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RESEARCH ARTICLE

GENETIC VARIABILITY AMONG SORGHUM (*Sorghum bicolor* L. Moench) ACCESSIONS

¹Akinyele, M. O., ²*Oyewale, R. O., ³Idowu, G. A., ²Ibrahim, H. M. and ⁴Afolabi, S. G.

¹National Biotechnology Development Agency, Abuja, Nigeria

²Department of Crop Production, Federal University of Technology, Minna, Niger state, Nigeria

³National Agricultural Seed Council, Abuja, Nigeria

⁴Department of Soil Science and Land Management, Federal University of Technology, Minna, Niger State, Nigeria

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ABSTRACT

Sorghum (*Sorghum bicolor* L. Moench) is an important staple food crops and provide bulk of raw materials for the livestock and many agro-allied industries in the world. There are collections of sorghum genotypes in some research institutes and most of these collections lack information on its morphology- agronomic traits that could be used by researchers to improve sorghum production in Nigeria. However, breeding for high yield crops requires information on the nature and magnitude of variation in the available materials and the relationship of yield with other agronomic characters. Grain yield in sorghum is quantitative in nature and polygenically controlled, effective yield improvement and simultaneous improvement in yield components are imperative. Selection on the basis of grain yield character alone is usually not very effective and efficient. Genetic variability studies provide the basic information regarding the genetic properties of the population based on which the breeding methods are formulated for further crop improvement. For any progress in plant breeding, there is the need to study the genetic variability which cannot be easily quantified. Genetic improvement for quantitative traits depends on the nature and amount of variability present in any genetic stock and the extent to which the desirable traits are heritable. Knowledge of association between yield and its component traits and among the component parameters themselves can improve the efficiency of selection in plant breeding. Therefore, there is the need to characterize as much as possible sorghum genotypes available in Nigeria to identify traits for yield against future sorghum improvements for better food production and security.

*Corresponding author: Oyewale, R. O.

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INTRODUCTION

Sorghum (*Sorghum bicolor* (L.) Moench) is the fifth most important cereal crop worldwide after wheat (*Triticum aestivum*) rice (*Oryza sativa*), maize (*Zea mays*) and barley (*Hordeum vulgare*) (Tesfamichael et al., 2015). It forms the most important dry land cereal crop for the semi-arid and tropics together with maize and pearl millet (*Pennisetum glaucum* (L.). It is grown in at least 86 countries in an area of 38 million hectares with annual grain production of about 5.8 million tonnes (FAOSTAT, 2014). It is the most widely cultivated crop in the northern guinea savanna zones of Nigeria (Maikasuwa and Ala, 2013). Sorghum in Nigerian savanna zones is grown on an estimated area of 4.5 million hectares with annual production output of about 6 million tons (Sani et al., 2013). USAID, (2011) reported that, 2015/2016 Sorghum production estimate was 6.2 million tons, a minimal decrease of two percent from the 2014/2015 figure of 6.3 million tonnes. Nigeria among the West African sub region is the highest sorghum producer accounting for 71% of the regional total output in the world (Sani et al., 2013). Sorghum belongs to the Poaceae family and the tribe of Andropogoneae

(Tiny et al., 2014). It has been argued that the Andropogoneae tribe has a centre of variability in Katanga (Congo) region of tropical Africa (Esperance, 2009). As a member of the grass family, sorghum represents a robust, cane-like species. It has the ability to tiller, regrow following harvest, and produce a fibrous, deep root system. Sorghum leaf blades, similar in appearance to those of its close relative Maize may be up to a meter in length with a width of 10–15 cm (Kimber et al., 2013). The panicles and grains of the *Sorghum* species can vary widely in shape and size and represent a means for racial classification. Sorghum panicles are made up of perfect flowers, and it is considered a self-pollinated species; however, out crossing can be as high as 70 % in certain races in particular environments (Kimber et al., 2013). Cultivated sorghum had been classified into five races (Bicolor, Guinea, Caudatum, Durra and Kafir) and ten intermediate races corresponding to the pair wise combinations of major races. They are identified according to morphological traits, especially panicle, grain and glumes traits (Esperance, 2009). Sorghum race identification is more complicated and the taxonomy has been a subject of many modifications from centuries. Until now sorghum classification has been complex

for most of the new scientists. However, sorghum identification can rather be easily carried out in the field for familiar research or in the laboratory from head or even spike less specimens (Esperance, 2009). The principal colours of sorghum grains are white, brown, red, orange and yellow with a range of intermediate colours and in addition to cultivated sorghum, there are numbers of wild weed sorghum species (Esperance, 2009). Around the world, Sorghum is grown for the production of dense grain panicles; for food, and energy (Kimber *et al.*, 2013). Sorghum in Africa is processed into a wide variety of attractive and nutritious traditional foods, such as semi-leavened bread, couscous, dumplings and fermented and non-fermented porridges. It is the grain of choice for brewing traditional African beers (Taylor, 2003). Sorghum straw (stem fibers) can also be made into excellent wallboard for house building, as well as biodegradable packaging. It is also used in packaging materials for sensitive electronic equipment. A more recent use of sorghum is for ethanol. By-products from ethanol production, such as sorghum-DDGS (distillers dried grains with soluble), are also finding a place in the market (US Grain Council, 2006). In Nigeria, sorghum is mainly used in the form of flour or paste processed into two main dishes *Tuwo* a thick porridge and *Ogi* or *Akamu*, *Kunu* a thin diet or porridge. Other dishes that are sometimes made from sorghum include; a number of deep fried snacks, steamed dumplings and other boiled or roasted snack foods (Ratnavathi and Patil, 2013).

Origin and History of Sorghum: Sorghum, (*Sorghum bicolor* (L.) Moench) is a cultivated tropical cereal grass. It is generally believed, not universally considered to have first been domesticated in North Africa, possibly in the Nile or Ethiopian regions as recently as 1000 BC (Taylor, 2003). The largest diversity of cultivated and wild sorghums in Africa (Kimber *et al.*, 2013). Numerous varieties of sorghum were created through the practice of disruptive selection, selection for more than one level of a particular character within a population occurs, this result from a balance of farmer selection for cultivated traits and natural selection for wild traits, generating both improved sorghum types, wild types and intermediate types (Olembo *et al.*, 2010). These improved sorghum types were spread via the movement of people and trade routes into other regions of Africa, India (approx. 1500–1000 BC), the Middle East (approx. 900–700 BC) and eventually into the Far East (approx. AD 400). By the time sorghum was transported to America during late 1800s to early 1900s, diversity of new sorghum types, varieties and races were created through the movement of people, disruptive selection, geographic isolation and recombination of these types in different environments (Dillon *et al.*, 2007). There seems to be no argument that sorghum plants are African in origin, with the earliest known record of sorghum found in 8000 BP charred remains of sorghum at the Nabta Playa in Southern Egypt. Mary, (2016) confirmed that all of the races belong to the same biological species and are fully fertile when hybridized. For the cereal sorghums, they identify four wild races and five cultivated races (Harlan and Stemler 1976).

Botanical Description of Sorghum

Sorghum is a member of the grass family, poaceae, a group of flowering plants that also includes such important agricultural grains as wheat, rice, maize and sugar cane. Grasses typically have hollow stem (called culms) that are plugged at intervals (the nodes) with leaves arising at the nodes. There are small,

wind pollinated flowers arranged into spikelets, the fruit is a caryopsis with the seed and fruit merged together to form a hard dry grain. The genus sorghum is one of about 600 genera in poaceae, this genus is in the sub family panicoideae and the tribe Andropogoneae. It is a close relative of sugar cane (*Saccharum officinarum*), which belongs to the same tribe. There are about 30 species in the sorghum genus, some of which are raised for grain and many of which used as fodder plants either cultivated or as part of pasture. Species are native to tropical and subtropical region of all continents in addition to the South west pacific and Australasia. Sorghum in nature is a short-day plant and photoperiod sensitive. Elite germplasm, however, has been bred to be photoperiod insensitive with multiple maturity classes based on manipulation of various known maturity genes (Kimber *et al.*, 2013). Similar to maize in early stages, and with maize like leaves and tall stalks, sorghum differs considerably from maize in later stages. The inflorescence and grain (fruit/seed) is a panicle and sorghum is characterized by spikelets borne in pairs (FAO, 1995). Sorghum has more side shoots than maize, putting out new shoots and producing several head bearing culms from the nodes. Sorghum also has a more extensively branched root system. A waxy coating on the leaves and stems help to retain water even in intense heat. While sorghum is a perennial grass, it is treated as annual and can be harvested more than one times during the year (FAO, 1995).

Economic Importance of Sorghum: Sorghum grain is used for animal feed in USA and India (Obilana, 2005). Sorghum is one of the most important staple food crops in Nigeria and its production surpasses all other crops (Baiyegunhi and Fraser, 2009). The leaves and grains are also used for livestock feeds and the stalks for thatching houses and making fences as reported by National Agricultural Extension Research and Liaison Services (NAERLS, 1996). Sorghum is a very valuable industrial crop for brewing alcoholic and non-alcoholic drinks as well as in the baking and confectionery industries in Nigeria. According to Aduba, *et al.* (2013). Sorghum has greater untapped potentials than any other crop. It even postulated that if the twentieth century was the century of wheat, rice and maize, then the twenty-first century could become the century of sorghum. So of all the cereal crops, sorghum contributes about 50% of the calories in Nigeria generally and about 73% in the savanna regions of the country in particular (Aduba *et al.*, 2013). Sorghum has greater untapped potentials than any other crop, the potential for sorghum to be the driver of economic development in Africa especially Nigeria cannot be over emphasized (Sani *et al.*, 2013).

Genetic Variability Studies in Sorghum: Jain *et al.* (2012) revealed highly significant differences among the genotypes. High heritability accompanied with high GA as percent of mean was observed for days to 50% flowering, plant height, number of leaves per plant, leaf length and fodder yield per plant suggested that these characters are under additive gene action and gives better scope for selection. Jain and Patel (2012) evaluated 102 land races of forage sorghum for days to 50 % flowering, plant height, number of leaves per plant, leaf length and fodder yield per plant. High heritability accompanied with GA as percentage of means was observed in most of the characters studied. Khandelwal *et al.* (2015) studied 224 genotypes of sorghum for genetic parameters and characters association among yield components, the result revealed that phenotypic coefficients of variation (PCV) were

higher than genotypic coefficients of variation (GCV) for all the traits investigated. Jain *et al.* (2016) studied 32 sorghum genotypes for yield and yield component traits, reported existence of diversity in nine quantitative traits among the genotypes.

Correlation Coefficients Studies in Sorghum: Most of the quantitative traits of economic importance show correlated responses. As yield is dependent on several component characters, the knowledge of characters association is useful to plant breeder for formulating effective and efficient breeding programmes, correlation study provides a measure of association amongst characters. They may also be appreciated when highly heritable traits are positively and closely associated among themselves and with grain yield. Thus, breeding procedures based on improvement of one or more sub-traits would lead to tangible improvement in grain yield than straight selection in grain yield. Bahaa *et al.* (2013) conducted experiment on twenty sorghum landraces noticed that positive and highly significant correlation between grains yield per panicle and panicle length, panicle width, thousand seed weight. It was discovered that most of the traits with positive relationship also have high heritability Deepalakshmi and Ganesamurthy (2007) recorded high heritability accompanied with high GA as per cent of mean for the traits days to fifty per cent flowering, plant height, leaves per plant, leaf length, ear head weight, number of primaries per panicle, 100 grain weight, grain mould score and single plant yield, thus suggesting that these characters would be under influence of additive gene action, and could be improved through selection. Seed yield was positively and significantly correlated with days to maturity, number of leaves per plant, ear head weight and number of primaries per panicle, but there was negative significant correlation with grain mould score.

Path Analysis Studies in Sorghum

Path analysis is done with the aim of understanding the direct and indirect Contributions of different traits towards the grain yield. The direct contribution of each Component to the yield and the indirect effects and its association with other traits cannot be differentiated by simple correlations. Path coefficient analysis fulfils this gap. Deepalakshmi and Ganeshmurthy (2007) conducted experiment on sixteen sorghum genotypes, observed that plant height had negative direct effect on grain yield per plant. Vijaya *et al.* (2012) conducted experiment on 25 *rabis* orghum genotypes where panicle weight, 100-seed weight and plant height contributed high direct positive effect on grain yield. Information on genetic diversity has been successfully used for efficient germplasm management and utilisation (Frankel, 1989; Blakeney, 2002), genetic finger printing and genotype selection (Bucheyeki *et al.*, 2009) in crop improvement. Morphological descriptors, biochemical and molecular markers are the methods used for measuring genetic diversity among crop species (Mehmood *et al.*, 2008). Morphological traits provide a simple way of measuring genetic diversity while studying genotype performance under normal growing conditions, but are influenced by environmental factors (Fufa *et al.*, 2005). Geleta and Labuschagne (2005) underlined the importance of using molecular markers as an additional tool for varietal description, as the genetic control of morphological traits is mostly polygenic and their expression depends on environmental factors. Molecular markers are invaluable for understanding the genetic make-up of agricultural crops. They

differ from morphological traits in that they usually occur in greater numbers, they can be distinguished without relying on the complete development of the plant and their expression is not altered by the environment (Abu Assar 2009). Knowledge of genetic similarity/dissimilarity not only generates a better understanding of germplasm sampling, but also has implications with regard to choice of parents for crosses and gene introgression from distantly related germplasm. Genetic diversity studies using a combination of techniques, such as morphological and molecular markers have been conducted in cotton (*Gossypiumhirsutum* L.) (Bie *et al.*, 2001; Wu *et al.*, 2001), wheat (*Triticumaestivum* L.) (Cox and Murphy, 1990; Vieira *et al.*, 2007) and oilseed crops (Riaz *et al.*, 2003). Morphological, SSR and AFLP markers have also been compared to assess genetic diversity in maize (*Zea mays* L.) (Beyene *et al.*, 2005) and have generated useful information about genetic diversity. Both morphological and molecular markers analyses are informative tools for estimating genetic distances (Vieira *et al.*, 2007). Thus, this study aimed to compare the use of both morphological and AFLP markers to assess biodiversity in sorghum accessions.

Molecular Marker Technologies: Conventional plant breeding is time consuming and highly dependent on environmental conditions. The application of molecular markers is crucial and more effective and efficient for selection of traits of interest in breeding programs as well as to assess genetic diversity, since these markers are not influenced by environment (Shehzad *et al.*, 2009). Molecular markers play a major role in the determination of genetic variability and relationships. Different kinds of markers have been used in many studies of sorghum (Shehzad *et al.*, 2009) for example restriction fragment length polymorphism (RFLPs) (Ahnert *et al.*, 1996), random amplified polymorphic DNA (RAPDs) (Iqbal *et al.*, 2010), simple sequence repeats (SSRs) (Bucheyeki *et al.*, 2009) and amplified fragment length polymorphism (AFLPs) (Ritter *et al.*, 2007). These molecular markers have been used efficiently to characterize and estimate the genetic diversity among sorghum germplasm. Genetic erosion resulted in loss of biodiversity in crop plants which has emphasized the importance of characterizing germplasm accessions, including local landraces and other plant materials for genetic resource conservation (Todorovska *et al.*, 2005). Hence, consideration must be given to evaluate crop genetic resources such as landraces as well as breeding material at DNA level for characterization, evaluation, maintenance and improvement of the crop (Todorovska *et al.*, 2005). Among the marker platforms, SSR markers are widely used and are advantageous as markers for diversity studies due to their high level of polymorphism, wide availability and ease of detection (Schloss *et al.*, 2002). SSR markers are highly polymorphic (Uptmooret *et al.*, 2003) even among closely related cultivars, which demonstrate that they are highly informative. These markers are co-dominant and can be analyzed by a rapid, technically simple, specific and inexpensive polymerase chain reaction (PCR) based assay that requires only small amounts of DNA. Furthermore, they are abundant (Varshney *et al.*, 2005) and uniformly dispersed in the plant genome. The study can indicate which landraces are suitable for rescue, agronomic evaluation and for short- and long-term breeding strategies in the country. DNA markers have been applied to assess and characterise genetic diversity within and among crop species and these will help in identifying important sources of genetic variability (Menz *et al.*, 2004; Todorovska *et al.*, 2005; Kumar *et al.*, 2008). Genetic erosion resulted in loss of biodiversity

which has emphasised the importance of characterising germplasm accessions, including local landraces and other plant materials for genetic resource conservation (Food and Agriculture Organization (FAO), 1998; Todorovska *et al.*, 2005). Hence, consideration must be given to evaluate genetic resources such as landraces as well as breeding material at DNA level for characterization, evaluation, maintenance and improvement of genetic diversity (Todorovska *et al.*, 2005). Conservation of genetic diversity within and among a species is very important to achieve genetic gain towards targeted goals and food security (Gray, 1996). Molecular marker technologies contribute towards studying genetic diversity. It has numerous advantages, including the availability of large numbers of markers that cover the entire genome and their expression are not affected by the environment (Gepts, 1993). Genetic diversity in sorghum has been estimated using several types of molecular markers such as, RFLP (Deu *et al.*, 2006), RAPD (Uptmoor *et al.*, 2003), AFLP (2003); Menz *et al.*, 2004) and SSR (Brown *et al.*, 1996; Menz *et al.*, 2004; Casa *et al.*, 2005). In each of these studies, the authors studied a precise subset of sorghum germplasm. Microsatellites or SSR markers are particularly attractive for studying genetic differentiation because they are co-dominant (Akkaya *et al.*, 1992) and abundant in the genome (Lagercrantz *et al.*, 1993). However, there are some limitations, previous genetic information is needed, huge upfront work required, problems associated with PCR (Kubik *et al.*, 2001). A few studies have been devoted to assessing patterns of sorghum genetic variation at country level for example in Kenya (Nkongolo *et al.*, 2008), Sudan (Assar *et al.*, 2005), Ethiopia (Mekbib *et al.*, 2009), Malawi (Nkongolo *et al.*, 2008) and at regional levels in Western Africa (Ktavii *et al.*, 2014). The strength of the current study lies in the fact that no previous regional studies have been conducted in Eastern and Central Africa, even though it is suggested to be the center of origin of the crop. This information is important in understanding the domestication and evolution of sorghum. Therefore, this study examines the extent of genetic diversity among sorghum collections from Eastern and Central Africa is using SSR markers and to study the genetic population structure diversity distribution within and between countries.

Before the advent of GBS-based single nucleotide polymorphism (SNP) markers and additionally other whole-genome profiling markers (for example DaRT), microsatellite markers are the most used worldwide in the characterization of sorghum genetic resources and particularly in genetic diversity analysis (Smith *et al.*, 2000; Bhosale *et al.*, 2011). In Ethiopia, AFLP markers were used in genetic characterization of sorghum germplasms (Ayana, *et al.*, 2000), SR markers were used for sorghum germplasm analysis (Adugna, 2014). Microsatellite markers when compared with AFLP markers, they are specific, codominant and multi-allelic and well known to allow a good discrimination of closely related sorghum accessions (Ghebru *et al.*, 2002). A clear and better picture of diversity can be obtained from molecular markers but it is also warranted that integration of genetic and phenotypic analysis could be a more powerful tool in elucidating genetic diversity and relationships among sorghum landraces. Previous studies (e.g. Adugna, 2014) have documented the existence of large genetic variation in land race collection of sorghum. Folkertsma *et al.* (2005) reported that the guinea race of sorghum from various parts of the world were highly diverged from each other and that there is a predominance of rare alleles. Additional studies on sorghum land races from Burkina

Faso (Barro-Kondombo *et al.*, 2010) and Cameroon corroborated the occurrence of extensive genetic diversity even in relatively small region of sorghum cultivation and in germplasm involving landraces.

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