mathrm{GCA_f} + \delta^2\mathrm{GCA_m} + \delta^2\mathrm{SCA_{fm}} + \delta^2\mathrm{e}}$$

Baker's ratio was determined as follows:

$$\text{Baker's ratio} = \frac{\delta^2\mathrm{GCA_f} + \delta^2\mathrm{GCA_m}}{\delta^2\mathrm{GCA_f} + \delta^2\mathrm{GCA_m} + \delta^2\mathrm{SCA_{fm}}}$$

Where

H^2 -Broad sense heritability

$\delta^2 GCA_f$ -Variance of general combining ability females

$\delta^2 GCA_m$ - Variance of general combining ability males

SCA_{fm} - Variance of specific combining ability males and females

$\delta^2 e$ - Error variance.

h^2 -Narrow sense heritability

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CHAPTER FOUR

4.0 FARMERS' PERCEPTION ON SWEET SORGHUM (*Sorghum bicolo*r [L] Moench) AND POTENTIAL OF ITS UTILIZATION IN KENYA

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4.1 Abstract

Studies on farmer's perception of technologies constitute a useful link between both descriptive and prospective research lines. They generate information about the farmer and their socio-economic conditions that influence technology adoption. The area of study was Western and Coastal regions of Kenya. A purposive sampling technique was used to select the farmers who were organized into groups. Descriptive and inferential statistical tests were used to analyze the data. It was observed that 72.9% of the respondents were male, 95.7% married and 46% had formal education. Most of the farmers (73.0%) cultivated below 2 acres of farmland. About 40% of the respondents were aware of existence of sweet sorghum varieties while 50% of them were aware of sweet sorghum processing technologies. Farmer's inability to have contact with extension agents affected their perception and awareness of the technologies. Farmers appreciate the potential of sweet sorghum and existence of capacity for its exploitation.

Key words: Farmers, perception, potential, sweet sorghum.

4.2 Introduction

Farmers' subjective assessments of agricultural technologies influence adoption behavior (Nowak, 1992). Understanding the farmers is an initial step towards the search for an effective and sustainable way to make agricultural research more relevant to them (Kudadjie *et al.,* 2004).

Many researchers have reported on the negative consequences of not including farmers in setting up research and policy agenda (Derera *et al*., 2006). Variables which affect farmers' access to information, and hence their perception formation (e.g. extension, education, media exposure, etc.), are typically used in economic models of the determinants of adoption decisions (Feder *et al.*, 1985; Shakya and Flinn, 1985; Kebede *et al*., 1990; Poison and Spencer, 1991; Strauss *et al.*, 1991). Situational studies are very important in generating information about the farmer and their socio-economic conditions that influence on cultivar adoption. This information can be gathered using participatory research techniques used to gather information prior to, during and after technology deployment (Matata *et al*., 2001). The situational studies can also help to explain the anticipated adoption pattern, which aid future breeding projects for the farmers. Stakeholders views in Zimbabwe and South Africa on development of sorghum for bio-energy has been reported (Makanda, 2009). Sweet sorghum has wider adaptability and offers comparable grain yields (Reddy *et al*., 2008). Sweet sorghum is best suited for ethanol production because of its higher fermentable sugar content in the stalk compared to sugarcane (Reddy *et al*. 2008). Other utilization can include processing it into syrup, grains for human consumption, stillage fibre and animal feed. National and international research centers have reported significant yield increases in many crops. However, farmers remain unaware and have low perception to take full advantage of these technologies (Ekpere, 1995). An ineffective extension service has been partly blamed for this deficiency as well as lack of support services among other factors that make it unprofitable for farmers to accept and implement new technologies (International Institute of Tropical Agriculture 1993, unpublished). No matter how well new technologies work on research stations, if farmers do not have access to them, their development would have been in vain (Bremer *et al.*,

1989). It is acknowledged that some feedback on farmer reaction to a new technology is desirable in order to refine that technology.

Research concerning the production of biofuels has focused on the technical and economic feasibility, as well as the potential supply of alternative sources of biofuel feedstocks (De la Torre Ugarte *et al.,* 2007; Graham *et al.,* 2007; Perlack *et al.,* 2005; Nelson, *et al*., 2010). A significant short-coming of many of these studies is that while they provide a useful frame of reference, they do not examine the necessary economic and institutional conditions under which such a large-scale undertaking would be plausible (Rajagopal *et al*., 2007). That is, how likely it is that farmers are willing to adopt biofuel crops with underdeveloped or nonexistent markets. Rajagopal *et al*., (2007) indicated that there still exists a need to understand the factors that lead to the adoption of biofuel technologies by farmers.

The agricultural research system must therefore conceptualize an effective mechanism and capacity to measure the farmers' perception of new technologies. Studies on farmer's perception of technologies constitute a useful link between both descriptive and prospective research lines.

The overall objective of the study was to analyze the perception of farmers on the potential of sweet sorghum and feasibility of its utilization. In order to meet this objective, the following specific objectives were formulated;

- To identify the demographic characteristics of the farmers
- To determine the level of perception of farmers on sweet sorghum and feasibility of its utilization
- To ascertain the level of awareness on existing infrastructure and their exploitation in sweet sorghum processing

4.3 Materials and Methods

A survey was conducted in Homabay, Ndhiwa, and South Nyanza Sugar zone, Kakamega, Mumias, Busia, Nyando and Kwale from October to November, 2012.These are predominantly sugarcane growing areas with established infrastructure for sugar processing. From each region farmers were identified purposively based on whether they are growing sorghum and have potential of growing sweet sorghum. Once the farmers were identified, they were assembled in one place briefed on the project and thereafter the questionnaire administered. A total of 70 farmers were interviewed. Data was collected on sorghum production, cultivars grown, the preferred cultivar traits, farmers' awareness and perceptions on use of sweet sorghum as a bio-energy crop, and farmers' preparedness to grow sweet sorghum cultivars, potential of sweet sorghum for sugar and allied products and challenges and opportunities for bio-ethanol production in Kenya. The data collected were analyzed using both the descriptive and inferential statistical tools such as frequency counts and percentages to indicate the proportion of responses to certain variables. Chi-Square tests and Pearson Product Moment Correlation were used to test for significant relationship between awareness and perception of farmers on utilization of sweet sorghum at 0.05 level of significance.

4.4 Results and discussion

4.4.1.Socio Economic characteristics of the farmers.

Socio-economic characteristics of farmers are important factor in determining the perception and awareness of some farming practices and adoption of the farming technology. Table 4.1 shows the proportion of factor levels of some of the socio-economic characteristics of the farmers. It can be pointed out from the results that males (72.9%) were more prominent in farming activities than

females. The high percentage of male farmers may be due to their access to farmland and their position as head of family. These results agree with the work of Oguntola (1998) who concluded that farming is a male-dominated profession. The lower proportion of female farmers could be due to previous land ownership system which discriminated against women. The high percentage (95.7%) of the farmers that were married may be as a result of the belief of the local people that married people are more responsible. In addition, most people probably married in order to raise large families that would supply labour on the farm. The distribution of age (Table 4.1) revealed that 48.6 % of the farmers were aged between 35 to 50 years. Approximately 46 % of the farmers had secondary education. This reflects fairly high levels of literacy of people in the area. These gives a strong combination characteristics that when fully utilized, there would be a high awareness and perception and hence high adoption and productivity of technologies (Strauss *et al.*, 1991)

Table 4.1: Socio-economic characteristics of the farmers surveyed on perception of sweet sorghum in Kenya

Variable	**Factor**	**Counts (Percentage)**
Gender	Male	51 (72.9)
	Female	19 (27.1)
Marital Status	Married	67 (95.7)
	Single	3 (4.3)
Education Level	None	3 (6.7)
	Primary	28 (40.0)
	Secondary	32 (45.7)
	Tertiary	1 (1.4)
	Missing	6 (8.6)
Occupation	Farming	56 (80.0)
	other occupation	14 (20.0)
Age	up to 35	12 (17.2)
	35 to 50	34 (48.6)
	over 50	21 (30.2)

4.4.2. Farmers' Land ownership

The cross tabulation in Table 4.2 shows the acreage of land farmers have and their ownership. It is pointed out that 73 % of the farmers have only up to 2 acres of land. The land tenure system, fragmentation of farmland and human activities such as the building of roads and industries may force people to have small farm size. Very few farmers have 10 and above acres (4.9%). 61.4% of the farmers that were interviewed own parcels of land. These may be farmers that had access to farmland because they were indigenous to the area or they were leaders of families. 10.3 % of the farmers lease land for their farming activities. Some farmers (27.9%) use their communal land although majority of them (54.6%) had less than 2 acres.

Table 4.2: Land ownership of sorghum farmers

		Land Ownership			
		Own (%)	**Leased (%)**	**Communal(%)**	**Total (%)**
Land sizes in acre	0 to 2	32 (78.0)	7 (100)	10 (54.6)	49 (73.5)
	2 to 5	4 (9.8)	0 (0.0)	4 (21.1)	8 (11.8)
	5 to 10	3 (7.3)	0 (0.0)	3 (15.8)	6 (8.8)
	10 & above	2 (4.9)	0 (0.0)	2 (10.5)	4 (5.9)
	Total	41 (61.5)	7 (10.3)	19 (27.9)	67 (100.0)

4.4.3. Preferred common sorghum varieties currently grown by the farmers.

There are a number of sorghum varieties that are currently grown by farmers. Some of the varieties are known to the farmers as listed in Table 4.3 below while majority of the farmers (54%) are not aware of the variety they are growing. Amongst the varieties that are known to the farmers, Seredo is the most common (21% of the farmers are growing it). Majority (77%) of the farmers who were interviewed liked the varieties that they are currently growing.

Table 4.3: Common sorghum variety preference by farmers.

		Whether the farmer like the current variety					
		Yes		**No**		**Total**	
		No.	%	No.	%	No.	%
Sorghum variety grown	Seredo	12	80	3	20	15	21
	Ochuti	6	100	0	0	6	9
	Brown	3	100	0	0	3	4
	Hybrid	2	100	0	0	2	3
	Jowi Jamwomo	2	100	0	0	2	3
	Andiwo	1	100	0	0	1	1
	Japidi	0	0	1	100	1	1
	Gadam	0	0	1	100	1	1
	Obamo	1	100	0	0	1	1
	Local	27	71	11	29	38	54
	Total	**54**	**77**	**16**	**23**	**70**	

No. =Number, % =Percentage

4.4.4. Effect of famers having a choice on variety on the preference of the variety

A Chi-Square analysis was used to determine if having a choice on variety of sorghum will have an association with the preference of the variety the farmer grows. The Chi square test (Table 4.4) resulted to a test statistics of 0.949 and a P-value of 0.333. Using the assumed null hypothesis of no association, there is no significant evidence of association between having a choice on variety and preference of the variety grown. (Figure 4.1 and Table 4.4)

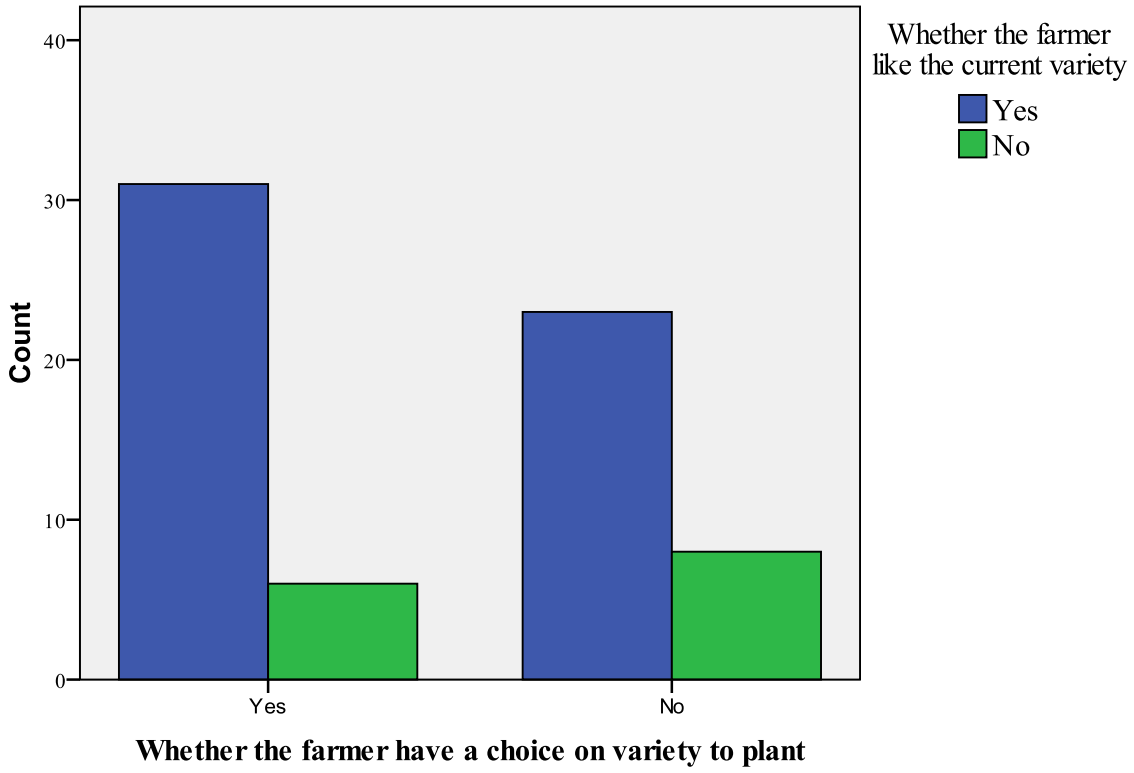

Figure 4.1: Association of having a choice on variety and preference of the variety

Table 4.4: Effect of famers having a choice on variety on the preference of the variety

Chi-Square Tests

	Value	df	Asymp. Sig. (2-sided)
Pearson Chi-Square	.949a	1	.330
Continuity Correction	.453b	1	.501
Likelihood Ratio	.946	1	.331
Fisher's Exact Test			
No. of Valid Cases	68		

a. 0 cells (.0%) have expected count less than 5. The minimum expected count is 6.38.

b. Computed only for a 2x2 table

4.4.5. Reasons for common sorghum varieties preference

Some of the reasons for sorghum variety preferences are shown in Table 4.5. Higher percentage of farmers prefers the varieties that they are currently growing because of their palatability. The percentages are computed based on the number of farmer's responses per variety.

Table 4.5: Reasons for common sorghum variety preferences

		High Yield		Early maturity		High bird tolerance		High Drought tolerance		Palatable		Total
		No.	%	No.	%	No.	%	No.	%	No.	%	
Variety	Seredo	7	(47)	6	(40)	7	(47)	2	(13)	13	(87)	15
	Ochuti	2	(33)	0	(0)	3	(50)	0	(0)	5	(83)	6
	Brown	0	(0)	1	(33)	1	(33)	0	(0)	1	(33)	3
	Hybrid	2	(100)	1	(50)	0	(0)	0	(0)	2	(100)	2
	Jowi Jamwomo	1	(50)	0	(0)	0	(0)	0	(0)	2	(100)	2
	Andiwo	0	(0)	0	(0)	0	(0)	0	(0)	1	(100)	1
	Japidi	0	(0)	0	(0)	1	(100)	0	(0)	1	(100)	1
	Gadam	0	(0)	0	(0)	0	(0)	0	(0)	0	(0)	1
	Obamo	1	(100)	1	(100)	0	(0)	1	(100)	1	(100)	1
	Local	9	(39)	7	(30)	5	(22)	7	(30)	17	(74)	23

4.4.6. Farmers' awareness of the sweet sorghum production potentials

The farmers' awareness on the potentials of sweet sorghum was measured in different aspects as shown in Figure 4.2 .Amongst the aspects measured, approximately 40 % of the farmers are aware that sweet sorghum varieties exist while 50 % of the farmers acknowledge that there is need for the adjustment of the sugarcane machinery to be used in producing fuel and sugar from sweet sorghum.

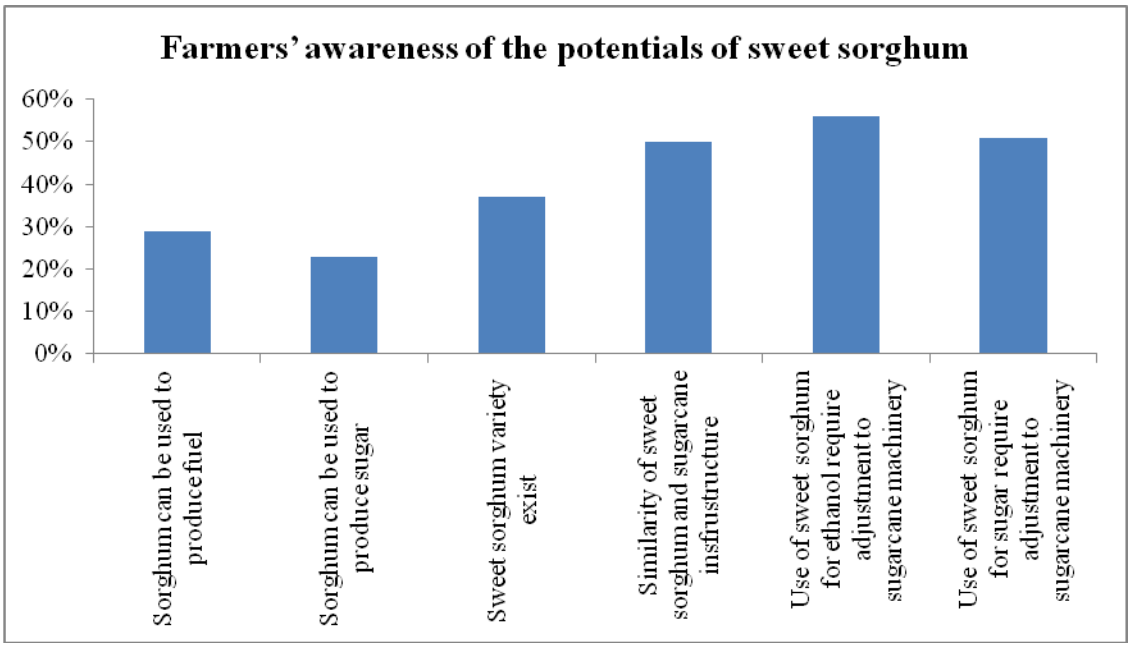

Figure 4.2: Farmers' awareness of the potentials of sweet sorghum

There was some evidence of significant association between level of education and the awareness that the farmers had about the similarity of sweet sorghum and sugarcane infrastructure (Chi-square statistics=31.313, P-value (PV) =0.012) and between level of education and the awareness that the use of sweet sorghum for ethanol requires adjustment to sugarcane machinery (Chi-square statistics=52.359, PV=0.000). However infrastructural challenges were identified as potential limitations to exploitation of sweet sorghum for bio-ethanol production.

4.4.7. Farmers' perception on sweet sorghum production

Sweet sorghum being a new technology in the sugar industry, majority of farmers (60%) as shown in Figure 4.3, are willing to venture in the farming of sweet sorghum with the aim of selling the stalks only. They are also willing to take part in the development of the sweet sorghum and it products by allowing small mills in their farms (80%), promoting and marketing sweet sorghum (90%) and willing to be contracted for sweet sorghum production (85%).

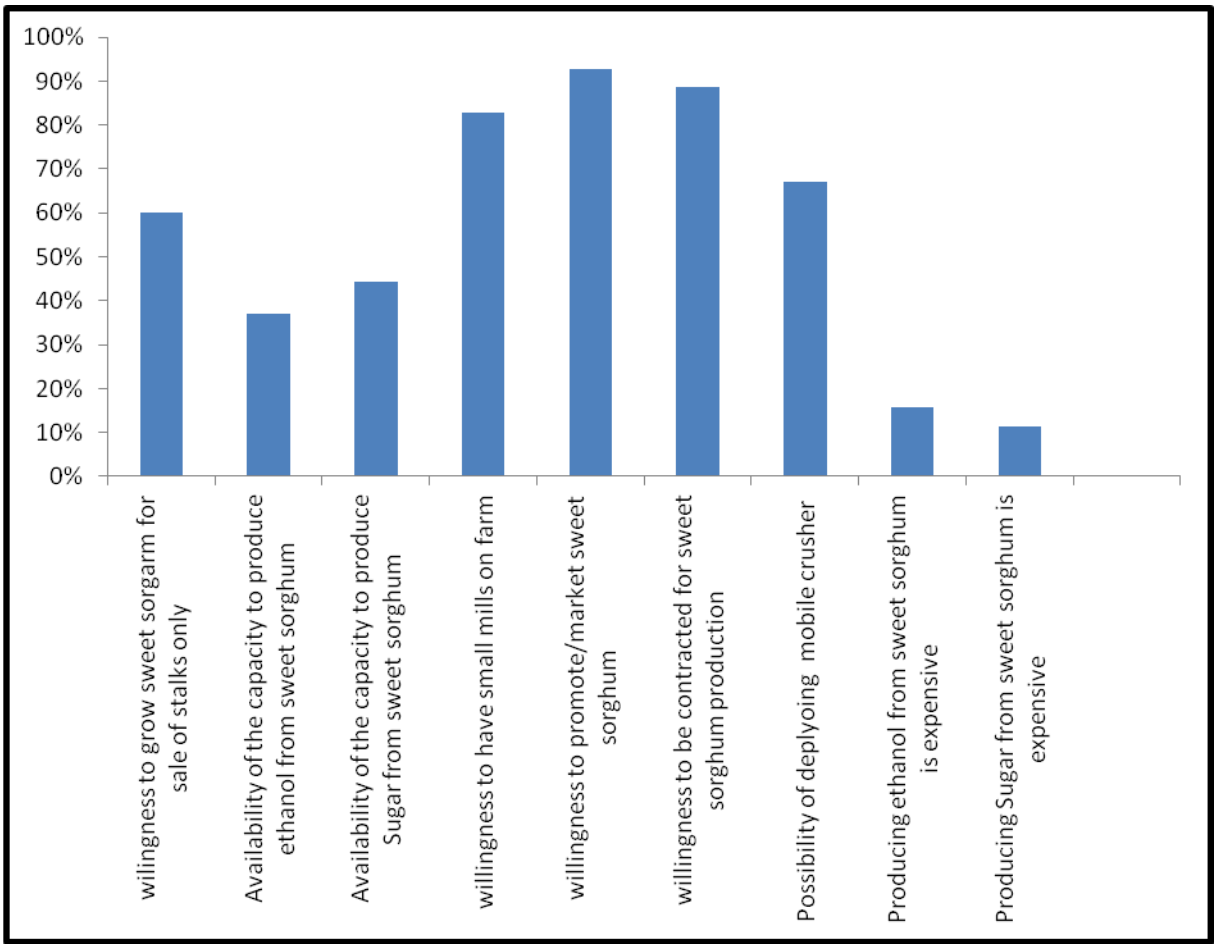

Figure 4.3: Farmers' perception on sweet sorghum production and related technologies

These results are consistent as shown in Figure 4.3. Further analysis on the farmers' awareness and perception on sorghum production was done using Chi-square test. The results show that there is a strong association between the farmer's willingness to promote/market sweet sorghum and their awareness on sorghum varieties they plant (Chi-square statistic = 26.564 and P-Value=0.001). Also a significant association was realized between the willingness to promote/market sweet sorghum and awareness that there is similarity of sweet sorghum and sugarcane infrastructure. (Chi-square statistic = 23.331 and P-Value=0.003).

These results are in line with the impact of acquiring knowledge by farmers. Amongst the respondents that were interviewed, only 30% of them had received some training on sorghum farming (Figure 4.4).

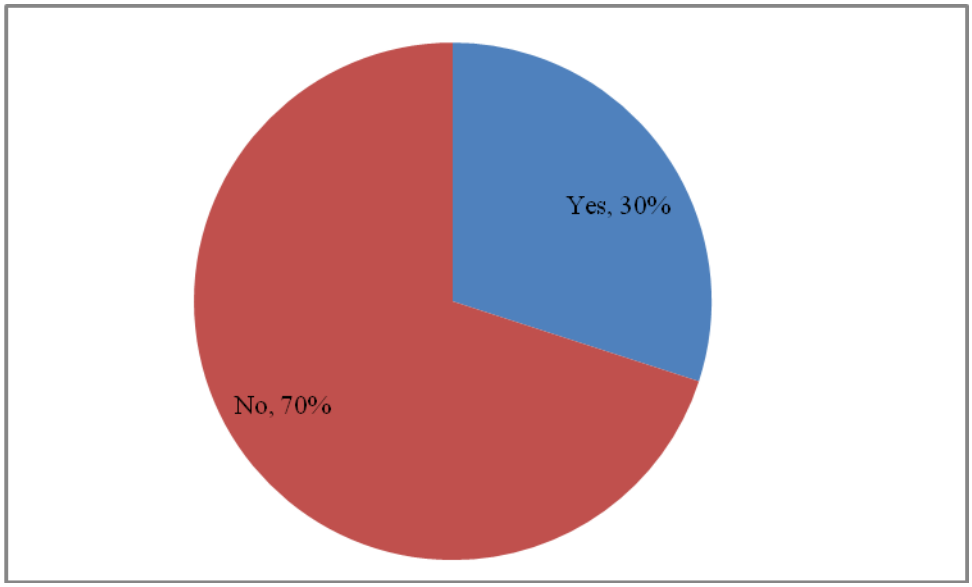

Figure 4.4: Whether farmer receive advice on sorghum farming

4.5 Conclusions

Farmers were aware of sweet sorghum and accompanying technologies however their perception was constrained by some socio-cultural factors.

Due to strong association of level of education and the awareness that the farmers had about the similarity of sweet sorghum and sugarcane infrastructure, sweet sorghum farming will thrive amongst educated farming community.

Many farmers grow local sorghum varieties at the expense of hybrids.

Farmers appreciate the potential of sweet sorghum and existence of capacity for its exploitation and are likely to adopt it.

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CHAPTER FIVE

5.0 ASSESSMENT OF GENETIC DIVERSITY FOR IMPROVEMENT OF SWEET SORGHUM (Sorghum bicolor (L.) Moench) GENOTYPES FOR SUGAR AND ALLIED PRODUCTS

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5.1 Abstract:

Sweet sorghum (*Sorghum bicolor* (L.) Moench) is a cultivated sorghum recognized as potential alternative source of bio-fuel due to its high fermentable sugar content in the stalk. It provides renewable energy products, industrial commodities, food and animal feed. Sweet sorghum gene pool creation has not received much attention mainly because it was not considered to be among important crops in Kenya, and the pedigree information is scarce. The objective of the study was to assess the genetic diversity and determine relationships among a collection of sweet sorghum genotypes using SSR markers. Eighty six sweet sorghum cultivars from Argentina, Brazil, Kenya (ICRISAT and Moi University), United States of America and Zambia were genotyped with 11 SSR markers that generated 86 alleles with an average of 8 alleles per locus. Polymorphism information content (PIC) value was 0.53 indicating a moderate diversity with a range of 0.09–0.89. The variability among the populations was low as 3 % but amounted to 22% and 75 % within individual genotypes and among individuals respectively. Clustering analysis based on the genetic similarity (GS) grouped the 86 sweet sorghum genotypes into 2 distinct clusters. The

study also revealed the genetic relationship of cultivars with unknown parentage to those with known parentage. Information generated from this study can be exploited to select parents for hybrid development to maximize sugar content and total biomass and for development of segregating populations to map genes controlling sugar content in sweet sorghum.

Keywords: Genetic distance, genetic diversity, simple sequence repeats (SSR) markers, sweet sorghum

5.2 Introduction

Interest in exploiting sweet sorghum (*Sorghum bicolor* (L.) Moench) as a biofuel crop is growing due to its rich stalk sugar content (Wang *et al*., 2009). Many breeding programs are working towards development of high-yielding varieties and hybrids with higher sugar content, resistance to diseases, drought tolerance and good agronomic traits (Klein *et al*., 2008). Significant breakthrough has been made in developing and releasing sorghum hybrids and varieties for commercial cultivation both in India and elsewhere (Kumar *et al*., 2011). Genetic similarity estimates among genotypes are important in selecting parental combinations for creating segregating populations to maintain genetic diversity in a breeding programs (Becelaere *et al*., 2005), develop mapping populations for detecting quantitative trait loci (QTL) (Varshney, 2011) and categorize lines into heterotic groups for hybrid crop breeding (Menz *et al.*, 2004). Simple sequence repeats (SSR), also known as microsatellites, are based on tandem repeats of one to six core nucleotide elements. Different studies have recommended the use of SSR markers in analyses of genetic diversity due to their high degree of polymorphism (Geleta *et al*., 2006; Ali *et al*., 2008; Shehzad *et al*., 2009). Simple sequence repeats are co dominant markers dispersed throughout the genome, and have multiple alleles that often have conserved loci between related species (Brown *et al*., 1996; Schulman, 2006). The SSRs are able to discriminate among closely

related individuals, and have advantage over other markers in their ability to trace pedigrees in plants (Powell *et al*., 1996). In sorghum, several studies have been conducted involving SSR markers either alone or in combination with other marker types (Casa *et al*., 2005; Ali *et al*., 2008; Klein *et al*., 2008; Murray *et al*., 2009). In sweet sorghum, genetic diversity has been successfully determined using amplified fragment length polymorphism (AFLP) and simple sequence repeat (SSR) markers. Due to their high values for polymorphic information content (PIC) and the Shannon diversity index (Geleta *et al*., 2006), these marker types were adequate for use in these species. Sweet sorghum gene pool creation has not received much attention mainly because it was not considered to be among important crops in the Kenya, and the pedigree information is scarce. The study was therefore undertaken to assay genetic diversity among a collection of sweet sorghum genotypes using SSR markers to identify specific genotypes exhibiting highest levels of polymorphism i.e. genotypes bearing a maximum of favourable alleles with the aim of improving the genetic base of the current cultivated sweet sorghum to support a breeding programme that will provide Kenyan and African farmers with high-yielding sweet sorghum varieties.

5.3 Materials and Methods

5.3.1. Germplasm collection

A set of 86 cultivars of sweet sorghum genotypes (Table 5.1) were selected as follows: Four from Argentina, seven from Brazil, 29 from Kenya (ICRISAT and Moi University), four from United States of America and 42 from Zambia.

Table 5.1: 86 sweet sorghum genotypes used in genetic diversity study, their genotype identification (GI), genotype name and region of collection

No.	GI	Name	Region	No.	GI	Name	Region
1	ZM1	SDS 89426	ZAMBIA	**44**	BR2	MXS 5648	BRAZIL
2	ZM2	PRGC	ZAMBIA	**45**	BR3	MXS 5637	BRAZIL
3	ZM3	ICSV 1089	ZAMBIA	**46**	BR4	MXS 5635	BRAZIL
4	ZM4	ICSV 1090	ZAMBIA	**47**	BR5	MXS 5646	BRAZIL
5	ZM5	MACIA	ZAMBIA	**48**	BR6	MXS 5647	BRAZIL
6	ZM6	ZSV-18	ZAMBIA	**49**	BR7	MXS 5639	BRAZIL
7	ZM7	ZSV-30	ZAMBIA	**50**	ARG1	MALON	ARGENTINA
8	ZM8	ZSV-31	ZAMBIA	**51**	ARG2	PAISANO	ARGENTINA
9	ZM9	SDS 4378	ZAMBIA	**52**	ARG3	ARG 151 DP	ARGENTINA
10	ZM10	SDS 1023	ZAMBIA	**53**	ARG4	ARG 165 BI	ARGENTINA
11	ZM11	SDS 876	ZAMBIA	**54**	USA1	NK 7829	USA
12	ZM12	SDS3845	ZAMBIA	**55**	USA2	NK 8830	USA
13	ZM13	SDS 3846	ZAMBIA	**56**	USA3	KS 5989	USA
14	ZM14	SDS2690	ZAMBIA	**57**	USA4	NK 8416	USA
15	ZM15	SDS2691	ZAMBIA	**58**	KARI1	GADAM	KENYA
16	ZM16	KSV-10	ZAMBIA	**59**	KARI2	MTAMA 2	KENYA
17	ZM17	KSV-7	ZAMBIA	**60**	ICR1	IESV 93046	KENYA
18	ZM18	SDS 4380	ZAMBIA	**61**	ICR2	IS 2331	KENYA
19	ZM19	ZSV-12	ZAMBIA	**62**	ICR3	NTJ 2	KENYA
20	ZM20	WP-13	ZAMBIA	**63**	ICR4	IESV 92008	KENYA
21	ZM21	ZM 2489	ZAMBIA	**64**	ICR5	IESV 93042	KENYA
22	ZM22	ZM 2499	ZAMBIA	**65**	ICR6	IESV 92038	KENYA
23	ZM23	ZM 2511	ZAMBIA	**66**	ICR7	IESV 91104	KENYA
24	ZM24	ZM 2518	ZAMBIA	**67**	MU1	IESV 91018	KENYA
25	ZM25	ZM 2536	ZAMBIA	**68**	MU2	IESV 92038	KENYA
26	ZM26	ZM 2547	ZAMBIA	**69**	MU3	IS 23496	KENYA
27	ZM27	ZM 2560	ZAMBIA	**70**	MU4	ICSV 700	KENYA
28	ZM28	ZM 2562	ZAMBIA	**71**	MU5	ST 17	KENYA
29	ZM29	ZM 2578	ZAMBIA	**72**	MU6	IESV 92099	KENYA
30	ZM30	ZM 2580	ZAMBIA	**73**	MU7	S 35	KENYA
31	ZM31	ZM 2584	ZAMBIA	**74**	MU8	IESV 92006	KENYA
32	ZM32	ZM 2592	ZAMBIA	**75**	MU9	ICSA 38	KENYA
33	ZM33	ZM 2602	ZAMBIA	**76**	MU10	IESV 92047	KENYA
34	ZM34	ZM 2610	ZAMBIA	**77**	MU11	ICSB 657	KENYA
35	ZM35	ZM 2625	ZAMBIA	**78**	MU12	IESV 92070	KENYA
36	ZM36	ZM 3869	ZAMBIA	**79**	MU13	IESV 9202	KENYA
37	ZM37	ZM 3935	ZAMBIA	**80**	MUI14	IESV 92207	KENYA
38	ZM38	ZM 3990	ZAMBIA	**81**	MU15	ICSB 297	KENYA
39	ZM39	ZM 4668	ZAMBIA	**82**	MU16	E 36-1	KENYA
40	ZM40	ZM 4856	ZAMBIA	**83**	MU17	ICSB 502	KENYA
41	ZM41	ZM 5750	ZAMBIA	**84**	MU18	ICSB 324	KENYA
42	ZM42	Sima	ZAMBIA	**85**	MU19	ENT 18-2	KENYA
43	BR1	MXS 5636	BRAZIL	**86**	MUI20	ICSB 592	KENYA

5.3.2. DNA extraction

Total genomic DNA was extracted from young leaves (12 days old) of five plants of each line planted in the green house at Biosciences eastern and central Africa (BeCA) ILRI hub in Nairobi, Kenya. Five (5) leaves of plants per genotype were harvested and pooled into microtubes tubes. DNA was extracted from leave samples using a modified CTAB protocol (Mace et al. 2003). Two steel beads were put in each well of strip tubes (Greentree Scientific, USA) that were processed in a Geno Grinder 2000. The samples were placed in microtubes and then, 450 µL preheated (65ºC) Extraction Buffer (EB) (3% (w/v) CTAB, 1.4 M NaCl, 0.2% (v/v) β-Mercapto-ethanol and 20 mM EDTA) was added and ground using the Geno-grinder. The quantity and quality of the DNA were checked using a Nano-drop spectrophotometer.

5.3.3. PCR and SSR assay

Eleven SSR primers from a reference microsatellite kit used to assess genetic diversity of *Sorghum bicolor* (Billot *et al*., 2012) were selected based on their clear polymorphic patterns and on their position in the sorghum genome, covering ten linkage groups or chromosomes. Upon dilution of DNA samples to 20 ng µL-1, a 10 µL PCR mix consisting of 20 ng of DNA, 10 X reaction buffer, 2 mM $MgCl_2$, 0.2 mM dNTPs, 2 pmols of forward and reverse primers, 0.5 U *Taq* polymerase was prepared for each genotype .Temperature cycling was carried out using the GeneAmp PCR systems 9700 (PE-Applied Biosystems), using a program with an initial denaturation at 94°C for 60 s followed by 35 cycles at 94°C for 60 s, 55°C for 60 s, and 72°C for 60 s with a final hold at 72°C for 20 min. After the PCR, a few accessions in each primer were randomly selected and their PCR products (3 ul) run on agarose (2 %) gel electrophoresis stained with gel red (2.5 ul) at a voltage of 100 V for 30 minutes. Genotyping was carried out by capillary electrophoresis using the ABI PRISM 3730 (Applied Biosystems). DNA fragments were

denatured and size-fractioned using capillary electrophoresis. The SSRs used in the study represented di-, tri-, tetra- and penta- nucleotide repeat units. The peaks were sized and the alleles identified using Gene-Mapper software and the internal GS500 (-250) LIZ size standard.

5.3.4. Cluster analyses

Dissimilarity indices were estimated using allelic data by simple allele matching and cluster analysis based on unweighted neighbor-joining (Gascuel, 1997) were carried using DARwin 5.0 dissimilarity analysis software. To ascertain the statistical strength of genetic relationships identified through this analysis, bootstrapping of the data (10,000 permutations) was performed. The total number of alleles detected, the number of common alleles with allelic frequencies of at least 5%, the observed size range (in base pairs; bp), the allele size differences (in bp), the polymorphism information content (PIC) values (Smith *et al*., 2000), and frequencies of unique alleles were calculated for each SSR marker using PowerMarker Version 3.25 (Liu and Muse, 2005).

5.3.5. Data analysis

Alleles were called and identified using Gene-Mapper version 3.7. Data was subjected to Allelobin software to check the quality of the SSR markers. Data generated from Allelobin was analyzed using Power-Marker version 3.25 to calculate the Polymorphic Information Content (PIC), heterozygosity, number of alleles for each marker, percentage of polymorphic loci estimates, and genetic diversity among the genotypes and their genetic distances. Allele and genotype frequencies were scored using haplotype diversity values with PowerMarker version 3.25. Darwin Version 5.0 software was used to calculate the principle coordinate analysis (PoCA) and clustering among the genotypes. To determine the genetic relationships and differentiation;

the 86 sweet sorghum accessions were clustered based on the matrix of genetic similarities using the Un-weighted Pair Group Method using the Arithmetic Averages (UPGMA) algorithm. Dissimilarity Index was calculated from allelic data by simple matching. The distances were computed for microsatellite data (11 loci) and trees constructed using the neighbour-joining method with DARwin Version 5.0 software. The genetic distance between genotypes was subjected to sequential agglomerative hierarchical nested (SAHN) with un-weighted, pair-group analysis (UPGMA) using Dice's indices as provided in DARwin 5.0. Major clusters were generated from Nei (1987) genetic distance matrices. Analysis of molecular variance (AMOVA) (Excoffier et al. 1992) was used to partition SSR variation among groups. Significance levels for variance component estimates were computed by a non-parametric permutation procedure using 100 permutations. AMOVA and Fst indices were calculated using the GenAlEx program, version 6.5 (Peakall and Smouse, 2012).

5.4 Results

5.4.1. Marker characterization and allele frequencies

The allele sizes among the genotypes for the 11 microsatellites varied from 98 to 244 bp (Table 5.2).

Table 5.2: The 11 SSR markers used in this study, the dyes used to label them, repeat motif, chromosome number and allele size range

Marker	Dye label	Repeat motifs	Chromosome number	Allele size range(bp)
Xcup 53	NED	(TTTA)5	1	186-198
Xcup 63	VIC	(GGATGC)4	2	133-145
mSbCIR 329	6-FAM	(AC)9	5	109-117
mSbCIR 246	6-FAM	(CA)7	7	98-100
Xtxp 021	PET	(AG)18	4	169-199
Xcup 14	6-FAM	(AG)10	3	211-225
Xcup 02	VIC	(GCA)6	9	192-204
Xtxp 273	NED	(TTG)20	8	169-199
Xtxp 012	6-FAM	(CT)22	4	161-205
Xtxp 145	VIC	(AG)22	6	208-244
mSbCIR 283	PET	(CT)8(GT)8	10	113-139

Number of repeats in the SSR motif had strong correlation with allele number and their polymorphism information content (Table 5.3). The 11 SSRs revealed a total of 86 alleles with a mean of 8 alleles per marker (Table 5.3).

Table 5.3: Summary of allele frequency, allele number and diversity indices of 86 sweet sorghum genotypes

Marker name	Major allele frequency	Genotype number	Allele number	Gene diversity	Heterozygosity	PIC
Xcup 53	0.90	6	4	0.19	0.03	0.18
Xcup 63	0.95	5	5	0.09	0.01	0.09
mSbCIR 329	0.38	16	9	0.75	0.14	0.72
mSbCIR 246	0.43	4	3	0.65	0.84	0.57
Xtxp 021	0.49	12	9	0.68	0.07	0.64
Xcup 14	0.54	13	10	0.64	0.25	0.60
Xcup 02	0.54	8	7	0.64	0.01	0.60
Xtxp 273	0.73	8	7	0.43	0.10	0.39
Xtxp 012	0.19	32	19	0.90	0.35	0.89
Xtxp 145	0.73	6	6	0.44	0.00	0.41
mSbCIR 283	0.36	10	7	0.75	0.06	0.71
Mean	**0.57**	**11**	**8**	**0.56**	**0.17**	**0.53**

The allele number ranged from 3 (mSbCIR 246) to 19 (Xtxp 012). All the markers were polymorphic (Table 5.3). The PIC value over the 11 SSR markers averaged 0.53, ranging from 0.09 for marker Xcup 63 to 0.89 for marker Xtxp 012. The mean level of heterozygosity per SSR marker was 0.17. (Table 5.3). Heterozygosity level ranged from 0.00 for marker Xtxp 145 to 0.84 for marker mSbCIR 246. Marker Xtxp 012 had the highest gene diversity of 0.9 while marker Xcup 63 had the lowest gene diversity of 0.09. The mean gene diversity per SSR marker was 0.56 (Table 5.3).

5.4.2. Population structure

There was a clear genetic differentiation among individuals within populations, and within the individuals using significance tests based on 1,000 permutations (Table 5.4).

Table 5.4: AMOVA partitioning SSR variation, among populations, among individuals within populations, and within individuals in 86 sweet sorghum genotypes

Source	d.f	Sum of squares	Variance components	P-value	Percentage of variation	Fst
Among Populations	4	38.10	0.12	0.03	3.4	
Among Individuals within populations	81	497.58	2.68	<0.001	75.0	0.034
Within Individuals	86	66.50	0.77	<0.001	21.6	
Total	**171**	**602.18**	**3.58**		**100.0**	

The variability among the populations was low as 3 % but amounted to 22% and 75 % within individual genotypes and among individuals respectively. The F_{ST} value was 0.034.

All the loci were polymorphic with Zambian population having the highest number of private alleles (Table 5.5).

Table 5.5: Table of Genetic diversity for each sweet sorghum populations analyzed in this study

Genetic Diversity						
Population	**Sample size**	**% Polymorphic Loci**	**Private alleles**	**Na(SE)**	**He(SE)**	**Ho(SE)**
Zambia	42	100	2	5.909(0.576)	0.564(0.076)	0.166(0.082)
Kenya	29	100	1	3.636(0.650)	0.511(0.074)	0.151(0.071)
Brazil	7	72.7	0	2.727(0.469)	0.348(0.088)	0.176(0.071)
Argentina	4	72.7	0	2.000(0.270)	0.330(0.074)	0.182(0.076)
USA	4	81.8	0	2.273(0.273)	0.431(0.072)	0.242(0.092)
Overall mean	**17**	**85.4**		**3.655(0.374)**	**0.437(0.035)**	**0.183(0.034)**

He = Expected heterozygosity; Na= No. of different alleles; Ho =Observed heterozygosity; SE= Standard Error

5.4.3. Genetic diversity within regions

The allele frequency based pair-wise genetic distances between the countries calculated using Power- Marker version 3.25 revealed the relatedness of genotypes on a country by country basis (Table 5.6).

Table 5.6: Genetic distance matrices between countries calculated according to Nei (1987) for the 86 sweet sorghum genotypes

Country	**Zambia**	**Brazil**	**Argentina**	**United States of America**	**Kenya**
Zambia	0.000				
Brazil	0.223	0.000			
Argentina	0.156	0.203	0.000		
United States of America	0.188	0.185	0.196	0.000	
Kenya	0.070	0.303	0.203	0.228	0.000

Genotypes from Kenya and Brazil were the most distant at 0.303 whereas genotypes from Kenya and Zambia were the closest at 0.070.

5.4.4. Cluster analysis

The pair-wise dissimilarity indices among the sweet sorghum genotypes were estimated using allelic data by simple allele matching followed by cluster analysis using unweighted neighbor-joining algorithm. All the 86 genotypes fell into two (I and II) clusters corresponding mainly to their geographic origin and pedigree. The biggest cluster, cluster I had 83 genotypes with cluster II having 3 genotypes (Z35 and Z42 from Zambia and K86 from Kenya), Figure 5.1.

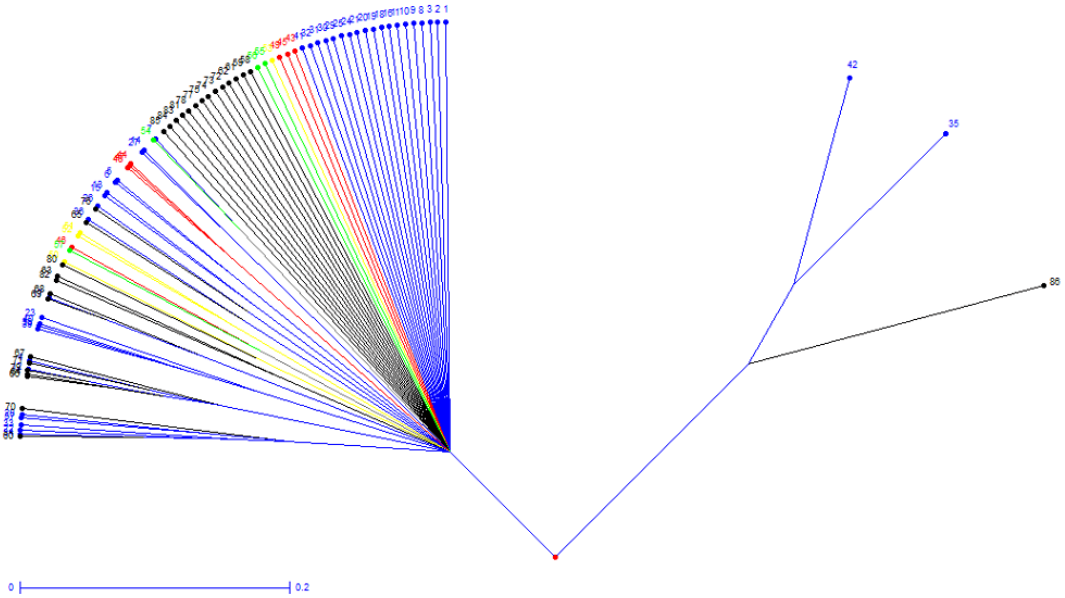

Figure 5.1: Dendogram (radial axis) of 86 sweet sorghum genotypes revealed by cluster analysis of genetic similarity estimates generated by Nei coefficient based on 11 SSR markers

5.5 Discussion

With the availability of complete sorghum genomic sequence (Paterson *et al*., 2009) simple sequence repeats (SSRs) have become the preferred markers of choice for studying genetic diversity of sorghum owing to their co-dominance, multi-allelic nature, ease of use and repeatability. Eight alleles per marker shown in Table 5.3 was higher than the average of 5.9 previously reported in elite sorghum lines (Smith *et al*., 2000) but lower than the average reported in the inbreds of sorghum (Menz *et al*. 2004) who reported 8.7 alleles per locus. The average

number of alleles revealed per SSR locus detected was higher to that detected by Schloss et al. (2002). This could be due to levels of polymorphism of SSR markers, the diversity of genotypes and the sensitivity of DNA fragment separation systems. Mean level of heterozygosity per SSR marker of 0.17 in Table 5.3 is similar to what was reported by Ngugi and Moraa (2012). PIC provides an estimate of the discriminatory power of a locus or loci by the number of alleles expressed and the relative frequencies of those alleles. According to PIC values, 2 markers (Xcup 53 and Xcup 63) were slightly informative (PIC<0.25 with mean = 0.095), 2 markers (Xtxp 273 and Xtxp 145) were reasonably informative (0.25<PIC<0.5, mean PIC = 0.42), 5 markers (mSbCIR 246, Xcup 14, Xcup 12, mSbCIR 283 and mSbCIR 329) were highly informative (0.5<PIC>0.75, mean PIC = 0.64). The SSR Xcup 63 has been identified as rare in an earlier study (Ali *et al.*, 2008). The F_{ST} value of 0.034 in Table 5.4 indicates negligible genetic differentiation among the population analyzed. F_{ST} values up to 0.05 indicate negligible genetic differentiation whereas >0.25 means very great genetic differentiation within the population analyzed. This demonstrates that sweet sorghum cultivars from Argentina, Brazil, Kenya, United States of America and Zambia, are very closely related. Efforts to widen genetic variability and improve sweet sorghum through exchange of cultivars for breeding within these countries are unlikely to yield useful results. Similar observation had also been made by Geleta *et al*. (2006)

Marker series Xtxp 012, Xtxp 021 which were of genomic origin were highly polymorphic compared to gene-based marker series Xcup 53 and Xcup 63 (Table 5.3). Genic SSRs have been reported to be less polymorphic compared with genomic SSRs in crop plants because of greater DNA sequence conservation in transcribed regions (Schloss *et al*., 2002).

SSRs with di-nucleotide repeats are the most polymorphic marker class followed by tri-, tetra- and penta-repeat units. A direct relationship exists between marker information content and the

number of repeat units (Weber 1990; Innan *et al*., 1997; Schloss *et al*., 2002). The gene diversity observed in this study (Mean PIC = 0.53) is closer to the diversity value (0.40, 0.46, 0.62, 0.58) reported by Ali *et al.* (2008), Schloss *et al.* (2002), (Agrama & Tuinstra, 2003) and Smith *et al.* (2000), respectively. The SSR loci mSbCIR 329, Xcup 14 and Xcup 12 were rich in allelic diversity exhibiting (9-19) alleles with highest PIC of 0.89. Thus, these primers could be of great use in DNA fingerprinting to characterize sweet sorghum genetic stocks in view of the emerging needs for Distinctiveness, Uniformity and Stability (DUS) characterization and plant varietal registration.

5.6 Conclusions

Pairs of genotypes, which can be exploited to select parents for hybrid development to maximize the sugar content and total biomass and for development of segregating populations to map genes controlling sugar content in sweet sorghum, were identified (Crossing genotypes in cluster I with the ones in cluster II). The results presented here demonstrate that sweet sorghum cultivars from Argentina, Brazil, Kenya, United States of America and Zambia, are very closely related. Efforts to widen genetic variability and improve sweet sorghum through exchange of cultivars for breeding within these countries are unlikely to yield useful results.

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CHAPTER SIX

6.0 GENOTYPE × ENVIRONMENT INTERACTION ON SUGAR AND BIOMASS PRODUCTION IN SWEET SORGHUM (*Sorghum bicolor (L*). Moench) IN WESTERN KENYA

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6.1 Abstract

Genotype x environment interaction was determined from field experiments conducted to evaluate sweet sorghum genotypes in Western Kenya during the 2011, 2012, 2013 rainy season of April to July at Alupe, Kibos, Homa Bay and Spectre International farm and crosses evaluated in 2014. The materials used in the study consisted of sixteen sweet sorghum genotypes and two sorghum genotypes sourced from ICRISAT and KARI. The treatments were laid out in a Randomized Complete Block Design (RCBD) and replicated three times. Data were collected on sorghum traits in accordance with the procedure outlined in the ICRISAT sorghum descriptor. The study revealed that genotype by environment interaction had significant influence on most of the traits. This indicates that selection for plant height, girth, brix juice, juice volume and stalks weigh cannot be carried out across the four environments, suggesting that selection for these traits have to be carried separately in each of the four environments.

Key words: Biomass, brix, environment, genotype, sweet sorghum.

6.2 Introduction

Genotype x Environment interaction can be defined as the differential response of varying genotypes under change(s) in the environment (Mather and Caligari, 1976). It refers to instances where the joint effects of genotype and environment are significantly greater or significantly

reduced, than would be predicted from the sum of the separate effects (Andrew *et al*., 1998). In order to exploit the existing variability and develop new high yielding cultivars, sorghum improvement efforts under diverse environmental conditions are needed (Faisal and Aisha, 2011). There are many reports on G × E and stability studies in sorghum (Majisu and Dogget, 1972; Chapman *et al.,* 2000; Haussmann *et al.,* 2000; Kenga *et al.,* 2004). Studying G × E for yield using 12 sorghum genotypes of diverse origin across 25 environments, Alagarswamy and Chandra (1998) found that 12% of the variation was due to genotypes, 61% due to environment while G × E accounted for 27%. Chapman *et al.* (2000) reported that most of the G × E in sorghum was a result of the genotype by location by year, but suggested breeders to deal with the genotype by location type over a fixed number of seasons.

The prevalence of environmental causes of variation over the genetic effects does not suggest that the importance of genotype should be minimized (Faisal and Aisha, 2011). However, global warming and climatic changes will reduce the productivity of many crops around the world. So that a considerable attention should be given to the effect of genotype- environment interaction in the plant breeding programs especially in the developed countries (Ghazy.Mona *et al.,* 2012). Developing high yielding cultivars is mainly depending upon existing genetic variation among the germplasm under existing breeding programs. The relative performance of cultivars for quantitative traits such as yield and the other characters, which influence yield, vary from an environment to another. Consequently, to develop a variety with high yielding ability and consistency, attention should be given to the importance of stability performance for the genotypes under different environments and their interactions (Ghazy.Mona *et al*., 2012). The interaction between genotype and environment has an important bearing on breeding for better

varieties (Allard and Bradshaw, 1964). It is therefore important to conduct multi-location testing, quantify G × E and conduct stability analyses to select superior materials in sorghum.

6.3 Objectives

The objective of the study was to investigate the influence of genotype by environment interaction on sugar and biomass yield of sweet sorghum in Western Kenya.

6.4 Materials and Methods

6.4.1. Test materials

A total of sixteen varieties and two checks from ICRISAT (IESV 92038/2-SH, NTJ2, IESV 92008 DL, IESV 93042-SH, IS 2331, IESV 91-018 LT, IESV 91104 DL, IESV 93046, Kenya Agricultural Research Institute (KARI) (KARI Mtama 2, GADAM,) Argentina (Malon, Paisano, Argensor 151 DP, Argensor 165 BIO) and United States of America(NK 5989-29005, NK 7829-29006, NK 8416-19075, NK 8830-29007) were evaluated in Randomized Complete Block Design with three replications in three seasons; 2011, 2012 and 2013 for seasons one, two and three respectively. Ten parents and their crosses were also evaluated in three environments in 2014.

Each entry was raised in four rows of 3 m length with a spacing of 70 cm × 20 cm. Sowing was done manually by placing 3 seeds in holes spaced 20 cm apart. Data were obtained from plants harvested from the two inner rows of each plot. Care was taken to reduce border effects due to unequal competition of cultivars by the appropriate use of sorghum buffer rows. Nitrogen fertilizer was added at a rate of 100 kg N/ha. All the package of practices were followed to raise a good and healthy crop.

6.4.2.Study sites

Four study sites were used; Kibos, CYMMIT farm; altitude 1190 meters above the sea level (masl), average daily temperature is 24 ^{0} C, rainfall per annum is 1441mm and the soils are planosol. Alupe; altitude is 1165 masl, average daily temperature is 22.2 ^{0} C, rainfall per annum is 1550 mm and the soils are acrisol. Spectre International farm-Kisumu**,** The soil type is chromic vertisol described as poorly drained, very deep, very dark grey to black, very firm, cracking clay. The average daily temperature is 23.1ºC. The annual average rainfall per annum is 1353mm. The altitude is 1164 masl. Homa Bay; soil types are black cotton, cracking and swelling montmorillonite. The altitude is 1190 masl. The mean daily temperatures are 25.8 ºC. The annual rainfall per annum ranges from 900 – 1200mm. The materials were evaluated for three seasons. Data collected included days to 50 per cent flowering, plant height (cm), stem thickness (cm), cane weight (g), juice volume (ml), brix % at 75, 90 and 120 days after planting, pol % juice, purity% juice, panicle height at harvest (cm), panicle diameter (cm) and 100-seed weight (g).

Sampling was done in the following manner: Flowering date was recorded when 50 % of the plot had flowered. The length of the plant from the ground to the panicle tip was measured to estimate plant height. Stem diameter was measured 20 cm above ground. The juice volume was measured using measuring cylinder. The fresh main stalk was pressed and 2–3 droplets of juice were collected on a sucrose- sensitive refractometer to measure the brix. Pol analysis was done using polarimetric method. Six gm of basic lead acetate was added to 300 ml of juice in clarification process. The juice was filtered through a Whatman filter paper No. 91. The pol reading was then fitted in the formula below to obtain Pol at 20^{0}C; POL20 = PT {1+ 0.000185(T-20) - 0.000003 (T-20)2}.

6.4.3.Data analyses

The data obtained on all the characters over four environments and three seasons was subjected to GenStat 14th edition to perform the analysis of variance (ANOVA). The analysis used the Linear Model for randomized completely block design.

$Y_{ij} = \mu + r_i + g_j + e_{ij}$ Where: Y_{ij}= Observed effect for i^{th} replication and j^{th} genotypes

μ = grand mean of the experiment r_i= effect due to the ith replication

g_j= effect due to jth genotype e_{ij} = effects due to the residual or random error of the experiment

Other analysis done included AMMI and IPCA.

6.5 Results

6.5.1. Performance of genotypes based on brix and biomass

Higher Brix value during 2011 season one (Table 6.1) was obtained from sweet sorghum cultivated in Kibos (13.9).Among the genotypes subjected to evaluation, IS2331 recorded highest brix in Kibos (17.19). Pol percentage (Table 6.1) which is an indicator of sucrose percent varied from 5. 83 to 6.65 percent (environmental mean) across the two environments. Genotype IS2331 showed the highest pol % (10.8) while the lowest pol percentage was recorded by IESV 91018 LT (3.4%).The high coefficient of variation for purity % and pol % of 39% and 46% respectively in Kibos can be attributed to large variation among the genotypes with respect to the two attributes. For instance, the top performing genotype IS 2331 recorded purity % of 61.9 and pol % of 10.8 while the lowest performing genotype IESV 92008 DL registered purity % and pol % of 23.4 and 4.1 respectively under the same environment.

IESV 93046 and IS 2331 were the tallest varieties across the four locations (Table 6.2) registering mean height of 269.64 cm and 252.76 cm respectively. IESV 93046 was the best performing genotype in terms of juice volume (1199 ml) and brix % (14.2)

Environment wise Homa Bay was best performing registering highest genotypic mean of brix % and pol % of 14.8 and 8.8 respectively (Table 6.3).

Purity is important when sugar is to be produced from the juice. Alupe, Kibos, Homa Bay and Spectre environments varied for purity percent (Table 6.3) as evident from the varying environment mean (21 to 58.6 %). Purity percent was maximum in Homa Bay (58.6) and the least was observed in Kibos (21). Among the genotypic means for purity IESV 93046 exhibited the highest value of 73.6 % in Homa Bay.

From Table 6.3 Spectre environment registered the maximum environmental mean (590.2 ml) in terms of juice yield, whereas Alupe environment was the least favored (384ml). Among the test genotypes, maximum juice yield was recorded by IESV 93046 (1550 ml) at Spectre International farm.

Table 6.1: Performance of genotypes for sugar and biomass related traits across two environments during 2011 season one

	Brix % juice		Girth 3(mm)		Height 3(cm)		Purity %		Pol % juice 2	
Location	Alupe	Kibos	Alupe	Kibos	Alupe	Kibos	Alupe	Kibos	Alupe	Kibos
GADAM	9.34	11.23	14.6	16.6	105.33	110.33	27.23	44.43	2.74	4.82
IESV 91018 LT	9.66	10.84	15.5	16.5	210.67	212.67	28.35	32.13	3.49	3.60
IESV 91104 DL	12.9	13.75	14.53	17.73	190.33	181.33	42.2	61.11	5.73	11.07
IESV 92008 DL	15.27	15.07	14.5	17.15	167.33	188.33	59.06	23.42	9.10	4.10
IESV 92038/2 SH	13.4	12.93	14.5	18.37	165..67	173.67	42.67	48.2	5.78	6.17
IESV 93042 SH	11.93	12.56	15.6	16.6	163.67	175.67	47.04	43.7	6.49	5.86
IESV 93046	13.06	14.43	13.87	17.87	270.00	278.20	51.07	57.99	6.78	8.34
IS 2331	15.86	17.19	15.47	17.47	261.00	272.33	62.56	61.97	9.91	10.84
KARI MTAMA 2	11.24	13.35	14.63	15.63	171.50	181.00	34.05	52.2	3.92	7.03
NTJ 2	11.44	14.21	13.97	16.97	200.24	237.67	38.18	30.47	4.41	4.63
Mean	12.41	13.956	14.717	17.08	193.34	201.12	43.24	45.56	5.83	6.65
LSD	4.707	2.356	1.728	1.604	15.75	14.65	29.2	31.03	5.03	5.30
CV%	22.11	9.84	6.84	5.76	11.35	11.15	39.36	39.71	50.23	46.53

Table 6.2: Mean of agronomic and quality parameters of sweet sorghum genotypes across locations in 2012 season two at 120 days after planting

Variety	Height (cm)	Girth (mm)	Brix (%)	Pol (%)	Juice Volume(mm)	Purity (%)	Grain Weight (g)	Stalk Dry weight (g)
ARGENSOR 151 DP	139.49	18.3	9.45	4.01	452	37.68	203.6	346.9
ARGENSOR 165 BIO	217.49	19.5	11.11	4.78	796	39.44	302.4	659.4
GADAM	107.01	18.3	9.43	4.32	215	40.87	146	300.6
ICSV 91018 LT	220.53	22.5	7.56	2.34	1161	26.41	250	567.7
ICSV 9104 DL	196.71	20.3	12.7	6.74	607	46.76	269.6	402.3
ICSV 92008 DL	178.85	19.2	12.56	6.44	698	45.52	313.4	456.4
ICSV 92038/2 SH	166.55	20.5	11.56	5.72	536	43.38	286	532.8
ICSV 93042 SH	167.6	20	11.08	5.52	532	44.45	237.8	414.5
ICSV 93046	269.64	20.4	14.24	9.47	1199	57.30	290.7	448.2
IS 2331	252.76	19.2	13.32	7.72	694	53.00	251.7	587.8
KARI MTAMA 2	167.88	18.7	11.11	6.05	190	45.68	183.7	404.1
KS 5989-29005	120.18	21.2	9.77	4.73	232	43.64	219.1	420.6
MALON	117.06	20.5	8.91	3.87	279	37.33	220.6	417.4
NK 7829-29006	95.59	22	8.94	5.48	168	47.63	202.9	383.3
NK 8416-19075	114.9	18.3	9.04	4.64	111	44.91	192.3	295.6
NK 8830-29007	100.86	21.3	8.84	4.13	178	42.07	203.2	384.0
NTJ 2	202.47	20	10.12	4.07	731	36.09	273	453.4
PAISANO	115.18	21.3	9	4.22	411	42.51	252	555.4
P-values	0.016	<.001	<.001	<.001	0.035	<.001	<.001	<.001
Lsd	21.194	2.20	2.307	2.432	317.8	13.59	134.20	159.61
Sed	10.721	1.11	1.167	1.229	160.8	6.87	67.89	80.74
CV %	8.0	6.8	13.6	28.8	38.6	19.6	34.8	26.3

Table 6.3: Mean of quality parameters of sweet sorghum genotypes by locations in 2012 season two at 120 days after planting

	Brix (%)				Pol (%)				Juice Volume(ml)				Purity %			
VARIETY	**AL**	**HB**	**KB**	**SP**	**AL**	**HB**	**KB**	**SP**	**AL**	**HB**	**KB**	**SP**	**AL**	**HB**	**KB**	**SP**
ARGENSOR 151 DP	7.37	14.21	8.97	7.24	2.57	8.46	2.84	2.15	147	372	867	422	35.1	59.5	27	29.5
ARGENSOR 165 BIO	10.02	14.77	10.13	9.52	3.97	8.24	3.49	3.43	537	923	843	880	32.3	55.1	34	36.1
GADAM	11.14	12.25	5.17	9.17	5.41	7.18	0.50	4.18	143	212	328	178	46.5	57.5	14	45.5
IESV 91018 LT	10.57	9.17	3.90	6.60	4.09	2.96	0.44	1.85	890	1293	1103	1357	38.4	31.1	8.9	27.2
IESV 9104 DL	17.11	16.35	6.11	11.24	11.52	9.34	1.23	4.89	718	680	390	640	67.1	57.1	20	43.4
IESV 92008 DL	13.10	17.25	7.45	12.45	6.91	10.91	1.52	6.42	555	688	640	908	52.4	62.3	16	51.6
IESV 92038/2 SH	13.99	16.22	6.06	9.97	7.78	9.86	1.01	4.23	463	635	468	577	55.7	60.7	15	42
IESV 93042 SH	14.37	14.23	5.43	10.27	8.83	7.59	1.10	4.54	463	650	299	717	59.6	53	21	44.1
IESV 93046	17.26	18.51	5.34	15.83	12.07	13.63	0.84	11.34	933	1423	890	1550	69.4	73.6	15	71.2
IS 2331	16.83	16.83	8.04	11.57	11.70	11.12	2.31	5.77	612	885	493	787	68.6	66	28	49.9
KARI MTAMA 2	11.46	18.56	5.62	8.78	6.71	12.50	1.46	3.51	90	157	228	283	51.6	67.4	24	39.4
KS 5989-29005	9.13	15.14	5.88	8.95	4.51	9.52	1.53	3.37	137	188	303	300	49	62.9	25	37.5
MALON	6.57	14.21	5.96	8.89	3.12	8.29	0.87	3.19	133	317	279	385	42.1	57.8	14	35.3
NK 7829-29006	5.44	14.46	6.32	9.52	6.48	9.14	1.89	4.39	85	183	193	210	54.6	63.2	28	44.7
NK 8416-19075	7.46	13.58	6.61	8.52	5.64	7.37	1.80	3.72	52	98	215	78	51.9	55.9	28	44.2
NK 8830-29007	6.91	14.13	5.72	8.61	3.03	8.45	1.38	3.64	103	227	200	183	43.2	59.4	23	42.3
NTJ 2	11.74	12.94	6.21	9.61	5.67	6.25	0.79	3.57	540	943	607	833	47.7	48.1	12	36.5
PAISANO	6.58	13.66	7.34	8.42	2.37	8.81	2.08	3.63	320	452	538	335	35.3	64.4	28	42
Location Means	**10.95**	**14.80**	**6.46**	**9.73**	**6.24**	**8.87**	**1.50**	**4.32**	**384.0**	**573.7**	**493.6**	**590.2**	**50.0**	**58.6**	**21**	**42.36**
P-values	<.001	<.001	<.001	<.001	<.001	<.001	<.001	<.001	<.001	<.001	<.001	<.001	<.001	0.004	<.001	<.001
Lsd	2.674	2.746	3.492	1.960	3.009	2.645	3.406	1.828	147.9	195.5	218.5	210.8	15.03	20.59	11.35	13.63
Sed	1.314	1.350	1.718	0.9645	1.467	1.454	1.674	0.8995	72.77	96.11	107.5	103.7	7.314	10.02	5.655	6.708
CV %	14.47	17.18	17.25	13.06	26.81	31.89	27.25	26.24	23.18	17.05	21.95	21.01	16.80	23.36	11.92	19.38

AL=Alupe, HB=Homa Bay, KB=Kibos and SP=Spectre International Farm

Among the parents and crosses evaluated in 2014, for Alupe, the top stem brix performers at 90 days were parental lines IESV 93046 (21.2%) and IS 2331(20.5%) followed by two hybrids NK8416-19075xIESV92008DL (20.30%) and PAISANOxIESV92008DL (20.30%) (Table 6.4). Grain weight yields per six panicles ranged from as low as 66.5 grams of the hybrid NK8416-19075xIESV 93046 to as high as 275.50 grams of the hybrid PAISANOxIESV92008 DL. For plant height, the tallest entry was hybrid MALONxIS2331 (297.0 cm) and the shortest was a cross involving GADAMxIESV93046 (125cm). In terms of girth, cross involving GADAMxIS2331 registered the biggest girth (1.8cm) and the thinnest entry was PAISANOxIESV 93046 (1.43cm).

In Homa Bay (Table 6.5) the top stem brix % performers at 90 days were two hybrids MALONxARGENSOR 165 BIO and PAISANOxNTJ2 (20.3 %). Grain weight yields per six panicles ranged from as low as 269 grams of the hybrid KS5989-29005XIS233 to as high as 515.00 grams of the hybrid MALONxIESV92038/2-SH.For plant height, the tallest entry was hybrid NK8416 19075xIESV92038/2-SH standing at 279 cm and the shortest entry was NK8416-19075x IESV93042-SH(134cm) . In terms of girth, hybrids performed better than parental lines with entry NK8416-19075x IESV 92008 DL leading at 1.8 cm.

In Kibos (Table 6.6) the top stem brix performer at 90 days was NTJ 2 (21.3 %) followed closely by MALONxNTJ 2 (20.75%). Grain weight yields per six panicles ranged from as low as 161.50 grams of the parent IESV 93046 to as high as 532.00 grams of the hybrid PAISANOxIESV92038/2-SH.For plant height, the tallest entry was hybrid NK8416-19075xIESV92038/2-SH (283 cm) and the shortest entry was MALON (125 cm). In terms of girth entry IESV 92008 DL led at 1.88 cm and the thinnest entry was a cross KS5989-2900xIS 2331(1.44cm).

Mean performance of entries across locations in 2014 (Table 6.7) showed that the top three stem brix performers at 90 days were two parents NTJ 2(20.65) and IESV 93046(20.60%) and a hybrid MALONxNTJ 2(20.57%). In terms of grain weight per six panicles the best performing entry was hybrid PAISANOxIESV92038/2-SH (410.3 grams) and the least performance in terms of grain was recorded by IESV93046 (152 grams). The tallest entry was a hybrid MALONxIS2331 (261.1 cm) and the shortest entry was a cross between GADAMxIESV92038/2-SH (150.3 cm). Based on girth, KS 5989-29005xIESV93046 led at 1.74 cm and the thinnest entry was KS 5989-29005x IS2331 with a mean girth of 1.47cm.

Table 6.4: Mean performance of parents and hybrids for sugar and biomass related traits in Alupe in 2014

VARIETIES	Brix 75DAP	Brix 90DAP	Fld wt/6 pncl	Girth 60DAP	Girth 90DAP	Grn wt/6 pncl	Plt ht 60	Plt ht 90	Pncl dmt	pncl ht harv	100 SW
GADAM	15.60	19.10	275.50	1.54	1.64	192.00	116.00	137.00	4.85	22.20	2.35
GADAMxIESV92008DL	15.40	18.60	277.00	1.49	1.54	195.00	103.00	145.50	4.85	19.80	2.49
GADAMxIESV92038/2-SH	15.50	18.60	328.50	1.66	1.63	226.00	100.00	125.00	5.45	21.10	2.16
GADAMxIESV93046	15.85	19.85	174.50	1.56	1.68	94.00	113.30	158.00	4.55	22.10	2.18
GADAMxIS2331	15.20	19.25	254.00	1.58	1.81	181.00	113.00	152.30	4.40	20.90	2.48
GADAMxNTJ2	14.95	19.45	225.50	1.57	1.66	135.00	115.00	171.30	4.45	18.80	2.17
IESV92008DL	15.45	19.60	296.50	1.35	1.44	212.50	122.80	179.00	5.45	21.60	2.18
IESV92038/2-SH	16.20	20.25	247.50	1.71	1.86	170.50	129.80	215.00	5.05	20.70	2.50
IESV93046	15.45	21.20	220.50	1.49	1.66	134.50	119.40	160.90	4.90	22.70	1.84
IS2331	15.10	20.55	153.50	1.51	1.59	96.50	105.00	150.20	4.55	20.50	2.30
KS5989-29005	15.55	19.50	241.00	1.56	1.55	169.00	115.60	165.00	4.55	26.20	2.11
KS5989-29005xIESV92008DL	15.45	18.95	235.50	1.51	1.61	158.50	113.60	164.00	4.15	21.10	2.55
KS5989-29005xIESV92038/2-SH	15.50	19.30	194.00	1.57	1.63	122.00	105.20	174.00	4.15	18.80	2.30
KS5989-29005xIS2331	15.55	18.90	190.50	1.49	1.53	104.00	93.00	171.60	4.50	24.90	1.46
KS5989-29005xNTJ2	15.40	19.00	210.50	1.44	1.59	119.00	107.70	173.70	4.65	22.90	2.02
KS598929005xIESV93046	15.65	19.45	222.50	1.67	1.74	185.00	110.00	159.00	4.55	20.70	2.21
MALON	14.90	19.90	207.00	1.41	1.62	125.50	89.00	134.00	5.00	23.10	1.91
MALONxIESV92008DL	15.10	19.75	299.00	1.52	1.67	204.00	107.70	172.70	5.35	21.90	2.36
MALONxIESV92038/2-SH	14.85	19.55	306.00	1.48	1.53	221.50	129.80	190.00	5.15	22.50	2.41
MALONxIESV93046	15.35	19.60	161.50	1.18	1.48	199.00	115.40	182.90	4.10	19.10	1.92
MALONxIS2331	15.35	19.00	356.50	1.48	1.55	99.00	123.90	297.00	4.65	21.10	2.13
MALONxNTJ2	15.75	20.20	222.00	1.45	1.66	145.00	122.50	187.00	4.54	20.80	2.67
NK8416-1905xIESV92008DL	15.40	20.30	238.00	1.34	1.77	119.00	119.00	166.00	4.70	22.40	2.31
NK8416-1905xNTJ2	15.05	20.00	218.50	1.55	1.59	143.50	122.20	177.00	4.35	23.20	2.13
NK8416-19075	15.20	19.00	241.00	1.56	1.54	166.50	119.50	138.60	4.75	25.30	2.11
NK8416-19075xIESV92038/2-SH	15.15	19.25	233.50	1.58	1.53	168.00	142.30	204.00	4.75	24.50	2.17
NK8416-19075xIESV93046	16.15	19.75	171.00	1.69	1.61	66.50	132.80	170.00	4.30	20.00	1.97
NK8416-19075xIS2331	15.70	18.65	214.50	1.72	1.75	139.50	112.60	161.90	4.45	22.70	2.15

NTJ2	15.45	19.25	298.00	1.67	1.70	202.50	119.60	163.10	4.50	20.10	2.56
PAISANO	14.70	19.55	304.50	1.51	1.63	169.50	104.80	150.50	5.65	23.70	1.93
PAISANOxIESV92008DL	15.10	20.30	349.00	1.45	1.58	275.50	108.00	185.90	4.90	21.10	2.29
PAISANOxIESV92038/2-SH	15.45	19.55	264.50	1.42	1.57	168.50	91.80	160.00	4.80	21.20	2.44
PAISANOxIESV93046	15.40	19.00	193.50	1.23	1.43	118.50	100.10	151.90	4.55	20.10	2.01
PAISANOxIS2331	15.45	19.10	210.00	1.38	1.56	128.00	133.10	187.00	4.20	20.50	1.87
PAISANOxNTJ2	15.75	19.30	256.00	1.46	1.63	132.00	114.00	151.20	5.25	27.50	2.29
SEREDO	16.10	19.95	290.00	1.45	1.45	196.00	119.50	170.70	4.55	39.70	1.85
L.S.D	0.87	1.59	127.10	0.36	0.25	113.76	34.1	55.1	1.33	4.83	0.94
C.V %	2.8	3.9	25.7	12	7.8	36.2	14.7	16.2	14.0	10.9	21.4

DAP=Days after planting, Fld wt/6 pncl=Field weight of six panicle, Grn wt/6 pncl=Grain weight six panicles**,** Pncl dmt=Panicle diameter, Pncl ht harv =Panicle height at harvest, 100 SW =100 seed weight, LSD=Least significant different, C.V =Coefficient of variation

Table 6.5: Mean performance of parents and hybrids for sugar and biomass related traits in Homa Bay in 2014

Varieties	Plt ht 60DAP	Plt ht 90DAP	Girth 60DAP	Girth 90DAP	Brix 75DAP	Brix 90DAP	pncl ht harv	Pncl dmt	Fld wt/6pancl	Grn wt/ 6pancl	100SW
GADAM	168	178	1.4	1.5	18	18.2	20.0	6.1	549	346	2.7
GADAMXIESV92008DL	184	231	1.7	1.9	18	19.4	17.9	5.1	453	282	2.6
GADAMXIESV92038/2-SH	205	236	1.8	1.8	18	19.0	19.6	6.0	479	282	2.7
GADAMXIESV93046	151	162	1.5	1.6	17	18.9	22.2	5.5	509	338	2.7
GADAMXIS2331	155	193	1.7	1.5	17	19.7	22.2	5.0	464	297	2.5
GADAMXNTJ 2	118	147	1.7	1.6	17	18.4	24.9	5.3	519	332	2.3
IESV92008DL	155	175	1.3	1.7	16	18.9	21.4	5.0	505	326	2.3
IESV92038/2-SH	207	268	1.3	1.7	17	19.1	21.1	5.1	476	425	2.3
IESV93046	201	244	1.5	1.7	18	19.7	26.6	6.4	432	295	2.8
IS2331	133	154	4.7	1.6	17	18.4	21.8	7.1	500	342	2.5
KS5989-29005	200	246	1.6	1.7	18	18.7	21.9	7.3	536	378	2.7
KS5989-29005XIESV92008DL	190	230	1.6	1.8	19	19.1	21.8	7.5	599	417	3.1
KS5989-29005XIESV92038/2-SH	180	205	1.6	1.7	18	20.0	25.5	7.2	649	395	2.8
KS598929005XIESV93046	173	221	1.7	1.7	18	19.6	23.1	6.7	466	358	2.9
KS5989-29005XIS2331	198	223	1.4	1.5	19	18.7	24.6	5.9	354	269	2.6
KS5989-29005XNTJ 2	207	232	1.3	1.6	18	19.7	21.8	6.3	446	333	3.2
MALON	166	189	1.7	1.7	19	19.7	23.9	7.4	759	482	2.9
MALONXARGENSOR 165 BIO	203	262	1.5	1.7	18	20.3	22.2	7.2	309	363	2.7
MALONXIESV92008DL	176	199	1.4	1.7	18	19.6	22.8	6.7	580	376	2.8
MALONXIESV92038/2-SH	161	232	1.8	1.7	18	18.9	23.7	7.7	802	515	2.9
MALONXIESV93046	176	204	1.3	1.6	16	19.8	21.5	6.8	503	345	3.1
MALONXIS2331	136	183	1.6	1.6	17	18.9	24.4	5.9	345	217	2.5
NK8416-1905XIESV92008DL	131	177	1.8	1.8	17	18.8	25.0	7.0	620	385	3.5
NK8416-1905XIESV93042-SH	125	134	1.4	1.5	16	18.7	27.3	6.7	585	371	2.4
NK8416-19075	183	229	1.5	1.7	17	19.3	27.5	5.7	669	424	2.4
NK8416-19075XIESV92038/2-SH	201	279	1.6	1.6	17	19.1	25.3	6.3	556	332	2.4
NK8416-19075XIESV93046	176	213	1.5	1.7	17	19.4	23.3	6.0	555	348	2.9

NK8416-19075XIS2331	150	166	1.3	1.6	16	19.2	25.3	6.5	558	385	2.6
NTJ 2	179	249	1.7	1.6	17	20.0	22.6	6.9	594	362	3
PAISANO	165	197	1.7	1.7	17	19.7	23.7	6.3	636	423	2.9
PAISANOXIESV92038/2-SH	206	260	1.7	1.7	16	20.0	24.0	6.0	455	260	2.9
PAISANOXIESV93042-SH	188	221	1.6	1.7	17	20.0	26.6	6.5	577	385	2.8
PAISANOXIESV93046	214	244	1.6	1.5	18	20.0	24.7	5.5	429	273	2.8
PAISANOXNTJ 2	189	227	1.6	1.6	16	20.3	21.7	6.1	448	375	2.7
SEREDO	185	207	1.6	1.8	17	19.3	22.9	6.7	589	402	3.1
L.S.D	63.2	69.6	1.69	0.19	2.76	1.32	4.84	1.44	245.3	187.3	0.82
C.V %	18.0	16.4	12.8	6.0	7.9	3.4	10.2	11.5	23.0	26.5	14.9

DAP=Days after planting, Fld wt/6 pncl=Field weight of six panicle, Grn wt/6 pncl=Grain weight six panicles**,** Pncl dmt=Panicle diameter, Pncl ht harv =Panicle height at harvest, 100 SW =100 seed weight, LSD=Least significant different, C.V =Coefficient of variation

Table 6.6: Mean performance of parents and hybrids for sugar and biomass related traits in Kibos in 2014

VARIETIES	Brix 75DAP	Brix 90DAP	Fld wt/6pancl	Girth 60DAP	Girth 90DAP	Grn wt/ 6pancl	Plt ht 60DAP	Plt ht 90DAP	Pncl dmt	pncl ht harv	100SW
GADAM	17.00	18.85	451.80	1.81	1.62	315.50	131.50	171.30	6.00	22.40	3.13
GADAMXIESV92008DL	17.00	19.05	628.30	1.34	1.56	419.50	138.40	156.30	7.25	23.20	2.25
GADAMXIESV92038/2-SH	17.00	18.55	430.30	4.67	1.63	307.00	148.20	163.00	6.30	23.90	2.86
GADAMXIESV93046	17.90	19.00	557.80	1.48	1.67	349.50	144.00	170.50	6.60	21.40	2.52
GADAMXIS2331	17.15	19.70	692.80	1.67	1.69	448.00	162.60	188.90	7.10	23.50	3.03
GADAMXNTJ2	17.00	19.30	567.30	1.67	1.61	398.00	175.40	226.50	7.15	21.30	2.88
IESV92008DL	18.75	19.90	716.30	1.69	1.88	480.50	182.00	204.00	7.00	22.40	3.03
IESV92038/2-SH	17.30	20.45	546.80	1.53	1.62	476.00	180.30	210.00	6.60	24.50	2.96
IESV93046	18.00	20.30	298.30	1.41	1.55	161.50	214.50	276.00	5.70	17.90	2.09
IS2331	17.90	19.50	346.80	1.36	1.57	231.50	200.90	248.90	5.75	19.00	3.56
KS5989-29005	18.05	18.90	402.30	1.28	1.56	239.50	151.40	173.50	5.35	24.20	2.03
KS5989-29005XIESV92008DL	17.25	20.00	521.80	1.63	1.75	290.00	168.20	235.00	6.60	25.70	3.10
KS5989-29005XIESV92038/2-SH	16.45	20.00	582.30	1.60	1.63	364.00	205.10	230.10	6.60	23.80	2.58
KS5989-29005XIS2331	17.70	19.10	337.30	1.44	1.44	245.00	220.30	250.00	5.50	23.30	2.71

KS5989-29005XNTJ2	16.45	19.70	547.80	1.67	1.69	383.50	188.00	218.90	6.50	24.20	2.89
KS598929005XIESV93046	18.00	19.40	562.30	1.62	1.75	341.00	205.10	268.00	7.00	23.20	2.80
MALON	15.35	18.40	519.80	1.43	1.50	320.00	119.50	125.80	5.50	26.20	2.40
MALONXIESV92008DL	17.40	19.05	542.30	1.54	1.75	392.00	209.10	237.70	6.70	22.00	2.99
MALONXIESV92038/2-SH	17.75	19.75	607.30	1.67	1.65	373.50	161.10	208.60	6.60	22.80	2.70
MALONXIESV93046	17.95	19.00	441.30	1.78	1.81	264.50	189.20	273.00	5.50	19.40	2.32
MALONXIS2331	18.50	19.20	377.80	1.32	1.59	255.00	209.60	243.00	6.00	21.30	3.27
MALONXNTJ2	18.55	20.75	571.30	1.28	1.51	371.50	197.70	205.50	6.15	21.40	2.94
NK8416-1905X92008DL	17.65	19.70	552.80	1.54	1.66	358.00	156.00	169.00	4.70	23.20	2.13
NK8416-1905XNTJ2	17.70	20.15	619.80	1.68	1.71	398.50	198.00	252.00	6.40	32.40	2.93
NK8416-19075	17.80	18.65	583.80	1.70	1.57	382.50	142.40	155.30	6.50	26.00	2.28
NK8416-19075XIESV92038/2-SH	17.20	18.80	546.30	1.61	1.60	469.00	207.20	283.00	5.80	25.90	2.36
NK8416-19075XIESV93046	17.30	19.55	445.80	1.49	1.54	296.00	179.30	216.90	5.90	26.00	2.68
NK8416-19075XIS2331	17.25	19.85	494.80	1.61	1.71	359.50	176.20	223.50	6.70	25.50	2.81
NTJ2	18.45	21.35	443.80	1.56	1.69	468.50	192.10	250.00	7.50	27.50	3.81
PAISANO	16.05	18.65	476.80	1.82	1.68	307.50	135.20	160.90	5.40	24.40	3.37
PAISANOXIESV92008DL	16.40	19.50	603.80	1.82	1.81	371.50	171.80	196.60	6.80	19.80	2.55
PAISANOXIESV92038/2-SH	16.60	19.25	814.30	1.65	1.82	532.00	159.80	189.50	7.30	22.15	3.18
PAISANOXIESV93046	17.20	19.30	485.30	1.29	1.69	287.00	206.90	264.00	5.60	20.50	2.34
PAISANOXIS2331	17.69	18.88	669.80	1.38	1.59	461.80	202.10	273.60	6.51	27.70	2.53
PAISANOXNTJ2	17.25	18.90	366.80	1.28	1.72	244.00	178.30	205.00	6.20	24.50	2.77
SEREDO	17.30	19.05	632.30	1.64	1.66	402.50	166.40	191.90	7.10	24.40	3.02
L.S.D	2.08	1.25	225.6	0.39	0.23	154.1	46.31	52.86	1.86	4.80	0.61
C.V %	5.9	3.2	21.6	12.6	6.9	21.6	13.0	12.2	14.5	10.1	10.9

DAP=Days after planting, Fld wt/6 pncl=Field weight of six panicle, Grn wt/6 pncl=Grain weight six panicles**,** Pncl dmt=Panicle diameter, Pncl ht harv =Panicle height at harvest, 100 SW =100 seed weight, LSD=Least significant different, C.V =Coefficient of variation

Table 6.7: Mean performance of parents and hybrids for sugar and biomass related traits across locations in 2014

VARIETIES	Plt ht 60DAP	Plt ht 90DAP	Brix 75DAP	Brix 90DAP	Girth 60DAP	Girth 90DAP	Pncl dmt	Pncl ht harv	Fld wt/6 pncl	Grn wt/ 6 pncl	100SW
GADAM	126.3	159.8	16.53	18.93	1.723	1.626	5.615	22.34	392.6	274.1	2.866
GADAMXIESV92008DL	126.5	152.7	16.46	18.9	1.394	1.549	6.446	22.06	510.5	344.3	2.327
GADAMXIESV92038/2-SH	132.1	150.3	16.5	18.57	2.609	1.633	6.015	22.97	396.1	279.9	2.622
GADAMXIESV93046	133.7	166.3	17.21	19.28	1.504	1.669	5.914	21.64	429.3	263.9	2.406
GADAMXIS2331	146	176.6	16.5	19.55	1.643	1.725	6.196	22.63	545.7	358.6	2.843
GADAMXNTJ2	155.2	208	16.31	19.35	1.636	1.624	6.246	20.47	452.7	309.9	2.639
IESV92008DL	162.2	195.6	17.64	19.8	1.575	1.73	6.481	22.13	575.6	390.8	2.744
IESV92038/2-SH	163.4	211.7	16.93	20.38	1.589	1.701	6.081	23.23	446.5	373.7	2.806
IESV93046	182.6	237.4	17.14	20.6	1.439	1.587	5.432	19.51	272.1	152.5	2.002
IS2331	168.8	215.8	16.96	19.85	1.412	1.579	5.348	19.51	281.9	186.3	3.133
KS5989-29005	139.4	170.6	17.21	19.1	1.372	1.552	5.082	24.87	348.2	215.9	2.055
KS5989-29005XIESV92008DL	149.9	211.2	16.65	19.65	1.588	1.698	5.78	24.16	425.8	246	2.913
KS5989-29005XIESV92038/2-SH	171.6	211.3	16.13	19.77	1.594	1.63	5.78	22.13	452.1	283	2.486
KS5989-29005XIS2331	177.7	223.7	16.98	19.03	1.483	1.471	5.165	23.84	288	197.8	2.292
KS5989-29005XNTJ2	161.1	203.7	16.1	19.47	1.59	1.658	5.881	23.77	434.7	294.9	2.597
KS5989-29005XIESV93046	173.2	231.5	17.21	19.42	1.637	1.744	6.18	22.37	448.4	288.8	2.601
MALON	109.3	128.5	15.2	18.9	1.426	1.541	5.333	25.16	414.9	254.9	2.234
MALONXIESV92008DL	175.1	215.9	16.63	19.28	1.532	1.723	6.248	21.97	460.7	329	2.774
MALONXIESV92038/2-SH	150.6	202.4	16.78	19.68	1.604	1.609	6.115	22.7	506.3	322.6	2.603
MALONXIESV93046	164.5	242.8	17.08	19.2	1.575	1.698	5.031	19.3	347.5	242.6	2.186
MALONXIS2331	180.9	261.1	17.44	19.13	1.371	1.575	5.548	21.24	370.5	202.8	2.883
MALONXNTJ2	172.5	199.3	17.61	20.57	1.337	1.559	5.611	21.2	454.2	295.7	2.85
NK8416-19075XIESV92008DL	143.6	168	16.9	19.9	1.476	1.694	4.7	22.93	447.3	278	2.189
NK8416-19075XNTJ2	172.6	226.9	16.81	20.1	1.637	1.669	5.714	29.32	485.3	313.1	2.661
NK8416-19075	134.7	149.7	16.93	18.77	1.657	1.562	5.914	25.77	468.9	310.2	2.218
NK8416-19075XIESV92038/2-SH	185.5	256.5	16.51	18.95	1.601	1.578	5.448	25.43	441.4	368.2	2.297
NK8416-19075XIESV93046	163.7	201.2	16.92	19.62	1.553	1.563	5.364	23.99	353.7	219.2	2.441
NK8416-19075XIS2331	154.9	202.9	16.73	19.45	1.646	1.722	5.947	24.57	400.8	285.8	2.584

NTJ2	167.8	220.9	17.44	20.65	1.6	1.693	6.495	25.02	394.9	379.4	3.39
PAISANO	125	157.4	15.6	18.95	1.716	1.661	5.484	24.17	419	261.3	2.885
PAISANOXIESV92008DL	150.4	193	15.96	19.77	1.698	1.733	6.164	20.24	518.4	339.4	2.46
PAISANOXIESV92038/2-SH	137	179.6	16.21	19.35	1.573	1.734	6.463	21.83	630.1	410.3	2.927
PAISANOXIESV93046	171.1	226.4	16.6	19.2	1.27	1.603	5.248	20.37	387.5	230.6	2.225
PAISANOXIS2331	169.3	231.5	16.42	18.77	1.345	1.582	5.57	24.84	499.3	319	2.31
PAISANOXNTJ2	156.8	187	16.75	19.03	1.341	1.687	5.882	25.51	329.6	206.5	2.606
SEREDO	150.7	184.8	16.9	19.35	1.574	1.587	6.246	29.53	517.6	333.4	2.627
L.S.D	47.28	47.0	1.56	0.83	0.56	0.14	1.33	3.14	209.8	148.3	0.58
C.V %	26.9	21.0	8.3	3.8	22.0	7.7	20.3	12.0	42.6	45.6	19.9

DAP=Days after planting, Fld wt/6 pncl=Field weight of six panicle, Grn wt/6 pncl=Grain weight six panicles**,** Pncl dmt=Panicle diameter, Pncl ht harv =Panicle height at harvest, 100 SW =100 seed weight, LSD=Least significant different, C.V =Coefficient of variation

6.5.2. Analysis of variance

Across location and seasons analysis of variance (Table 6.8) showed that genotypes and seasons were significantly different ($P<0.001$) for all the major traits evaluated. Locations x seasons interactions were significantly different ($P<0.001$) for brix % juice, juice volume and purity %. Location x variety interactions were significantly different ($P<0.001$) for girth, stalk weight and juice volume. Higher interactions of location by season by variety was significant ($P<0.05$) for brix %. For brix % juice, genotypes, environments and interactions accounted for 8.9, 31 and 5.5 % of the sum of squares treatment respectively (Table 6.8).

Analysis of variance for Additive Main Effect and Multiplicative Interaction (AMMI) model showed significant differences amongst treatments, genotypes, environments and interactions between genotypes and environments ($P>0.001$) (Table 6.9). For girth, genotypes, environments and interactions accounted for 60.8, 28.1 and 10.9 % of the sum of squares treatment respectively. For brix % juice, genotypes, environments and interactions accounted for 14.4, 69.6 and 16.0 % of the sum of squares treatment respectively. For purity %, genotypes, environments and interactions accounted for 19.1, 60.1 and 20.7 % of the sum of squares treatment respectively.

When the analysis was split into Interaction Principle Component Axes (IPCA), both IPCA-1 and IPCA-2 showed significant different mean brix % juice ($P<0.01$) and captured 62.2 and 25.1 % of the sum of squares for interaction (Table 6.9)

Table 6.8: General ANOVA for sugar and biomass traits across location and seasons

Change	d.f.	Girth	Explained percentage	Stalk weight	Explained percentage	Brix % Juice	Explained percentage	Juice Volume	Explained percentage	Purity %	Explained percentage
Location	3	2308.77***	19.59	1.29**	1.65	817.70***	31.02	310314.00*	1.92	4101.40*	4.62
Season	1	16494.31***	46.65	27.40***	11.67	495.85**	6.27	7617835.00***	15.72	7617.20**	2.86
Location x Season	3	7.89ns	0.07	3.06ns	3.91	104.58***	3.97	607658.00***	3.76	12516.30***	14.10
Location/season/rep	16	4.02**	0.18	0.17ns	1.16	42.26***	8.55	42435.94ns	1.40	622.97*	3.74
Variety	17	19.00***	0.91	5.48**	39.67	41.61***	8.94	992040***	34.80	2120.90***	13.53
Location x Variety	51	2.01***	0.29	0.23ns	4.99	8.53ns	5.50	47190ns	4.97	451.70ns	8.65
Season x Variety	17	4.85***	0.23	1.72***	12.45	12.36ns	2.66	396022***	13.89	776.60ns	4.96
Location x Season x Variety	51	0.45ns	0.06	0.30***	6.51	10.54*	6.80	55770***	5.87	482.20	9.23
Residual	284	1.80	1.45	0.14	16.93	7.32	26.29	27732	16.25	358.50	38.22
Total	427	82.81		0.55		18.52		113491		623.88	

ns=not significant *Significant at 0.05, ** significant at 0.01, *** significant at 0.001

Table 6.9: AMMI ANOVA for sugar and biomass traits across locations

Source	df	Girth	Explained Percentage	Brix % Juice	Explained Percentage	Purity %	Explained Percentage
Treatments	71	9.99***		40.59***		1011***	
Genotypes	17	25.41***	60.88	24.40***	14.40	809***	19.16
Environments	3	66.66***	28.19	668.50***	69.60	14382***	60.11
Block	8	4.19ns		59.36***		1288***	
Interactions	51	1.52ns	10.92	9.05**	16.00	292***	20.73
IPCA I	19	3.28ns	80.39	15.09***	62.26	458***	58.50
IPCA II	17	0.80ns	17.55	6.81ns	25.16	327**	37.36
Residuals	15	0.11ns	2.19	3.93ns	12.80	41ns	4.13
Error	136	2.24		5.23		139	
Total	215	4.87		18.92		470	

ns=not significant *Significant at 0.05, ** significant at 0.01, *** significant at 0.001

Figure 6.1 presents AMMI biplot providing a visual expression of the relationships between the first interaction principal component axis (IPCA1) and means of genotypes and environments based on brix % juice.

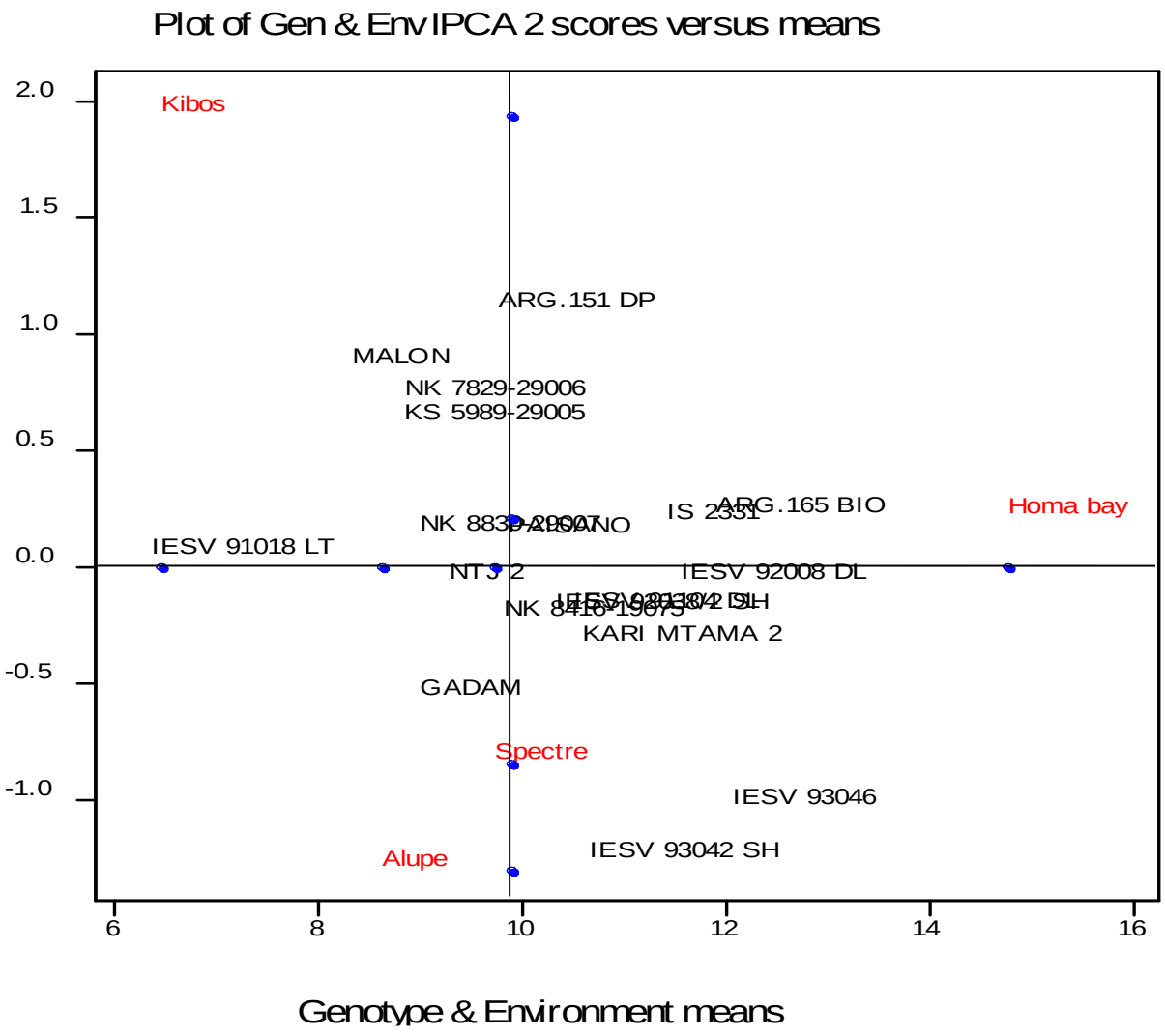

Figure 6.1: AMMI biplot of interaction principal component axis-1 (IPCA-1) against mean brix % juice of 18 genotypes and four environments

The AMMI biplot (Figure 6.1) showed four groupings of genotypes; IESV 91018 LT, generally low brix and stable; NK 7829-29006 and KS 5989-29005, low brix and unstable. The other two groups included NK 8830-29700 and NTJ 2 that had moderate brix yield and stable and IESV

93046 that had high brix yield but unstable. Homa Bay showed high brix yields and high stability while Kibos was low yielding and very unstable environment. However Spectre was more stable than Alupe.

Figure 6.2 presents AMMI biplot providing a visual expression of the relationships between the first interaction principal component axis (IPCA1) and means of genotypes and environments based on girth.

The AMMI biplot (Figure 6.2) showed four groupings of genotypes; NK 8416-19075 thin and unstable; ARGENSOR.151 DP thin and stable. The other two groups included IESV 93046 and NTJ 2 that had moderate girth and stable and NK 7829-29006 and KS 5989-29005 that were thick but unstable. Homa Bay and Alupe showed high stem girth and high stability while Spectre and Kibos were low girth and unstable environments.

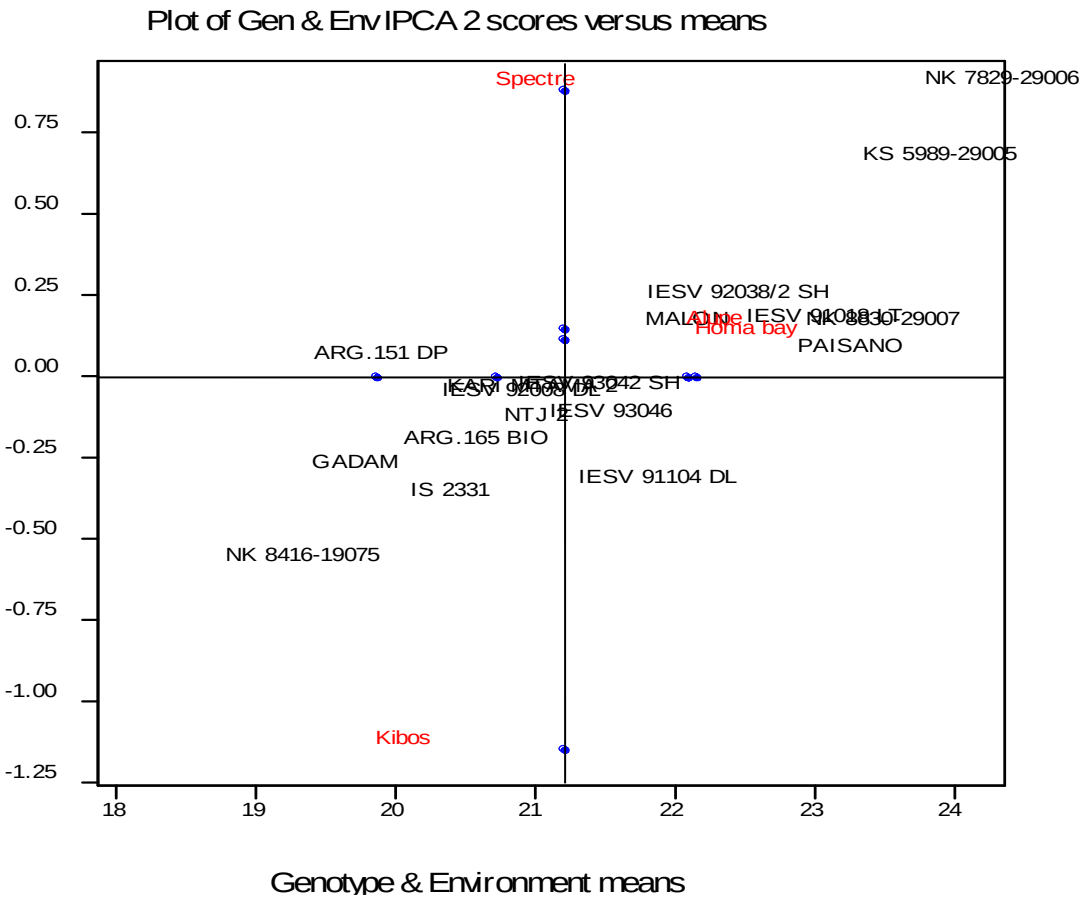

Figure 6.2: AMMI biplot of interaction principal component axis-1 (IPCA-1) against mean girth of 18 genotypes and four environments

Figure 6.3 presents AMMI biplot providing a visual expression of the relationships between the first interaction principal component axis (IPCA1) and means of genotypes and environments based on purity % juice.

The AMMI biplot (Figure 6.3) showed three groupings of genotypes; IESV 91018 LT low purity percent and unstable; IS2331 moderate purity % and stable; IESV 93046 high purity and unstable. Homa Bay environment registered high purity % but was unstable while Kibos recorded low purity % and was equally unstable.

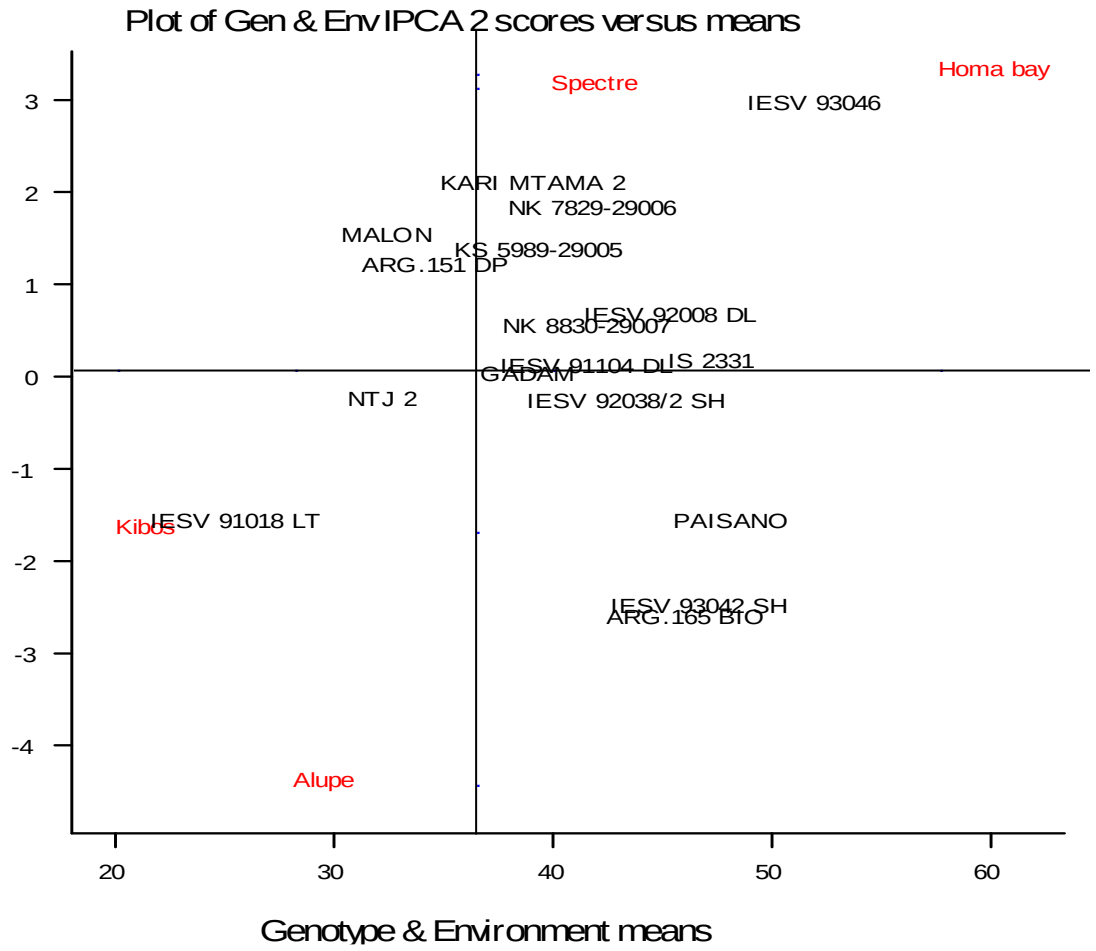

Figure 6.3: AMMI biplot of interaction principal component axis-1 (IPCA-1) against mean purity % of 18 genotypes and four environments

Results from AMMI analysis (Table 6.10) revealed that the best environment was Homa Bay recording the best overall mean for girth, brix % juice and purity percent juice. The best four genotypes in terms of brix were IESV 93046, IESV 92008 DL, IS 2331 and IESV 91104 DL. IESV 93046 can be considered stable and adaptable to wider environments in terms of sugar quality. Kibos consistently registered the lowest genotypes means on the parameters evaluated.

Table 6.10: First four AMMI selections per environment on the basis of girth, brix % and purity

Number	Environment	Mean	1	2	3	4
			Girth (mm)			
1	Homa bay	22.14	NK 8830-29007	PAISANO	KS 5989-29005	MALON
2	Alupe	22.08	NK 8830-29007	PAISANO	KS 5989-29005	MALON
3	Spectre	20.71	NK 7829-29006	KS 5989-29005	PAISANO	NK 8830-29007
4	Kibos	19.86	NK 7829-29006	KS 5989-29005	PAISANO	NK 8830-29007
			Brix%			
1	Homa bay	14.763	IESV 93046	IESV 92008 DL	IS 2331	IESV 91104 DL
2	Spectre	9.731	IESV 93046	IESV 92008 DL	IS 2331	IESV 91104 DL
3	Alupe	8.626	ARG.165 BIO	IESV 93042 SH	NK 8416-19075	PAISANO
4	Kibos	6.46	ARG.165 BIO	ARG.151 DP	IS 2331	IESV 92008 DL
			Purity % Juice			
1	Homa bay	57.6	IESV 93046	NK 7829-29006	IS 2331	KARI MTAMA 2
2	Spectre	39.9	IESV 93046	IESV 92008 DL	IS 2331	GADAM
3	Alupe	28.12	IESV 93042 SH	PAISANO	ARG.165 BIO	GADAM
4	Kibos	20.01	ARG.165 BIO	PAISANO	NK 8416-19075	IS 2331

6.5.3. Discussion

High brix was recorded by some genotypes (Table 6.3), IESV 93046 registered brix of 17.2% in Alupe and IESV 92008 DL registered brix of 17.2 % in Homa Bay. The results are closer to what was observed by Reddy et al (2005) of 16- 23% brix and slightly higher than that observed by Woods (2000) of 11.0-18.5% brix among genotypes evaluated. This variation could be attributed to stalk variety, different soils and climatic conditions.

Purity is important when sugar is to be produced from the juice. Alupe, Kibos, Homa Bay and Spectre environments varied for purity percent (Table 6.3) as evident from the varying environment mean (21 to 58.6). Mean purity percent was maximum in Homa Bay (58.6) and the least was observed in Kibos (21). Among the genotypic means for purity IESV 93046 exhibited the highest value of 73.6 % in Homa Bay. Similar report was given earlier by Woods (2000) where the apparent purity for the sweet sorghum varieties considered varied from 48.2% - 69.7% whereas that of sugarcane juice was 83.6%. Sucrose purity is used to calculate the ease with

which sucrose can be extracted and crystallized and 75% is required as the minimum (Woods, 2001). Among the genotypes evaluated IESV 93056 has potential of being exploited for sucrose extraction and crystallization.

Superior performance of genotypes in Homa Bay (Table 6.3) can be attributed to montmorillonite soils in this environment which are very efficient in nutrient uptake. Genotypes performed better in Spectre International farm than Kibos despite the fact that these environments have similar average daily temperatures and rainfall per annum. Very deep and firm clay soils at Spectre International farm might have contributed to better performance under this environment. Woods (2000) reported that sweet sorghum performance variation could be attributed to different soil conditions.

Good performance observed in crosses over parental lines (Tables 6.4, 6.5, 6.6 and 6.7) for both plant height and stem biomass at all sites could be attributed to the dominant and epistatic gene effects reported for these traits by Sleper and Poehlman (1996) and Rooney and Aydin (1999). In general this observation supports superiority of hybrids over pure line varieties for both sugar and biomass yield which is consistent with previous findings (Blum *et al*., 1990; Corn, 2008). Among males, IESV 93046 and IS 2331 showed consistently higher plant height across the environments (Tables 6.4, 6.5, 6.6 and 6.7). Males are generally taller than females as reported by Naik (1993) and Kadam *et al.* (2000).

Combined analysis of variance (Table 6.8) revealed highly significant ($P \leq 0.001$) variations among environments, genotypes, seasons, genotype x environment and environment x variety x season interaction. This result revealed that there was a differential yield performance among the sweet sorghum genotypes across testing environments and seasons. Maarouf and Moataz (2009) reported variation between sorghum genotypes with respect to fodder production. This indicate

that, simultaneous selection for girth, brix% , stalk weight and purity % is not possible across the four environments and that selection for each location have to be carried out separately. This limit their wider utilization, as reported by Pham and Kang (1988) who stated that, significant G x E for a quantitative trait is known to reduce the usefulness of the genotype means over all locations or environments for selecting and advancing superior genotypes to the next stage of selection.

Across location and seasons analysis of variance (Table 6.8) showed that genotypes and seasons were significantly different ($P<0.001$) for all sugar related traits. Seasons x variety interactions were significantly different ($P<0.001$) for girth, stalk weight and juice volume. Chapman *et al.* (2000) reported that most of the G × E in sorghum was a result of the genotype by location by year, but suggested breeders to deal with the genotype by location type over a fixed number of seasons. This difference among seasons can be attributed to heavy rains received in 2012.

When the interaction between environments and genotypes was significant further analysis was done using Additive Main Effects and Multiplicative Interaction (AMMI) model to determine adaptive response of specific genotypes to specific locations (Annicchiarico, 2002; Egesi and Asiedu, 2002).

Analysis of variance for Additive Main Effect and Multiplicative Interaction (AMMI) model showed significant differences amongst treatments, genotypes, environments and interactions between genotypes and environments ($P>0.001$) (Table 6.9). For brix % juice, genotypes, environments and interactions accounted for 14.4, 69.6 and 16.0 % of the sum of squares treatment respectively. These variations are closer to the ones reported by Alagarswamy and Chandra (1998) while studying G × E for yield using 12 sorghum genotypes of diverse origin across 25 environments. He found that 12% of the variation was due to genotypes, 61% due to environment while G × E accounted for 27%.

6.6 Conclusions

High performance demonstrated by genotypes IESV 93046 and IS2331 for stem brix and stem biomass shows their potential for exploitation for ethanol production.

Homa Bay is the best environment for sweet sorghum production.

The study indicates that selection for girth, brix % juice, purity % and stalks weigh cannot be carried out across the four environments, suggesting that selection for these traits have to be carried separately in each of the four environments.

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CHAPTER SEVEN

7.0 COMBINING ABILITY OF PARENTS AND HYBRIDS FOR SUGAR YIELD AND ITS ATTRIBUTING TRAITS IN SWEET SORGHUM [*Sorghum bicolor* (L.) Moench]

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7.1.Abstract

An investigation was carried out to assess the combining ability and nature of gene action in respect of sugar yield and its attributing traits in 25 new hybrids of sweet sorghum developed by crossing five (5) high sugar lines with five (5) low sugar lines in a North Carolina II mating design and grown in alpha lattice with two replications during long rains season of 2014 in western Kenya. The variance among the lines in respect of their general combining ability (GCA) was highly significant for brix and plant height at 90 days. Specific combining ability (SCA) variance was relatively higher in magnitude for grain weight and plant height indicating predominance of non-additive gene action in the genetic control of these traits. GADAM, MALON and PAISANO among the females and IESV93036, IS2331 and NTJ 2 among males were identified as good general combiners indicating their ability in transmitting additive genes in the desirable direction to their progenies. The best hybrids for total biomass and total sugar content were GADAMxIESV93036, GADAMxIS2331 and MALONxIS2331 and after adequate testing in many locations across the target production environments these hybrids can be recommended for commercial exploitation for ethanol production.

Key words: Combing ability, hybrid breeding, sweet sorghum

7.2. Introduction

Sweet sorghum [*Sorghum bicolor* (L.) Moench] is similar to grain sorghum with a sugar-rich stalk, similar to sugarcane. The sugar content varies from 16-23% brix. Besides having wide adaptability, rapid growth, high sugar accumulation and biomass production potential, it has great potential for jaggery, syrup and biofuel (ethanol) production and the grains from sweet sorghum can be used for food or feed. In any plant breeding programme, combining ability provides necessary information on nature and magnitude of gene action involved which helps for selection of parents for breeding programme. The line × tester mating design helps in assessing the combining ability of parents there by facilitating the selection of superior parents as well as cross combinations (Sprague and Tatum, 1942). An understanding of gene action involved in stem sugar accumulation and the associated traits may help in developing a viable breeding strategy. Schlehuber (1945) reported that genes with partial dominance action controlled sucrose content in hybrids. Baocheng *et al.* (1986) reported that genes with additive and dominance effects influenced stem sugar accumulation. Guiying *et al.* (2000) reported that recessive genes exhibiting additive effects controlled stem sugar accumulation in sorghum. Following a QTL analysis, Natoli *et al.* (2002) reported no significant segregation for genes with major effects on stem sugar percentage. However, studies by Ritter *et al*. (2008) suggested involvement of major genes in addition to genes with minor effects for stem brix. Moderate to high heritabity estimates, ranging between 40% and 96% (Guiying *et al.,* 2000), and predominance of genes with additive effects suggest that brix could be improved through selection.

The general combining ability (GCA) of each parent should be examined when the objective is the development of superior populations, while the specific combining ability (SCA) effects provide information about the performance of hybrids (Cruz and Regazzi, 1994). The differences

in GCA are mainly due to the additive genetic effects and higher order additive interactions, while the differences in SCA are attributed to the non-additive dominance and other types of epistasis (Falconer, 1989). This analysis therefore allows broad inferences on the nature of the gene effects for a trait under selection. A breeder can make use of this information to find the best strategy to select desirable parents or determine which breeding procedure will efficiently improve the performance of the traits of interest (Dudley and Moll, 1969).

Accordingly, the present study was carried out to assess the nature of combining ability and gene action in respect of sugar yield and its attributing traits in 25 hybrids resulting from 10 sweet sorghum parents.

Knowledge of combining ability of the parents, especially for hybrid cultivar development is important for optimizing the breeding strategy. Information on combining ability is needed to identify potentially superior parents and hybrids, and would help to define the pattern of gene effects in the expression of quantitative traits (Goyal and Kumar, 1991). Information on general combining ability studies for sugar yields are limited (Beil and Atkins, 1967; Malm, 1968; Makanda, 2009). However, there are reports of significant GCA and SCA effects for the associated traits, but their level of importance was dependent on the germplasm that was evaluated. Kenga *et al*. (2004) reported that SCA effects were predominant over GCA for grain yield and days to anthesis. Nevertheless, results obtained elsewhere do not necessarily give an indication of the behaviour of the genes in a different environment. Falconer and Mackay (1996) reported that combining ability and heritability information is pertinent to the set of genotypes and the environment where it has been tested.

In sweet sorghum, genetic improvement of sugars has not been intensively studied compared to that of sugarcane. Genetic enhancement of the crop for increased sugar yield is very critical to make sweet sorghum more profitable to the farmers and the industry, while sustaining grain yield, juice volume, plant height, plant girth and other important components. The choice of an efficient breeding programme depends largely on knowledge of the type of gene action involved in the expression of the character. The knowledge on nature of gene action for sugar yield and its component traits like brix% and juice content in the breeding material can provide useful information for selecting proper breeding procedure for future genetic enhancement. Inheritance of stalk biomass, brix percentage and stalk weight in sugar stalk was subject to both additive gene effect and non-additive gene effect, but mainly controlled by non-additive genes Zhou *et al*. (2005). However, the literature regarding inheritance of these traits and their genetic interactions in sweet sorghum is scanty. Several national research programs in the semi-arid regions have shown an increased interest in hybrids (Axtell et al., 1999). The immediate task that faced those breeding programs is to gain information on the combining ability of the various varieties and populations developed and improved over the years.

7.3. Objectives

The objective of this study was to assess the general and specific combining abilities in respect of stem sugar and biomass traits of ten parents and 25 hybrids of sweet sorghum at three locations.

7.4. Materials and Methods

Germplasm

Ten sorghum lines were divided into two groups based on their sugar content (High sugar sweet sorghum and low sugar sorghum).The five high sugar lines were designated as males and crossed with the five low sugar lines designated as females according to North Carolina Design II mating

scheme to generate 25 hybrids. The males were from the International Crops Research Institute for the Semi-Arid Tropics (ICRISAT) India and the female were constituted from introduced lines from Argentina, United States of America and Kenya. 25 hybrids, ten parents and one check were evaluated in three environments.

Experimental sites

Genotypes were evaluated in three study sites ; Kibos, CYMMIT farm; altitude 1190 masl, average daily temperature is 24 ^{0} C, rainfall per annum is 1441mm and the soils are planosols. Alupe; altitude is 1165 masl, average daily temperature is 22.2 ^{0} C, rainfall per annum is 1550 mm and the soils are acrisol. Homa Bay; soil types are black cotton, cracking and swelling montmorillonite. The altitude is 1190 masl. The mean daily temperature is 25.8 ^{o}C. The annual rainfall per annum ranges from 900 – 1200mm. The materials were evaluated during April-July 2014.

Table 7.1: Origin and roles of parental sorghum lines used to generate 25 F1 hybrids in a North Carolina design II mating scheme.

Line no.	Name	Origin	Role in crosses	Principal selection criteria
1	ICSV 93046	ICRISAT India	Male	High sugar
2	IS 2331	ICRISAT India	Male	High sugar
3	NTJ 2	ICRISAT India	Male	High sugar
4	IESV 92008 DL	ICRISAT India	Male	High sugar
5	IESV 92038/2-SH	ICRISAT India	Male	High sugar
6	MALON	Argentina	Female	High sugar
7	PAISANO	Argentina	Female	Low sugar
8	GADAM	KARI-Kenya	Female	Low sugar
9	KS 5989-29005	Argentina	Female	Low sugar
10	NK 8416-19075	USA	Female	Low sugar
11	Seredo	KARI-Kenya	N/A	Check

Experimental design

The experiments was laid out as alpha lattice replicated twice Seeds of each entry were planted at 0.70 m inter-row and 0.20 m intra-row spacing. There were 40 plants per plot. 100 kg ha^1 of calcium ammonium nitrate was used as source of nitrogen. The fields were kept weed free by hand weeding. At planting, stalk borer granules (dimethyl- (2, 2, 2-trichloro-1-hydroxyethyl) phosphonate) were used to control stalk borer damage. Stem sugar concentration was measured in brix %, using a hand-held pocket refractometer. Brix measurements were taken at the middle internode of each selected stalk. Stalk juice was pressed from the cut internode section into the sample stage of the refractometer using a pair of pliers. Both the pliers and the refractometer sample stage were rinsed with clean water and dried with tissue paper before the next sample was

measured to avoid carryover effects. Stem diameter was also measured from the mid internode sections using a veneer caliper. Other measurements that were taken included plant height, 100-seed weight (g) and grain yield per plant (g).

Data analyses

Data were analyzed using REML procedure in GenStat statistical package (Payne et al., 2007) following a fixed effects model: $Y_{ijkl} = \mu + s_i + r_j\,(s_i) + m_k + f_l + mf_{kl} + s_i \times m_{ik} + s_i \times f_{ij} + s_i \times mf_{ikl} + e_{ijkl}$ where: Y_{ijkl} = observed hybrid performance; μ = overall population mean; s_i = effect of the ith site; $r_j\,(s_i)$ = effects of the jth replication in the ith site; m_k= effect of the kth male parent; f_l = effect of the lth female parent; mf_{ikl} = interaction effect of the kth male and the lth female parents; and $e_{ijkl}$is the experimental error. The hybrid variation was partitioned into male and female parent main effects giving two independent estimates of GCA effects, while the male and female interaction estimates the SCA effects (Kearsey and Pooni, 1996). The GCA values for the parents and SCA effects for crosses and their standard errors were also estimated (Kearsey and Pooni, 1996).

To estimate heritability, the analogous broad-sense and narrow-sense coefficients of genetic determination were estimated as follows:

Broad-sense heritability;

$$H^2 = \frac{\delta^2\mathrm{GCA_f} + \delta^2\mathrm{GCA_m} + \delta^2\,\mathrm{SCA_{fm}}}{\delta^2\mathrm{GCA_f} + \delta^2\mathrm{GCA_m} + \delta^2\mathrm{SCA_{fm}} + \delta^2\mathrm{e}}$$

Narrow-sense coefficient genetic determination (heritability);

$$\mathrm{NS-CGD} \approx \mathrm{h}^2 = \frac{\delta^2\mathrm{GCA_f} + \delta^2\mathrm{GCA_m}}{\delta^2\mathrm{GCA_f} + \delta^2\mathrm{GCA_m} + \delta^2\mathrm{SCA_{fm}} + \delta^2\mathrm{e}}$$

Baker's ratio was determined as follows:

$$\text{Baker's ratio} = \frac{\delta^2 \text{GCA}_\text{f} + \delta^{2\,2} \text{GCA}_\text{m}}{\delta^2 \text{GCA}_\text{f} + \delta^2 \text{GCA}_\text{m} + \delta^2 \text{SCA}_\text{fm}}$$

Where

H^2 -Broad sense heritability

$\delta^2 \text{GCA}_\text{f}$ -Variance of general combining ability females

$\delta^2 \text{GCA}_\text{m}$ - Variance of general combining ability males

SCA_fm - Variance of specific combining ability females and males

$\delta^2 \text{e}$ - Error variance.

h^2 -Narrow sense heritability

7.5. Results

Analysis of variance

Location-wise analysis of variance was carried out on sugar and biomass related characters individually to find out the performance of genotypes at each of the three environments. Environment-wise analysis of variance for Kibos (Table 7.2) revealed significant differences ($P<0.05$) due to genotypes for brix %, significant difference ($P<0.001$) due to plant height at 90 days, significant difference ($P<0.01$) due grain weight of six panicles and significant difference ($P<0.01$) due to 100 seed weight.

Environment-wise analysis of variance for Homa Bay (Table 7.2) revealed significant differences($P<0.05$) due to genotypes for brix % at 90 days, significant difference ($P<0.001$) due to plant height at 90 days, significant difference($P<0.05$) due to grain weight of six panicles and significant difference($P<0.001$) due to 100 seed weight.

Location-wise analysis of variance for Alupe (Table 7.2) showed the significant differences ($P<0.05$) due to genotypes for plant height at 90 days.

Across location analysis of variance (Table 7.2) revealed significant difference ($P<0.001$) due to genotypes for plant height at 90 days, significant difference ($P<0.001$) due to brix % at 90 days, significant difference ($P<0.001$) due to grain weight of six panicles and significant difference ($P<0.001$) due to 100 seed grain weight.

Table 7.2: Analysis of variance for sugar and biomass related traits

Location	Stratum	d.f	Brix 75 DAP	Brix 90 DAP	Girth 60DAP	Girth 90DAP	Plt ht 60DAP	Plt ht 90DAP	pncl dmt	Pncl ht harv	Fld wt/ 6 pncl	Grn wt/6 pncl	100 SW
Kibos	REP1	1	10.07ns	1.24ns	0.49ns	0.02ns	211.30ns	359.20ns	0.47ns	78.15*	50138.00*	997.00ns	0.44*
	REP1.Block	4.99	1.72*	0.61ns					2.83***	7.27**		9000.00ns	
	Variety	35	1.15ns	0.80*	0.59ns	0.02ns	1420.00**	3231.70***	1.07ns	15.82***	25015.00*	15196.53**	0.34***
	Residual	29.01	0.64	0.32	0.71	0.01	501	618.7	0.51	2.16	11538	5406	0.08
	LEE	16.7	0.81	0.37					0.86	2.97		6054.4	
Homa Bay	REP1	1	8.36ns	1.50*	0.01ns	0.02ns	118.10ns	446.00ns	0.62ns	89.11*	48309.00ns	1891.00ns	0.49**
	REP1.Block	5	1.43ns	0.56ns				419.40ns	2.95***	9.09**	12164.00ns	8913.00ns	
	Varieties	35	1.12ns	0.82*	0.06ns	0.02ns	1402.10**	3738.24***	1.05ns	15.87***	25066.37*	14542.11*	0.34***
	Residual	30	0.74	0.32	0.04	0.01	533	730.4	0.5	2.24	11205	5386	0.08
	LEE	17.6	0.84	0.35	0.05	0.02	955.6	685.92	0.84	3.2	11342.25	5887.49	0.21
Alupe	REP1	1	0.03ns	0.64ns	0.04ns	0.02ns	0.10ns	16958.70***	0.62nsns	7.87ns	168.00ns	1624.00ns	0.01ns
	REP1.Block	4.99		1.38*	0.03ns								
	Varieties	35	0.24ns	0.65ns	0.03ns	0.02ns	282.00ns	1672.70*	0.30ns	25.66ns	5269.00ns	4006.00ns	0.13ns
	Residual	29.01	0.19	0.46	0.03	0.02	291.2	799.9	0.43	29.2	4036	2641	0.22
	LEE	16.7		0.59	0.03								
Across	Location	2	91.08*	0.19ns	0.33ns	0.04ns	92905.50***	46038.40ns	62.24**	29.30ns	1899695.00**	909358.00***	8.24*
	Location.REP1	3	6.07***	1.09ns	0.18ns	0.02ns	109.80ns	5921.30***	0.57ns	61.34**	32872.00*	1768.00ns	0.31ns
	Varieties	35	1.68***	1.70***	0.28ns	0.03**	2138.90***	6062.80***	1.26***	33.62***	37793.00***	23381.00***	0.58***
	Location. Varieties	70	0.35ns	0.35ns	0.20ns	0.01ns	482.60ns	998.30ns	0.40ns	10.85ns	8719.00ns	4090.00ns	0.11ns
	Residual	104	0.61	0.44	0.26	0.01	441.2	702.3	0.71	11.86	8947	4810	0.13
	Total	214	1.62	0.62	0.24	0.02	1591.9	2172.7	1.27	15.94	31596	16023	0.28

DAP=Days after planting, Fld wt/6 pncl=Field weight of six panicle, Pncl dmt=Panicle diameter, Pncl ht harv =Panicle height at harvest, 100 SW =100 seed weight, ns=not significant * Significant at P≤0.05 **Significant at P≤0.01***Significant at P≤0.001, LEE=Lattice effective error

Combining ability effects

The estimates of GCA effects of parents for sugar and biomass traits at Alupe are presented in Table 7.3. The magnitude of GCA estimates varied among parents. Female parent MALON had positive and significant (P<0.01) GCA effects for height at 90 days and male parents IESV92008 DL and IESV92038/2-SH had high and positive and significant effects (P<0.05) for grain weight of six panicles.

The estimates of SCA effects of crosses for sugar and biomass traits at Alupe are presented in Table 7.4. Positive and significant SCA (P<0.05) estimate for brix % was obtained from GADAMxIESV93036. SCA effect for MALONxIS2331 was significant (P<0.01) for height at 90 days. KS5989-29005xIESV930461 showed significant (P<0.05) SCA effect for six panicles grain weight.

The estimates of GCA effects of parents for sugar and biomass traits in Homa Bay are presented in Table 7.5. The magnitude of GCA estimates varied among parents. Male parent IESV 93046 and female parent KS5989-29005 were significant (P<0.05) for plant height at 90 days. The GCA effects due to female parent PAISANO was significant (P<0.05) for girth at 90 days.

The estimates of SCA effects of crosses for sugar and biomass traits in Homa Bay are presented in Table 7.6. SCA effects for GADAMxNTJ 2 was significant (P<0.05) and NK8416-19075xIESV92038/2-SH (P<0.01) for height at 90 days. SCA effect was significant (P<0.05) for grain weight of panicles for PAISANOxIESV92038/2-2H.

The estimates of GCA effects of parents for sugar and biomass traits in Kibos are presented in Table 7.7. GCA effect due female parent KS5989-29005 was significant (P < 0.05) for height at 90 days. Male parents IS 2331 and NTJ 2 had positive and significant (P < 0.05) for 100 seed weight.

The estimates of SCA effects of crosses for sugar and biomass traits in Kibos are presented in Table 7.8. GADAMxNTJ2 and NK8416-19075xIESV92038/2-SH had high and positive significant ($P \leq 0.05$) SCA effects for height at 90 days. PAISANOxIS2331 had significant SCA effect ($P \leq 0.01$) for panicle height.

Table 7.3**:** General combining ability (GCA) effects for sugar and biomass related traits in Alupe

	Brix 75 DAP	Brix 90 DAP	Girth 60 DAP	Girth 90 DAP	Plt ht 60 DAP	Plt ht 90 DAP	Pncl dmt	Pncl ht harv	Field w/6 pan	Grn wt/6 panicle	100SW
GADAM	0.005ns	-0.136ns	0.077ns	0.043ns	-8.876ns	-25.620*	0.083ns	-0.918ns	2.132ns	1.252ns	0.031ns
KS5989-29005	0.143ns	-0.326ns	0.000ns	-0.018ns	-9.116ns	-6.820ns	-0.212ns	-0.020ns	-25.628ns	-13.968ns	-0.129ns
MALON	-0.125ns	0.190ns	-0.066ns	-0.033ns	8.064ns	33.700**	0.119ns	-0.560ns	30.112ns	21.892ns	0.118ns
NK8416-19075	-0.047ns	0.254ns	0.084ns	0.064ns	12.324*	3.760ns	-0.092ns	1.060ns	-22.328ns	-21.868ns	-0.020ns
PAISANO	0.025ns	0.020ns	-0.096ns	-0.056ns	-2.396ns	-5.020ns	0.101ns	0.440ns	15.712ns	12.692ns	-0.001ns
IESV92008DL	-0.115ns	0.150ns	-0.021ns	0.021ns	-1.536ns	-5.400ns	0.151ns	-0.380ns	40.812ns	38.592**	0.220ns
IESV92038/2-SH	-0.115ns	-0.180ns	0.057ns	-0.031ns	2.024ns	-1.620ns	0.221ns	-0.020ns	26.412ns	29.392*	0.117ns
IESV93046	0.305ns	0.244ns	-0.032ns	-0.033ns	-3.416ns	-11.680ns	-0.247ns	-1.058ns	-65.168**	-32.348*	-0.210ns
IS2331	-0.049ns	-0.372ns	-0.014ns	0.027ns	-1.556ns	18.880ns	-0.134ns	0.460ns	10.332ns	-18.728ns	-0.203ns
NTJ2	-0.025ns	0.160ns	0.010ns	0.016ns	4.484ns	-0.180ns	0.009ns	1.000ns	-12.388ns	-16.908ns	0.076ns

Table 7.4**:** Specific combining ability (SCA) effects for sugar and biomass related traits in Alupe

	Brix 75 DAP	Brix 90 DAP	Girth 60 DAP	Girth 90 DAP	Plt ht 60 DAP	Plt ht 90 DAP	Pncl dmt	Pncl ht harv	Field w/6 pan	Grn wt/6 panicle	100SW
GADAMxIESV92008DL	0.105ns	-0.844ns	-0.047ns	-0.138ns	1.616ns	4.300ns	-0.023ns	-0.542ns	-4.832ns	3.348ns	0.061ns
GADAMxIESV92038/2-SH	0.205ns	-0.514ns	0.041ns	0.009ns	-4.944ns	-19.980ns	0.507ns	0.398ns	61.068ns	43.548ns	-0.166ns
GADAMxIESV93046	0.285ns	1.032*	-0.029ns	0.012ns	-15.904ns	3.980ns	-0.014ns	3.346*	-55.752ns	-92.412**	-0.255ns
GADAMxIS2331	-0.161ns	0.328ns	0.033ns	0.125ns	11.636ns	-13.180ns	-0.188ns	-0.282ns	2.648ns	46.668ns	0.475ns
GADAMxNTJ2	-0.435ns	-0.004ns	0.001ns	-0.008ns	7.596ns	24.880ns	-0.281ns	-2.922ns	-3.132ns	-1.152ns	-0.115ns
KS5989-29005xIESV92008DL	0.017ns	-0.304ns	0.049ns	-0.007ns	12.456ns	4.000ns	-0.428ns	-0.140ns	-18.572ns	-17.932ns	0.281ns
KS5989-29005xIESV92038/2-SH	0.067ns	0.376ns	0.029ns	0.069ns	0.496ns	10.220ns	-0.498ns	-2.800ns	-45.672ns	-45.232ns	0.134ns
KS5989-29005xIESV93046	-0.203ns	0.102ns	0.218ns	0.176ns	10.736ns	5.280ns	0.369ns	0.138ns	74.408ns	79.508*	0.366ns
KS5989-29005xIS2331	0.241ns	0.088ns	-0.241*	-0.217*	-24.224*	-27.980ns	0.344ns	2.520ns	-19.792ns	-14.412ns	-0.670*
KS5989-29005xNTJ2	-0.123ns	-0.264ns	-0.056ns	-0.021ns	0.536ns	8.480ns	0.214ns	0.280ns	9.628ns	-1.932ns	-0.110ns
MALONxIESV92008DL	-0.065ns	-0.020ns	0.118ns	0.071ns	-10.624ns	-27.820ns	0.441ns	1.200ns	-10.812ns	-8.292ns	-0.161ns
MALONxIESV92038/2-SH	-0.315ns	0.110ns	-0.001ns	-0.017ns	7.916ns	-14.300ns	0.171ns	1.440ns	10.588ns	18.408ns	-0.003ns
MALONxIESV93046	-0.235ns	-0.264ns	-0.210ns	-0.061ns	-1.044ns	-11.340ns	-0.411ns	-0.922ns	-42.332ns	57.648ns	-0.166ns

MALONxIS2331	0.119ns	-0.248ns	0.072ns	-0.059ns	5.596ns	72.200**	0.026ns	-0.440ns	77.168ns	-55.972ns	0.032ns
MALONxNTJ2	0.495ns	0.420ns	0.020ns	0.066ns	-1.844ns	-18.740ns	-0.227ns	-1.280ns	-34.612ns	-11.792ns	0.298ns
NK8416-19075xIESV92008DL	0.157ns	0.466ns	-0.206ns	0.069ns	-3.584ns	-4.580ns	0.002ns	0.080ns	-19.372ns	-49.532ns	-0.073ns
NK8416-19075xIESV92038/2-SH	-0.093ns	-0.254ns	-0.042ns	-0.110ns	16.156ns	29.640ns	-0.018ns	1.820ns	-9.472ns	8.668ns	-0.105ns
NK8416-19075xIESV93046	0.487ns	-0.178ns	0.151ns	-0.035ns	12.096ns	5.700ns	-0.001ns	-1.642ns	19.608ns	-31.092ns	0.017ns
NK8416-19075xIS2331	-0.269ns	-0.192ns	0.125ns	0.172ns	-18.264ns	-31.960ns	0.224ns	0.240ns	-5.092ns	41.488ns	0.271ns
NK8416-19075xNTJ2	-0.283ns	0.156ns	-0.029ns	-0.097ns	-6.404ns	1.200ns	-0.206ns	-0.500ns	14.328ns	30.468ns	-0.109ns
PAISANOxIESV92008DL	-0.215ns	0.700ns	0.085ns	0.005ns	0.136ns	24.100ns	0.009ns	-0.600ns	53.588ns	72.408*	-0.107ns
PAISANOxIESV92038/2-SH	0.135ns	0.280ns	-0.029ns	0.049ns	-19.624ns	-5.580ns	-0.161ns	-0.860ns	-16.512ns	-25.392ns	0.141ns
PAISANOxIESV93046	-0.335ns	-0.694ns	-0.130ns	-0.092ns	-5.884ns	-3.620ns	0.057ns	-0.922ns	4.068ns	-13.652ns	0.038ns
PAISANOxIS2331	0.069ns	0.022ns	0.009ns	-0.021ns	25.256*	0.920ns	-0.406ns	-2.040ns	-54.932ns	-17.772ns	-0.109ns
PAISANOxNTJ2	0.345ns	-0.310ns	0.064ns	0.060ns	0.116ns	-15.820ns	0.501ns	4.420ns	13.788ns	-15.592ns	0.037ns

Table 7.5: General combining ability (GCA) effects for sugar and biomass related traits in Homa Bay

	Brix 75 DAP	Brix 90 DAP	Girth 60 DAP	Girth 90 DAP	Plt ht 60 DAP	Plt ht 90 DAP	Pncl dmt	Pncl ht harv	Field w/6 pan	Grn wt/6 panicle	100SW
GADAM	-0.064ns	-0.266ns	-0.002ns	-0.059ns	-26.496**	-38.764***	0.591ns	-0.864ns	53.012ns	39.140ns	-0.035ns
KS5989-29005	-0.256ns	0.356ns	0.082ns	-0.008ns	17.444ns	19.196*	0.056ns	0.316*	-38.088ns	-37.260ns	0.124ns
MALON	0.740*	0.102ns	-0.022ns	0.008ns	11.604ns	12.516ns	-0.177ns	-2.100**	-34.668ns	-26.560ns	0.110ns
NK8416-19075	0.108ns	0.176ns	0.016ns	-0.014ns	1.184ns	10.176ns	-0.484ns	3.596***	-15.788ns	17.340ns	-0.139*
PAISANO	-0.530ns	-0.368ns	-0.074ns	0.073*	-3.736ns	-3.124ns	0.013ns	-0.950ns	35.532ns	7.340ns	-0.060ns
IESV92008DL	-0.150ns	0.012ns	0.037ns	0.050ns	-13.036ns	-22.124*	0.043ns	-0.700ns	27.132ns	8.340ns	-0.132ns
IESV92038/2-SH	-0.290ns	-0.178ns	0.075ns	0.013ns	-5.456ns	-6.204ns	0.153ns	0.230ns	53.432ns	51.240*	0.002ns
IESV93046	0.400ns	-0.136ns	-0.010ns	0.002ns	4.684ns	18.676*	-0.169ns	-1.424*	-23.788ns	-37.660ns	-0.208**
IS2331	-0.062ns	-0.010ns	-0.078ns	-0.059ns	8.064ns	9.116ns	-0.140ns	0.612ns	-48.708ns	-23.160ns	0.189**
NTJ2	0.100ns	0.312ns	-0.023ns	-0.007ns	5.744ns	0.536ns	0.113ns	1.280*	-8.068ns	1.240ns	0.148*

DAP=Days after planting, Fld wt/6 pncl=Field weight of six panicle, Pncl dmt=Panicle diameter, Pncl ht harv =Panicle height at harvest, 100 SW =100 seed weight, ns=not significant * Significant at P≤0.05 **Significant at P≤0.01***Significant at P≤0.001

Table 7.6: Specific combining ability (SCA) effects for sugar and biomass related traits in Homa Bay

	Brix 75 DAP	Brix 90 DAP	Girth 60 DAP	Girth 90 DAP	Plt ht 60 DAP	Plt ht 90 DAP	Pncl ht harv	Pncl dmt	Field w/6 pan	Grn wt/6 panicle	100SW
GADAMxIESV92008DL	-0.076ns	-0.144ns	-0.230ns	-0.089ns	-3.804ns	-3.856ns	1.284ns	0.249ns	5.488ns	14.160ns	-0.320*
GADAMxIESV92038/2-SH	0.064ns	-0.454ns	-0.081ns	0.027ns	-1.584ns	-13.076ns	1.054ns	-0.811ns	-218.812**	-141.240*	0.156ns
GADAMxIESV93046	0.354ns	0.264ns	-0.061ns	-0.105ns	-8.324ns	-24.256ns	-0.012ns	0.199ns	87.808ns	53.160ns	-0.003ns
GADAMxIS2331	-0.014ns	0.528ns	0.217ns	0.150*	-0.704ns	-2.496ns	0.272ns	0.283ns	145.828ns	74.160ns	0.138ns
GADAMxNTJ2	-0.326ns	-0.194ns	0.155ns	0.018ns	14.416ns	43.684*	-2.596ns	0.079ns	-20.312ns	-0.240ns	0.030ns
KS5989-29005xIESV92008DL	0.366ns	0.184ns	-0.031ns	0.049ns	-17.944ns	16.884ns	2.604ns	0.134ns	-9.912ns	-38.940ns	0.371*
KS5989-29005xIESV92038/2-SH	-0.294ns	0.374ns	-0.090ns	-0.028ns	11.376ns	-3.936ns	-0.226ns	0.024ns	24.288ns	-7.840ns	-0.278ns
KS5989-29005xIESV93046	0.566ns	-0.268ns	0.010ns	0.101ns	1.236ns	9.084ns	0.828ns	0.746ns	81.508ns	58.060ns	0.152ns
KS5989-29005xIS2331	0.048ns	0.126ns	0.042ns	-0.178*	22.256ns	-0.156ns	-2.328ns	-0.867ns	-147.172ns	-72.940ns	-0.132ns
KS5989-29005xNTJ2	-0.684ns	-0.416ns	0.069ns	0.056ns	-16.924ns	-21.876ns	-0.876ns	-0.036ns	51.288ns	61.660ns	-0.114ns
MALONxIESV92008DL	-0.480ns	-0.512ns	-0.013ns	0.038ns	28.796ns	26.264ns	1.320ns	0.467ns	7.168ns	52.360ns	0.275ns
MALONxIESV92038/2-SH	0.010ns	0.378ns	0.078ns	-0.024ns	-26.784ns	-18.756ns	1.190ns	0.257ns	45.868ns	-9.040ns	-0.144ns
MALONxIESV93046	-0.480ns	-0.414ns	0.270ns	0.143ns	-8.824ns	20.764ns	-0.556ns	-0.521ns	-42.912ns	-29.140ns	-0.314*
MALONxIS2331	0.532ns	-0.340ns	-0.121ns	-0.012ns	8.196ns	0.324ns	-0.692ns	-0.050ns	-81.492ns	-53.140ns	0.234ns
MALONxNTJ2	0.420ns	0.888ns	-0.214ns	-0.145ns	-1.384ns	-28.596ns	-1.260ns	-0.153ns	71.368ns	38.960ns	-0.050ns
NK8416-19075xIESV92008DL	0.402ns	0.064ns	-0.047ns	-0.031ns	-13.884ns	-40.096ns	-3.176ns	-1.226ns	-1.212ns	-25.540ns	-0.331*
NK8416-19075xIESV92038/2-SH	0.092ns	-0.646ns	-0.019ns	-0.052ns	29.736ns	57.984**	-1.406ns	-0.236ns	-34.012ns	42.560ns	-0.235ns
NK8416-19075xIESV93046	-0.478ns	0.062ns	-0.058ns	-0.100ns	-8.304ns	-32.996ns	0.348ns	0.186ns	-57.292ns	-41.540ns	0.295ns
NK8416-19075xIS2331	-0.216ns	0.306ns	-0.028ns	0.107ns	-16.884ns	-5.136ns	0.192ns	0.873ns	-8.472ns	2.460nsns	0.081ns
NK8416-19075xNTJ2	0.202ns	0.214ns	0.151ns	0.076ns	9.336ns	20.244ns	4.044**	0.404ns	100.988ns	22.060ns	0.189ns
PAISANOxIESV92008DL	-0.210ns	0.408ns	0.321*	0.033ns	6.836ns	0.804ns	-2.030ns	0.377ns	-1.532ns	-2.040ns	0.005ns
PAISANOxIESV92038/2-SH	0.130ns	0.348ns	0.112ns	0.077ns	-12.744ns	-22.216ns	-0.610ns	0.767ns	182.668*	115.560*	0.501**
PAISANOxIESV93046	0.040ns	0.356ns	-0.161ns	-0.039ns	24.216ns	27.404ns	-0.606ns	-0.611ns	-69.112ns	-40.540ns	-0.129ns
PAISANOxIS2331	-0.348ns	-0.620ns	-0.111ns	-0.067ns	-12.864ns	7.464ns	2.558ns	-0.240ns	91.308ns	49.460ns	-0.321*
PAISANOxNTJ2	0.390ns	-0.492ns	-0.161ns	-0.004ns	-5.444ns	-13.456ns	0.690ns	-0.293ns	-203.332**	-122.440*	-0.055ns

DAP=Days after planting, Fld wt/6 pncl=Field weight of six panicle, Pncl dmt=Panicle diameter, Pncl ht harv =Panicle height at harvest, 100 SW =100 seed weight, ns=not significant * Significant at P≤0.05 **Significant at P≤0.01***Significant at P≤0.001

Table 7.7: General combining ability (GCA) effects for sugar and biomass related traits in Kibos

	Brix 75 DAP	Brix 90 DAP	Girth 60 DAP	Girth 90 DAP	Plt ht 60 DAP	Plt ht 90 DAP	Pncl dmt	Pncl ht harv	Grn wt/6 panicle	Field w/6 pan	100SW
GADAM	-0.108ns	-0.286ns	0.465ns	-0.058ns	-28.360***	-40.772***	0.572ns	-0.9452ns	36.968ns	51.288ns	-0.036ns
KS5989-29005	-0.312ns	0.336ns	-0.054ns	-0.007ns	16.740*	17.908*	0.039ns	0.2368ns	-38.952ns	-39.232ns	0.123ns
MALON	0.678*	0.084ns	-0.152ns	0.008ns	10.280ns	10.848ns	-0.193ns	-2.1732***	-28.712ns	-35.672ns	0.109ns
NK8416-19075	0.048ns	0.156ns	-0.084ns	-0.014ns	0.480ns	8.888ns	-0.501ns	3.5168***	15.648ns	-16.932ns	-0.140ns
PAISANO	-0.306ns	-0.290ns	-0.175ns	0.070ns	0.860ns	3.128ns	0.084ns	-0.6352ns	15.048ns	40.548ns	-0.056ns
IESV92008DL	-0.212ns	-0.006ns	-0.093ns	0.051ns	-14.360ns	-23.792**	0.027ns	-0.7732ns	6.188ns	26.128ns	-0.133ns
IESV92038/2-SH	-0.352ns	-0.196ns	0.572ns	0.014ns	-6.780ns	-7.872ns	0.137ns	0.1568ns	49.088ns	52.428ns	0.001ns
IESV93046	0.352ns	-0.156ns	-0.170ns	0.004ns	2.820ns	16.668ns	-0.188ns	-1.5052*	-39.832ns	-25.512ns	-0.209**
IS2331	0.174ns	0.064ns	-0.155ns	-0.062ns	13.900ns	16.128ns	-0.072ns	0.9148ns	-14.532ns	-43.972ns	0.193*
NTJ2	0.038ns	0.294ns	-0.153ns	-0.006ns	4.420ns	-1.132ns	0.097ns	1.2068*	-0.912ns	-9.072ns	0.147*

DAP=Days after planting, Fld wt/6 pncl=Field weight of six panicle, Pncl dmt=Panicle diameter, Pncl ht harv =Panicle height at harvest, 100 SW =100 seed weight, ns=not significant * Significant at P≤0.05 **Significant at P≤0.01***Significant at P≤0.001

Table 7.8: Specific combining ability (SCA) effects for sugar and biomass related traits in Kibos

	Brix 75 DAP	Brix 90 DAP	Girth 60 DAP	Girth 90 DAP	Plt ht 60 DAP	Plt ht 90 DAP	Pncl ht harv	Pncl dmt	Field w/6 pan	Grn wt/6 panicle	100SW
GADAMxIESV92008DL	-0.032ns	-0.124ns	-0.697ns	-0.091ns	-1.940ns	-1.848ns	1.3652ns	0.268ns	7.212ns	16.332ns	-0.319*
GADAMxIESV92038/2-SH	0.108ns	-0.434ns	1.961*	0.026ns	0.280ns	-11.068ns	1.1352ns	-0.792ns	217.088**	-139.068*	0.157ns
GADAMxIESV93046	0.474ns	0.276ns	-0.648ns	-0.105ns	-8.620ns	-23.608ns	0.0372ns	0.210ns	86.652ns	55.252ns	-0.002ns
GADAMxIS2331	-0.268ns	0.456ns	-0.303ns	0.153ns	-6.000ns	-9.168ns	-0.0228ns	0.216ns	141.812ns	65.552ns	0.134ns
GADAMxNTJ2	-0.282ns	-0.174ns	-0.312ns	0.017ns	16.280ns	45.692*	-2.5148*	0.098ns	-18.588ns	1.932ns	0.031ns
KS5989-29005xIESV92008DL	0.422ns	0.204ns	0.105ns	0.048ns	-17.240ns	18.172ns	2.6832*	0.151ns	-8.768ns	-37.248ns	0.372*
KS5989-29005xIESV92038/2-SH	-0.238ns	0.394ns	-0.581ns	-0.029ns	12.080ns	-2.648ns	-0.1468ns	0.041ns	25.432ns	-6.148ns	-0.277ns
KS5989-29005xIESV93046	0.608ns	-0.246ns	0.177ns	0.100ns	2.480ns	10.712ns	0.9152ns	0.765ns	83.372ns	59.772ns	0.153ns
KS5989-29005xIS2331	-0.164ns	0.044ns	0.095ns	-0.174*	18.900ns	-5.648ns	-2.6548*	-0.939ns	-152.468ns	-79.728ns	-0.135ns
KS5989-29005xNTJ2	-0.628ns	-0.396ns	0.205ns	0.055ns	-16.220ns	-20.588ns	-0.7968ns	-0.019ns	52.432ns	63.352ns	-0.113ns
MALONxIESV92008DL	-0.418ns	-0.494ns	0.117ns	0.037ns	30.120ns	27.932ns	1.3932ns	0.483ns	8.172ns	54.512ns	0.276ns
MALONxIESV92038/2-SH	0.072ns	0.396ns	-0.419ns	-0.024ns	-25.460ns	-17.088ns	1.2632ns	0.273ns	46.872ns	-6.888ns	-0.143ns

MALONxIESV93046	-0.432ns	-0.394ns	0.430ns	0.142ns	-6.960ns	22.772ns	-0.4748ns	-0.502ns	-41.188ns	-26.968ns	-0.313ns
MALONxIS2331	0.296ns	-0.414ns	-0.044ns	-0.008ns	2.360ns	-6.688ns	-0.9948ns	-0.118ns	-86.228ns	-61.768ns	0.230ns
MALONxNTJ2	0.482ns	0.906ns	-0.083ns	-0.146ns	-0.060ns	-26.928ns	-1.1868ns	-0.137ns	72.372ns	41.112ns	-0.049ns
NK8416-19075xIESV92008DL	0.462ns	0.084ns	0.053ns	-0.032ns	-13.180ns	-38.808*	-3.0968*	-1.209ns	-0.068ns	-23.848ns	-0.330*
NK8416-19075xIESV92038/2-SH	0.152ns	-0.626ns	-0.546ns	-0.052ns	30.440ns	59.272**	-1.3268ns	-0.219ns	-32.868ns	44.252ns	-0.234ns
NK8416-19075xIESV93046	-0.452ns	0.084ns	0.072ns	-0.101ns	-7.060ns	-31.368ns	0.4352ns	0.205ns	-55.428ns	-39.828ns	0.296ns
NK8416-19075xIS2331	-0.424ns	0.224ns	0.169ns	0.111ns	-20.240ns	-10.628ns	-0.1348ns	0.801ns	-13.768ns	-4.328ns	0.078s
NK8416-19075xNTJ2	0.262ns	0.234ns	0.251ns	0.075ns	10.040ns	21.532ns	4.1232**	0.421ns	102.132ns	23.752ns	0.190ns
PAISANOxIESV92008DL	-0.434ns	0.330ns	0.422ns	0.037ns	2.240ns	-5.448ns	-2.3448ns	0.306ns	-6.548ns	-9.748ns	0.001ns
PAISANOxIESV92038/2-SH	-0.094ns	0.270ns	-0.414ns	0.080ns	-17.340ns	-28.468ns	-0.9248ns	0.696ns	177.652*	107.852ns	0.497**
PAISANOxIESV93046	-0.198ns	0.280ns	-0.030ns	-0.036ns	20.160ns	21.492ns	-0.9128ns	-0.679ns	-73.408ns	-48.228ns	-0.133ns
PAISANOxIS2331	0.560ns	-0.310ns	0.084ns	-0.081ns	4.980ns	32.132ns	3.8072**	0.040ns	110.652ns	80.272ns	-0.306ns
PAISANOxNTJ2	0.166ns	-0.570ns	-0.061ns	-0.001ns	-10.040ns	-19.708ns	0.3752ns	-0.364ns	-208.348*	-130.148*	-0.059ns

DAP=Days after planting, Fld wt/6 pncl=Field weight of six panicle, Pncl dmt=Panicle diameter, Pncl ht harv =Panicle height at harvest, 100 SW =100 seed weight, ns=not significant * Significant at P≤0.05 **Significant at P≤0.01***Significant at P≤0.001

The estimates of GCA effects of parents for sugar and biomass traits across locations are presented in Table 7.9. Male parents NTJ 2 had significant GCA effect ($P<0.01$) for brix % at 90 days. Female parent MALON had significant GCA effect ($P<0.01$) for plant height at 90 days.

The estimates of SCA effects of crosses for sugar and biomass traits across locations are presented in Table 7.10. The following crosses had significant ($P\leq0.01$) SCA effects for brix at 90 days; GADAMxIS2331 and GADAMxIESV92008 DL. NK8416-19075xIESV93046 had significant SCA effect ($P\leq0.001$) for plant height at 90 days. Cross KS5989-29005xIS2331 had a significant SCA effect ($P<0.001$) for girth at 90 days. PAISANOxIESV 93046 had significant and positive SCA effect ($P<0.001$) for 100 seed weight.

Table 7.9: General combining ability (GCA) affects for sugar and biomass related traits across locations

Parents	Brix 75 DAP	Brix 90 DAP	Girth 60 DAP	Girth 90 DAP	Plt ht 60 DAP	Plt ht 90 DAP	Pncl dmt	Pncl ht harv	Field w/6 pan	Grn wt/6 panicle	100SW
GADAM	-0.116ns	-0.207ns	0.178ns	-0.024ns	-21.296**	-35.020***	0.384**	-0.898ns	35.716*	24.564ns	-0.013ns
KS5989-29005	-0.110ns	0.115ns	0.010ns	-0.010ns	8.384ns	10.040ns	-0.055ns	0.104ns	-35.324*	-31.096*	0.039ns
MALON	0.436*	0.127ns	-0.080ns	-0.007ns	9.944ns	19.080**	-0.053ns	-1.556**	-12.304ns	-8.816ns	0.113**
NK8416-19075	0.066ns	0.187ns	0.006ns	0.013ns	4.784ns	7.600ns	-0.374**	2.646***	-19.384ns	2.484ns	-0.100*
PAISANO	-0.276ns	-0.223ns	-0.114ns	0.028ns	-1.816ns	-1.700ns	0.098ns	-0.294ns	31.296*	12.864ns	-0.039ns
IESV92008DL	0.058ns	-0.129ns	-0.082ns	-0.027ns	6.984*	14.700*	-0.175***	0.498**	-30.564**	-24.256**	0.059ns
IESV92038/2-SH	-0.150**	0.051ns	-0.025ns	0.039*	-9.656**	-17.060*	0.104***	-0.568***	32.356**	19.964*	-0.014ns
IESV93046	-0.244***	-0.185*	0.232*	-0.003ns	-3.416ns	-5.220ns	0.200***	0.176ns	44.996***	45.384***	0.040ns
IS2331	0.292***	0.007ns	-0.070ns	-0.009ns	1.224ns	7.820ns	-0.231***	-1.316***	-37.904**	-37.736***	-0.209*
NTJ2	0.044ns	0.255**	-0.054ns	0.000ns	4.864ns	-0.240ns	0.103**	1.212***	-8.884ns	-3.356ns	0.124ns

DAP=Days after planting, Fld wt/6 pncl=Field weight of six panicle, Grn wt/6 panicle=Grain weight six panicle, Pncl dmt=Panicle diameter, Pncl ht harv =Panicle height at harvest, 100 SW =100 seed weight, ns=not significant * Significant at P≤0.05 **Significant at P≤0.01***Significant at P≤0.001

Table 7.10: Specific combining ability (SCA) effects for sugar and biomass related traits across locations

Crosses	Brix 75 DAP	Brix 90 DAP	Girth 60 DAP	Girth 90 DAP	Plt ht 60 DAP	Plt ht 90 DAP	Pncl dmt	Pncl ht harv	Field w/6 pan	Grn wt/6 panicle	100SW
GADAMxIESV92008DL	-0.114ns	0.435**	-0.016ns	0.137***	1.536ns	-8.240ns	0.222ns	0.188ns	100.324*	70.836**	0.249*
GADAMxIESV92038/2-SH	0.054ns	-0.395**	-0.322ns	-0.107**	-1.224ns	-0.480ns	0.194ns	0.684ns	2.204ns	12.416ns	-0.192ns
GADAMxIESV93046	0.188ns	-0.489**	0.631**	0.020ns	-1.964ns	-14.720ns	-0.332ns	0.850ns	-124.636**	-77.304**	0.048ns
GADAMxIS2331	0.162ns	0.599***	-0.243ns	-0.058ns	-11.204ns	-14.560ns	-0.078ns	0.962ns	36.464ns	-7.284ns	-0.087ns
GADAMxNTJ2	-0.290ns	-0.149ns	-0.050ns	0.009ns	12.856ns	38.000**	-0.005ns	-2.686**	-14.356ns	1.336ns	-0.019ns
KS5989-29005xIESV92008DL	0.100ns	0.083ns	-0.036ns	-0.185ns	5.856ns	-11.300ns	-0.609**	-1.144ns	-110.936**	-63.304*	-0.314**
KS5989-29005xIESV92038/2-SH	0.238ns	0.033ns	0.041ns	0.029ns	-7.604ns	12.960ns	-0.033ns	1.782ns	-11.356ns	-30.224ns	0.341**
KS5989-29005xIESV93046	-0.188ns	0.389**	-0.212ns	0.003ns	7.856ns	1.220ns	-0.130ns	-0.992ns	2.204ns	-18.744ns	-0.139ns
KS5989-29005xIS2331	0.356*	-0.153ns	0.135ns	0.123***	4.816ns	8.380ns	0.702**	0.740ns	81.404ns	70.276**	0.225*
KS5989-29005xNTJ2	-0.506**	-0.351*	0.072ns	0.029ns	-10.924ns	-11.260ns	0.069ns	-0.388ns	38.684ns	41.996ns	-0.112ns
MALONxIESV92008DL	0.274ns	-0.309ns	-0.031ns	-0.030ns	5.196ns	22.160*	0.012ns	-0.544ns	-26.556ns	-51.484ns	0.165ns
MALONxIESV92038/2-SH	-0.328*	-0.339*	0.073ns	0.050ns	16.036*	8.620ns	0.434ns	1.252ns	0.524ns	30.496ns	0.129ns
MALONxIESV93046	-0.084ns	0.297ns	-0.112ns	-0.020ns	-14.704ns	-16.720ns	0.203ns	1.248ns	33.384ns	-1.324ns	-0.096ns
MALONxIS2331	-0.320*	-0.385*	0.161ns	0.073*	-5.444ns	10.640ns	-0.448ns	-0.660ns	-42.516ns	1.896ns	-0.264**
MALONxNTJ2	0.458**	0.737ns	-0.092ns	-0.073*	-1.084ns	-24.700*	-0.202ns	-1.298ns	35.164ns	20.416ns	0.067ns
NK8416-19075xIESV92008DL	-0.246ns	0.111ns	0.089ns	0.135***	-18.144*	-15.960ns	0.507*	-0.236ns	-13.876ns	5.516ns	0.144ns
NK8416-19075xIESV92038/2-SH	0.312*	0.211ns	-0.067ns	0.002ns	-10.304ns	-27.700*	-0.792**	-1.980*	-5.896ns	-31.904ns	-0.244*
NK8416-19075xIESV93046	0.016ns	-0.503**	-0.200ns	-0.072*	25.356**	48.860***	-0.141ns	-0.224ns	-24.336ns	32.876ns	-0.191ns
NK8416-19075xIS2331	-0.110ns	-0.025ns	0.055ns	-0.081*	-1.084ns	-19.480ns	0.206ns	-0.182ns	-29.236ns	-33.004ns	0.202ns
NK8416-19075xNTJ2	0.028ns	0.207ns	0.123ns	0.017ns	4.176ns	14.280ns	0.221ns	2.6208**	73.344ns	26.516ns	0.089ns
PAISANOxIESV92008DL	-0.014ns	-0.319*	-0.007ns	-0.056ns	5.556ns	13.340ns	-0.133ns	1.734ns	51.044ns	38.436ns	-0.245*
PAISANOxIESV92038/2-SH	-0.276ns	0.491**	0.275ns	0.026ns	3.096ns	6.600ns	0.197ns	-1.740ns	14.524ns	19.216ns	-0.034ns
PAISANOxIESV93046	0.068ns	0.307ns	-0.108ns	0.069ns	-16.544*	-18.640ns	0.400ns	-0.884ns	113.384**	64.496*	0.379***
PAISANOxIS2331	-0.088ns	-0.035ns	-0.108ns	-0.057ns	12.916ns	15.020ns	-0.382ns	-0.862ns	-46.116ns	-31.884ns	-0.075ns
PAISANOxNTJ2	0.310*	-0.443**	-0.053ns	0.019ns	-5.024ns	-16.320ns	-0.083ns	1.750ns	-132.836**	-90.264**	-0.026ns

DAP=Days after planting, Fld wt/6 pncl=Field weight of six panicle, Pncl dmt=Panicle diameter, Pncl ht harv =Panicle height at harvest, 100 SW =100 seed weight, ns=not significant * Significant at P≤0.05 **Significant at P≤0.01***Significant at P≤0.001

Genetic components effects and measures of gene action

Heritability estimates and measures of gene action at Kibos (Table 7.11) indicates that low coefficient of Baker's ratio (0.281) was recorded for brix % at 90 days.

Plant height at 90 days for general combining ability males (GCAm), general combining ability females (GCAf) and SCA had significant effects ($P<0.05$). The amount of Baker's ratio for plant height at 90 days, 100 seed weight and panicle height at harvest was > 0.5. High narrow sense heritability (0.56) was registered for plant height at 90 days.

Heritability estimates and Bakers ratio in Homa Bay (Table 7.11) indicated that SCA effect of brix % at 90 days was significant ($P<0.05$). Low coefficient of Baker's ratio (0.060) was observed for brix % at 90 days.

The amount of Baker's ratio for plant height in Homa Bay at 90 days was closer to 1 (more than 0.7). High narrow sense heritability (0.56) was noted for plant height at 90 days.

Heritability estimates at Alupe (Table 7.11) showed low coefficient of Baker's ratio (0.29) for brix % at 90 days. The amount of Baker's ratio for plant girth at 90 days and grain weight for six panicles were 1.

Heritability estimates and Bakers ratio for combined location (Table 7.12) indicated that GCAm and SCA effects for brix % at 90 days were significant ($P<0.05$). Low coefficient of narrow sense heritability (0.224) was recorded for brix % at 90 days.

The SCA effect for plant height at 90 days was highly significant ($P<0.001$). The amount of Baker's ratio for plant height at 90 days was closer to 1.

Table 7.11: Variance components, Heritability estimates and Bakers ratio for sugar and biomass traits within locations

Stratum	d.f.	Brix 75DAP	Brix 90DAP	Girth 60 DAP	Girth 90DAP	Plt ht 60 DAP	Plt ht 90 DAP	Pncl dmt	pncl ht harv	Fld wt/6 pncl	Grn wt/6 pncl	100SW
Kibos												
REP1	1	8.44ns	0.68ns	0.74ns	0.02ns	20.90ns	757.70ns	2.34ns	47.25ns	91124ns	13513.00ns	0.14ns
REP1.Block	3.56	1.62*	0.68ns		0.02ns			2.34ns	6.30*	13624ns	10872.00ns	
Female	4	2.63*	0.54ns	0.84ns	0.03ns	2344.10*	4601.40**	1.32ns	31.39***	12585.61ns	10318.58ns	0.13ns
Male	4	1.05ns	0.59ns	1.00ns	0.01ns	992.20ns	2713.90*	0.44ns	16.05**	15897.61ns	16233.69ns	0.30**
Female. Male	16	0.44ns	0.55ns	0.77ns	0.02ns	633.20ns	1795.70*	0.89ns	12.33**	30470.42*	13801.92ns	0.16**
Residual	16.44	0.5	0.35	1.18	0.01	656.1	800.8	0.68	2.02	11055	4649	0.05
LEE	16.83	0.82	0.46		0.01			1.13	3.24	13248.01	6572.35	
BR		1	0.281	0	0.369	1	0.697	0.271	0.614	0.052	0.369	0.558
NSH		0.501	0.107	0	0.23	0.552	0.56	0.054	0.547	0.031	0.254	0.46
BSH		0.501	0.38	0	0.624	0.552	0.804	0.201	0.892	0.607	0.688	0.825
Homa Bay												
REP1	1	0.68ns	99843.00ns	0.163ns	16867.00ns	0.03ns	982.60ns	2.40ns	53.11ns	0.02ns	730.00ns	6.67ns
REP1.Block	3.5	0.62ns	13823.00ns		10521.00ns			2.40*	7.89*	0.02ns	730.00ns	1.45ns
Female	4	0.76ns	12890.47ns	0.135*	9743.00ns	0.03ns	4610.90**	1.18ns	30.99***	0.03ns	2738.00*	2.79*
Male	4	0.54ns	14731.97ns	0.267**	15552.00ns	0.03ns	2552.60*	0.48ns	16.86**	0.01ns	884.00ns	0.70ns
Female. Male	16	0.57ns	29709.47*	0.165**	12866.00ns	0.06ns	1679.90ns	0.86ns	11.98	0.02ns	759.00ns	0.41ns
Residual	17.5	0.35	10473	0.048	4630	0.05	905	0.66	2.16	0.01	700	0.7
LEE	17.1	0.44	12276.64		6245.74			1.08	3.59	0.01	782	0.92
BR		0.433	0.06	0.511	0.383	0	0.734	0.176	0.624	0.35	0.933	1
NSH		0.205	0.037	0.426	0.261	0	0.56	0.033	0.548	0.222	0.495	0.448
BSH		0.472	0.625	0.833	0.681	0.176	0.763	0.185	0.879	0.635	0.53	0.448

Alupe												
REP1	1	0.05ns	0.21ns	13491.00***	0.28ns	0.85ns	3.76ns	0.11*	3110.00ns	0.03ns	14.00ns	77.30ns
REP1.Block	3.35	0.32ns	0.73ns							0.02ns		
Female	4	0.07ns	0.67ns	4402.00**	0.04ns	0.24ns	6.56ns	0.06ns	3596.00ns	0.02ns	5881.00ns	818.20*
Male	4	0.23ns	0.43ns	1884.00ns	0.28ns	0.40ns	7.40ns	0.01ns	8571.00**	0.02ns	14060.00*	84.10ns
Female. Male	16	0.21ns	0.55ns	1301.00ns	0.12ns	0.25ns	7.77ns	0.03ns	4329.00*	0.02ns	3727.00ns	327.80ns
Residual	17.65	0.2	0.33	1058	0.21	0.45	4.71	0.03	2128	0.01	4718	211.1
LEE	17.65	0.23	0.43	1776	0.17	0.37	6.15	0.03	3603	0.02	5184	290.5
BR		0	0.297	0.873	1	0	0.372	0.739	0.59	0.15	1	0.675
NSH		0	0.126	0.562	0.116	0	0.19	0.318	0.422	0.063	0.471	0.426
BSH		0.071	0.426	0.644	0.116	0	0.509	0.569	0.716	0.419	0.471	0.892

BR=Bakers ratio, NSH=Narrow sense heritability, BSH=Broad sense heritability, LLE=Lattice effective error, ns=not significant, DAP=Days after planting, Fld wt/6 pncl=Field weight of six panicle, Pncl dmt=Panicle diameter, Pncl ht harv =Panicle height at harvest, 100 SW =100 seed weight,* Significant at P≤0.05 **Significant at P≤0.01***Significant at P≤0.001

Table 7.12: Variance components, Heritability estimates and Bakers ratio for sugar and biomass traits across locations

Change	d.f.	Brix 75 DAP	Brix 90DAP	Fld wt/6 pncl	Girth 60DAP	Girth 90DAP	Gr wt/ 6 pncl	Height 60DAP	Height 90DAP	Pncl dmt	Pncl ht harv	100 SW
Location	2	55.82*	0.02ns	1443853.00**	0.46ns	0.04ns	646047.00**	73550.40***	36480.60ns	46.03**	59.09ns	4.36*
Location. Replicate	3	5.57***	0.78ns	45723.00**	0.29ns	0.03ns	7421.00ns	33.00ns	5077.00**	0.60ns	62.09***	0.19ns
Female	4	2.42ns	1.29ns	18998.00*	0.45ns	0.01ns	10234.00ns	4095.10*	11703.50**	2.41*	70.88**	0.23*
Male	4	1.15***	0.89*	37097.00***	0.50ns	0.01ns	29639.00***	1363.80*	4582.20ns	0.94***	28.76***	0.44ns
Female. Male	16	0.55ns	1.13**	32122.00***	0.31ns	0.04***	16273.00***	913.80*	3229.30***	0.98ns	14.80***	0.31***
Location. Female	8	0.96ns	0.43ns	4595.00ns	0.24ns	0.02*	3780.00ns	724.50ns	977.00ns	0.50ns	8.37*	0.04ns
Location. Male	8	0.07ns	0.15ns	2316.00ns	0.27ns	0.01ns	1998.00ns	240.60ns	1263.60ns	0.02ns	0.38ns	0.19ns
Location.Female.Male	32	0.15ns	0.15ns	12079.00ns	0.28ns	0.01ns	4547.00ns	351.00ns	773.50ns	0.35ns	5.54ns	0.07ns
Residual	62	0.6	0.4	9043	0.41	0.01	4552	521.9	923.3	0.83	3.6	0.1
Total	139	1.45	0.46	34285	0.35	0.02	15886	1690.4	2193.7	1.36	10.15	0.2
BR		0.719	0.396	0.495	0.826	0.061	0.538	0.762	0.696	0.643	0.797	0.418
NSH		0.280	0.224	0.259	0.121	0.026	0.350	0.431	0.496	0.266	0.535	0.209
BSH		0.389	0.566	0.523	0.146	0.418	0.650	0.566	0.713	0.414	0.671	0.501

BR= Baker's ratio, NSH=Narrow sense heritability, BSH=Broad sense heritability, DAP=Days after planting, ns=not significant, Fld wt/6 pncl=Field weight of six panicle, Pncl dmt=Panicle diameter, Pncl ht harv =Panicle height at harvest, 100 SW =100 seed weight,* Significant at P≤0.05 **Significant at P≤0.01***Significant at P≤0.001

7.6. Discussion

The significant mean values obtained from the analysis of variance for the individual location and across locations (Table 7.2) suggests that differences existed between the sorghum genotypes for most traits, indicating that they are highly variable This finding is in agreement with the findings of Bello D. *et al*. (2007), Basu (1981), and Abu-Gasim and Kambal (1985) for several quantitative traits in sorghum genotypes. Zaveri *et al*. (1989) also reported similar results in pearl millet. Bello D. *et al*. (2007).

The GCA effects of parents on sugar and biomass traits in this study are given and described in Tables 7.3, 7.5, 7.7 and 7.9. The significant GCA effects due to male parents IESV92008 DL and IESV92038/2-SH (Table 7.3) for Alupe for grain weight of six panicles implied that genes with additive effects were important for this traits and breeding progress could be achieved through selection of good parents. The GCA effects due to male parent IESV 93046 and female parent KS5989-29005 for Homa Bay (Table 7.5) were significant ($P<0.05$) for plant height at 90 days and for girth at 90 days; thus are good combiners for increasing tallness. Parents with significant negative GCA effects were good combiners in decreasing plant height. These results are consistent with earlier reports (Tadesse et al., 2008; Kenga et al., 2004). This implied that genes with additive effects were important for these traits and breeding progress could be achieved through selection of good parents.

GCA effects across locations (Table 7.9) due to male parents NTJ 2 for brix % at 90 days and female parent MALON plant height at 90 days were significant ($P<0.01$).The desirable GCA effect associated with brix for NTJ 2 indicates that it combines for increased brix and that genes with additive effects were important for this trait. The positive and significant GCA effects for

male parents IESV 93046 and IESV 92038/2-SH for grain weight of six panicles (Table 7.9) suggests that their use in sweet sorghum hybrid production could result in improved grain weight.

The specific combining ability of parents and traits in this study are given and described in Tables 7.4, 7.6, 7.8 and 7.10. SCA effects across locations (Table 7.10) for GADAMxIS2331 were significant ($P<0.001$) for brix % at 90 days. SCA effects for NK8416-19075x1ESV93046 were significant ($P<0.01$) for height at 90 days. For these traits, further gains can be achieved through hybridization capitalizing on non-additive gene effects.

The significant SCA and GCA effects and their significant interaction with site for plant height indicate potential for exploiting non-additive gene action for improving stem biomass. Schlehuber (1945) reported similar results where F1 hybrids were higher than that of either parent in stem biomass.

SCA effects across locations were also significant ($P\leq0.001$) for 100 seed weight and grain weight for panicles for PAISANOxIESV 93046 (Table 7.10).The use of this hybrid could result in improved seed weight.

Plant height at 90 days for Kibos and Homa Bay (Table 7.11) GCAm, GCAf and SCA significant effects indicate that both additive and non additive effects were important under environments of evaluation. This suggests that parental selection and combination for plant height performance can exploit both non additive and additive gene affects.

The amount of Baker's ratio for plant height at 90 days and panicle height at harvest (Table 7.12) was closer to 1 indicating additive effects play more significant roles. Low coefficient of Baker's ratio for the traits brix % at 90 days indicates the role of non-additive effects in controlling it.

High narrow sense heritability for plant height at 90 days indicates that the alleles that control plant height can be easily transmitted to the offspring through recombination.

Bakers ration was large in most traits (≥ 0.50) (Table 7.13) indicating predominance of additive effects. The closer the ratio is to unity, the greater the predictability of progeny performance based on GCA effects alone.

7.7. Conclusions

The study identified parental lines with positive GCA effects for improvement of plant height, brix and grain weight as IESV 93046, IS 2331 and NTJ 2 respectively.

The best SCA effects for improvement of brix were crosses GADAMxIESV93046 and GADAMxIS2331, for improvement of plant height were MALONxIS2331; MALONxNTJ 2 and for improvement of grain weight was PAISANOxIESV 93046.

The study confirmed earlier reports that genes with both additive and non-additive effects were important in controlling stem brix and biomass associated traits in sweet sorghum.

In general, the study also demonstrated high potential of hybrids over pure line cultivars, suggesting that productivity of sweet sorghum in western Kenya could be boosted by promoting hybrid cultivars.

7.8. References

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CHAPTER EIGHT

8.0 GENERAL DISCUSSION, CONCLUSIONS AND RECOMMENDATIONS

8.1. General discussion

Farmers' subjective assessments of agricultural technologies influence adoption behavior (Nowak, 1992). Often farmers are not included in setting up research agenda leading to negative consequences (Derera *et al*., 2006). Despite the fact that research centers have reported significant yield increases in many crops and in development of other technologies, farmers remain unaware and have low perception of the technologies to take full advantage of them (Ekpere, 1995). Breeding objectives in countries where traditional cropping systems are dominating have not been appropriately oriented towards the perceptions of farmers, specifically their needs and preferences for the difficult growing conditions of their regions (Witcombe et al., 2006; Mekbib, 2006).

In first result chapter (Chapter 4) aimed at scoping farmer's perception and the potential for sweet sorghum production as an alternative crop. The study revealed that majority of farmers (60%) are willing to venture in the farming of sweet sorghum with the aim of selling the stalks only. They were also willing to take part in the development of the sweet sorghum and it products by allowing small mills in their farms (80%), promoting and marketing sweet sorghum (90%) and willing to be contracted for sweet sorghum production (85%). The study concluded that farmers appreciate the potential of sweet sorghum and existence of capacity for its exploitation. Taken together these results show that there is scope for introduction of sweet sorghum as an alternative crop for the supply of the sugar and jaggary industry. Indeed, forward-thinking biofuel companies interested in sweet sorghum are working directly with seed companies to better understand and identify potential project applications, specifically with respect to selection of optimal hybrids, and to develop optimal processing, cultivation and supply chain parameters. By

working together, energy crop developers and industry participants can capitalize on a near-term opportunity to expand production; more fully utilize existing sugarcane assets and lower production costs.

As bio-fuels are produced from sweet sorghum, they offer enormous opportunities to improve the income levels of smallholder farmers in Kenya and other developing countries, which are predominantly agrarian economies. At community level, farmers will cultivate sweet sorghum and fetch more income while meeting their food needs. Local production of bio-fuels is projected to have a broad range of positive economic, social and environmental implications. At a national level, producing more bio-fuels will generate new technologies, new industries, new jobs and new markets assisting economic growth in rural areas besides reducing environmental pollution.

Sweet sorghum gene pool creation has not received much attention mainly because it is not considered to be among important crops in Kenya, and the pedigree information is incomplete. Currently there are no criteria (morphological traits or molecular markers) to differentiate sweet sorghums from grain sorghums (Murray *et al*., 2009), and most of the accessions lack the proper information to help distinguish between sweet and grain sorghum. Therefore when requesting sweet sorghum germplasm, one is limited to a few characters that are common in sweet sorghum like tall plants that are leafy (high biomass), and where available the brix degree, which also is subjective as there is no definite value for distinguishing grain sorghums from sweet ones (Murray *et al*., 2009).

The second result chapter (Chapter 5) is about assembling and molecular description of assembled gerpmplasm to be exploited in breeding efforts to meet farmer and niche needs identified in objective 1 and 2. The 11 SSR markers that were used to assess genetic diversity in sweet sorghum generated 86 alleles with an average of 8 alleles per locus. Polymorphism information

content (PIC) value was 0.53 indicating a moderate diversity with a range of 0.09–0.89. The variability among the populations was low as 3 % but amounted to 22% and 75 % within individual genotypes and among individuals respectively. Clustering analysis based on the genetic similarity (GS) grouped the 86 sweet sorghum genotypes into 2 distinct clusters. The study also revealed the genetic relationship of cultivars with unknown parentage to those with known parentage. Information generated from this study can be exploited to select parents for hybrid development to maximize sugar content and total biomass and for development of segregating populations to map genes controlling sugar content in sweet sorghum.

The constraints for large scale sweet sorghum cultivation are the limited availability of genotypes suited to different agro-climatic conditions with all built-in resistances for biotic and abiotic stresses, photoperiod sensitivity and non –availability of quantity of feedstock suited to off-season crushing in sugar industries(Ortiz *et al*., 2006).

Demand for renewable energy sources and biofuel, which would minimize pollution has risen sharply (Belum *et al*., 2007). However biofuel crops like sweet sorghum have not been extensively put under cultivation to bridge this demand.

The third result chapter (chapter 6) is about stability of selected sweet sorghum genotypes for western Kenya ecologies. Among the test genotypes, maximum juice yield was recorded by IESV 93046 (1550 ml) in Alupe. Mean purity percent was maximum in Homa Bay (58.6) and the least was observed in Kibos (21). Among individual genotypic means for purity IESV 93046 exhibited the highest value of 73.6 % in Homa Bay. IESV 93046 and IS 2331 were the tallest varieties across the four locations registering mean height of 269 and 252 cm respectively. Taken together these results show that Genotypes IESV 93046 and IS 2331 are the highest yielding and most stable in terms of brix and biomass. These varieties can be exploited for ethanol production.

The study indicates that genotype by location interaction and other high level interactions had significant influence on brix, plant height, and juice volume and stalk weight. Due to significant genotype by environment interaction on brix, plant height, juice volume and stalk weight, simultaneous selection for these traits is not possible across the four environments and that selection for each location have to be carried out separately in individual environment.

Sweet sorghum has received relatively little attention and application of technology, such as advanced breeding, compared to maize, soy and cotton, for instance. Commercial sweet sorghum seed varieties consist mostly of old, open-pollinated lines, or sweet varieties that were crossed with Sudan grass for use as forage. Such varieties do not provide economic yields of fermentable sugars under many circumstances. General and specific combining ability of sweet sorghum lines for brix has been scarcely reported in literature (Makanda, 2009).

The fourth result chapter (Chapter 7) is about breeding efforts towards development of sweet sorghum hybrids. From the results of this study, the GCA effects due to male parents IESV92008 DL and IESV92038/2-SH in Alupe were significant ($P<0.05$) for grain weight of six panicles. This implied that genes with additive effects were important for this traits and breeding progress could be achieved through selection of good parents. SCA effects for GADAMxIESV93036 were significant ($P<0.05$) for brix % at 90 days. SCA effects for MALONxIS2331 were significant ($P<0.01$) for height at 90 days. Bakers ration was large in most traits (>0.50) indicating predominance of additive effects. For these traits, further gains can be achieved through hybridization capitalizing on non-additive gene effects. Overall this study demonstrates potential of development of sweet sorghum hybrids.

8.2. General conclusions

Farmers were aware of sweet sorghum and accompanying technologies however their perception was constrained by some socio-cultural factors.

Farmers appreciate the potential of sweet sorghum and existence of capacity for its exploitation and are likely to adopt it.

Pairs of genotypes, which can be exploited to select parents for hybrid development and for development of segregating populations to map genes controlling sugar content in sweet sorghum, were identified.

Sweet sorghum cultivars from Argentina, Brazil, Kenya, United States of America and Zambia, are very closely related.

The study identified parental lines with positive GCA effects for improvement of plant height, brix and grain weight as IESV 93046, IS 2331 and NTJ 2 respectively.

The best SCA effects for improvement of brix were crosses GADAMxIESV93046 and GADAMxIS2331, for improvement of plant height were MALONxIS2331; MALONxNTJ 2 and for improvement of grain weight was PAISANOxIESV 93046.

The study confirmed earlier reports that genes with both additive and non-additive effects were important in controlling stem brix and biomass associated traits in sweet sorghum.

In general, the study also demonstrated high potential of hybrids over pure line cultivars, suggesting that productivity of sweet sorghum in western Kenya could be boosted by promoting hybrid cultivars.

8.3. Recommendations and future prospectives

Since the varieties respondents grow are mainly local varieties, a well coordinated sweet sorghum seed supply system should be promoted.

The small land holding units (<2 acres) that majority of the farmers (73.0%) have are not feasible for economic purposes and farmers should be encouraged to consolidate their lands to maximize on the economies of scale

Research institutes and extension agents should embark on enlightenment campaign on the importance of sweet sorghum as a multipurpose crop suitable for food, feed, fiber and fuel.

Due close genetic relationships among sweet sorghum cultivars from Argentina, Brazil, Kenya, United States of America and Zambia, they can only be improved by accessing additional new genes. It may therefore be necessary that the future breeding strategies are directed at crossing sorghum with some of its close relatives.

Since there was G x E influence for most of the characters evaluated, it is important to conduct multi-location testing to select superior materials in sweet sorghum.

The best hybrids for total sugar content and biomass were GADAMxIESV93036, GADAMxIS2331 and MALONxIS2331 and after adequate testing across many locations in the target production environments these hybrids can be recommended for commercial exploitation for ethanol production.

More focus should be on identifying diverse parental lines and use them in crossing programme to develop mapping population to identify QTL's.

Appendix 1: List of primers used for diversity assessment

Marker	Primer sequences (5′–3′)
Xcup53	F: GCAGGAGTATAGGCAGAGGC R: CGACATGACAAGCTCAAACG
Xcup63	F: GTAAAGGGCAAGGCAACAAG R: GCCCTACAAAATCTGCAAGC
mSbCIR329	F: GATCTTCACCAGGAACAGG R: ATGAGAGGAAAACATTGCTG
mSbCIR246	F: TTTTGTTGCACTTTTGAGC R: GATGATAGCGACCACAAATC
Xtxp021	F: GAGCTGCCATAGATTTGGTCG R: ACCTCGTCCCACCTTTGTTG
Xcup14	F: TACATCACAGCAGGGACAGG R: CTGGAAAGCCGAGCAGTATG
Xcup02	F: GACGCAGCTTTGCTCCTATC R: GTCCAACCAACCCACGTATC
Xtxp273	F: GTACCCATTTAAATTGTTTGCAGTAG R: CAGAGGAGGAGGAAGAGAAGG
Xtxp12	F: AGATCTGGCGGCAACG R: AGTCACCCATCGATCATC
Xtxp145	F: GTTCCTCCTGCCATTACT R: CTTCCGCACATCCAC
mSbCIR283	F: TCCCTTCTGAGCTTGTAAAT R: CAAGTCACTACCAAATGCAC

Appendix 2: Mean monthly temperature and rainfall data for Kibos Station No. 9034105 during the trial data. Source: Kenya Sugar Research Foundation AgroMet (2012, 2013 and 2014)