Sudan University of Science and Technology

College of Graduate Studies

Assessment of Variation Water Stress Tolerance, Genetic Diversity and Quality Triats in Grain Pearl Millet(*Pennisetum glaucum* L.) Genotypes

تقويم التباين للتحمل للاجهاد المائي والتباعد الوراثي وصفات الجودة لمحصول الدخن اللؤلوي الحبوب

By

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Dedication

This work is dedicated to the soul of my late parents Zainab Hadad and Mohamed Elsadig (ALLAH Mercy them).

To my son Mohamed and my lovely daughters Samah, Sara, Salooma and to my pitty brothers and sisters and to Engineer Abuobieda M alseid.

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الخلاصة

أجري هذا البحث بغرض دراسة التباين بين ثلاثين طرز وراثى من نبات الدخن اللؤلؤى الحبوب تحت ظروف الرى الطبيعى والاجهاد المائى والتباعد الوراثى باستخدام واسمات التصميم العشوائي وصفات الجودة. اجريت تجربتين حقليتين بجامعة السودان للعلوم والتكنولوجيا كلية الدراسات الزراعية شمبات لموسمين صيفيين متتاليين للاعوام 2012 و.2013 باستخدام تصميم القطاعات الكاملة العشوائية بثلاث مكررات مبنية على تنظيم القطع المنشقة. تم تجميع القراءات لكل من طول النبات وعدد الاوراق وقطر الساق ومساحة الورقة وعدد الايام ل50% ازهار ووزن القندول وعدد الحبوب فى القندول ووزن الف حبة وانتاجية الحبوب بالطن للهكتار. اجريت تجريبتين معمليتين لدراسة التباعد الوراثى باستخدام واسمات الحمض النووي عشوائية التضخيم المتعدد الاشكال وصفات الجودة والتى تشمل المكونات الكيميائية والمحتوى المعدنى والخصائص الفيزيائية لثلاثين طرز وراثى من الدخن باستخدام تصميم العشوائى الكامل. اظهرت النتائج وجود فروقات معنوية لمعظم الصفات المدروسة تحت ظروف الرى الطبيعى والاجهاد المائى للتحليلين الفردى والتجميعى فى الموسمين اظهر تحليل واسمات الحمض النووي عشوائية التضخيم المتعدد الاشكال وجود درجة عالية من التعدد الشكلي محددة وجود تباعد وراثى بين الطرز الوراثية للدخن. كما اظهرت النتائج جود تباين لكل صفات الجودة. سجلت اعلى انتاجية للحبوب بالطن هكتار الطرز الوراثى HSD10319 وكانت 2.51 و2.90 تحت ظروف الاجهاد و الرى الطبيعى علي التوالي . نتائج المكونات الكيميائية أظهرت أن أعلى قيم للبروتين (16.21%)، الكربوهيدرات (68.63%)، الألياف (15.37%)، الرطوبة (7.05%)، الدهون (5.21%) والرماد (1.70%). وأحرزت بواسطة الطرز الوراثية دخن ديمبي وهجين سودان 55555 وسودان اتنين وديمبي شنقل طوباي و هجين سودان 10294 وهجين سودان 10294 وهجين سودان 10294 علي التتابع .

Abstract

This research was conducted to study the variability under normal and stress watering conditions, genetic diversity by using RAPD markers and quality traits among thirty grain pearl millet (*Pennisetum glaucum* L.) genotypes. Two field experiments were conducted at Sudan University of Science and Technology, College of Agricultural Studies, Shambat, during two consecutive summer seasons of 2012 and 2013. A randomized complete block design (RCBD) with three replications based on split plot arrangements was used for the field experiments. Data was collected for ten growth and yield parameters included plant height (cm), number of leaves/plant, stem diameter (cm), leaf area (cm), Days to 50% flowering, panicle weight, number of grains \pancile, thousand seed weight (g) and grain yield ton/ha. Two different laboratory experiments were conducted, the first one to investigate genetic diversity among the thirty millet genotypes by using RAPD markers and the second one to investigate quality traits which included chemical compositions, mineral contents and physical properities of the thirty millet genotypes. The results of the separate and combined field experiments data showed that there were significant differences among most of studied traits for normal and stress watering conditions in the two seasons. Based on DNA molecular markers analysis, high level of polymorphism among genotypes was detected, indicating that the technique was efficient in determining the genetic diversity among millet genotypes. The results showed that there were significant differences between the 30 pearl millet genotypes for all the quality studied traits. The genotype HSD10319 scored the highest values grain yield of 2.51 and 2.90 ton\ha under water stress and normal watering, respectively. The means of the chemical compositions showed that, the highest values of protein (16.21%,), carbohydrates (68.63%), fiber (15.37%), moisture (7.05%), Fats (5.21%) and ash (1.70%) were obtained by the

genotypes Dembi millet, HSD55555, Sudan II, Dembi Shangal Toby, HSD10294 and HSD10294, respectively.

CHAPTER ONE

INTRODUCTION

Pearl millet (*Pennisetum glaucum{L.}* R. Br*)*is a grain crop belongs to the family Poaceae (Gramineae). It is an annual crop, cross pollinated and has chromosome number of (2n =14.).The genus *Pennisetum* is distributed through the tropics and sub tropics of the world. It includes about 140 species, one African species, *P. glaucum* (L.) was domesticated as the cereal pearl millet. The common names of pearl millet include pearl, bulrush, cattail or spiked millet and duckn (Gill, 1991). Today millet covers the food needs for more than 500 million people. The areas planted with millet are estimated at 15 million hectares annually in Africa and 14 million hectares in Asia and global production exceeds 10 million tons per agar (ICRISAT, 1987). Millet adapted to poor sand soils in dry areas, it is a summer cereal crop and produce a large number of tillers annually which contribute to the end product yield. In terms of annual production, pearl millet is the sixth most important cereal crop in the world following wheat, rice, maize, barely and sorghum (Stoskopf, 1985). The world production is around 33.4 million metric ton with an average grain yield of 699.0 kg ha^{-1} (FAO, 2002).

Pearl millet is a dual-purpose crop, its grains are used for human consumption and its fodder serves as feed for cattle in Asia and Africa, where more than 95% of the crop is produced. it is grown primarily as a grain crop and its grains are comparatively has high nutritive value than grains of other cereals.

In the Sudan, pearl millet is the preferred staple food for the majority of inhabitants in Western Sudan (Kordofan and Darfur States). Among the cereals, it comes second to sorghum in cultivated areas and total production. The crop is mainly raised under traditional farming methods, where the rainfall is between 200 – 800 mm (Abuelgasim, 1999) and the average yield

was 653 kg/ha (AOAD, 2008). The short rainy season and fluctuation in rainfall expose the crop to drought stress; therefore, there is a highly need to breed for drought tolerant and early maturing cultivars. Grain yield as a character in pearl millet as well as in all crop plants is quantitative in nature and is poly genetically controlled. Selection on the basis of grain yield character alone is usually not very effective and efficient. However, selection based on grain yield and its components and secondary characters could be more efficient and reliable. Knowledge of the genetic variability and interrelationship between yield and its components and among the component characters themselves can improve the efficiency of selection in plant breeding (Abuali *et al.*, 2012).

The grains of pearl millet is comparatively highly nutritive value more than the grains of other cereals crops, especially in carbohydrates, fats and mineral contents. Most of millet improvement programs in the Sudan were focused on obtaining millet varieties or hybrids characterized with high yield, early maturity, prevailing pests and diseases …etc (Abu Elgasim, 1989). On the other hand, meager studies were applied to compare the differences between millet genotypes in quality characters (protein, carbohydrates, minerals, vitamines …ect), (Subi, 2012).

Molecular marker technologies can assist conventional breeding efforts and are valuable tools for the analysis of genetic relatedness, identification and selection of desirable genotypes for crosses as well as for germplasm conservation in gene banks. Molecular markers, such as SSRs and RAPD have been widely used in germplasm evaluation in the world. The use of molecular marker to interpret population structure provides much greater resolution than other types of markers because of high level of polymorphism (Cho *et al.*, 2000).

The main objectives of the study were:

1- To study variability for some growth and yield traits in 30 pearl millet (*Pennisetum glaucum* L.) genotypes under tolerance to water stress.
2- To study molecular characterization of the 30 pearl millet genotypes inorder to investigate the genetic diversity among them using RAPD markers.
3. To investigate and to determine the chemical compositions, mineral contents and physical properties of the 30pearl millet genotypes.

CHAPTER TWO

LITERATURE REVIEW

2.1 General background

Although it is an important food crop in the Sudan, pearl millet did not received much attention to improve it prior to 1974 when a proper millet-breeding program was started. This program was strengthened in 1977 through cooperation with ICRISAT. The program objectives were centered around developing high yielding, drought tolerant, early-maturity verities with acceptable grain quality and resistance to prevailing pests and disease (Abu Elgasim, 1989). Pearl millet (*Pennisetum glaucum* L.R.Br*)* is a diploid species (2n = 14) believed to be originated in West Africa (Muhammad *et al.*, 2003). It is now widely cultivated in different parts of the world. Pearl millet is of a great importance in the semi-arid tropics, where it is the stable food for millions of people (FAO, 2008). The crop is grown commonly under the most difficult farming conditions, including those in drought-stricken area, where soil fertility is low and food supplies are dependent on rainfall. Pearl millet growing in areas suffer from erratic rainfall which has high within and between year variability (Abuali *et al.*, 2012).

2.1 Diffinition of drought (water stress or water dificiency)

The crop grown under unfavorable environments withstands the stress through different modifications. These include developmental, morphological and biological mechanisms (Turner and Begy, 1981). Plant adaptation mechanisms are classified into three major categories:

2.1.1 Drought escape

Which is particularly an important strategy for phenological development with- in the period of soil moisture availability to minimize the impact of

drought stress on crop production in environments where the growing season is short and terminal drought stress predominates (Turner, 1986). Also, later flowering can be beneficial in escaping early season drought when it is followed by rains (Ludlow and Muchow, 1990).

Kramer1980,reported that Drought is actually meteorological event which implies the absence of rainfall for a period of time, long enough to cause moisture – depletion in soil and water deficit with a decrease of water potential in plant tissues.In addition Begy, 1980 stated that drought acts as serious limiting factor in agricultural production by preventing a crop from reaching the genetically determined theoretical maximum yield (). The effect of drought on crop production is well known (Singh, 1990). The fact that most of the crops are sensitive to water deficits specially in the period from flowering to seed development stage is well documented (Salter, 1967). Even the crops grown in arid and semi arid regions such as pearl millet, sorghum and pigeon pea are affected by drought at the reproductive stage. Plant adaptation to drought stress (as measured by grain yield) depends on different traits, response, the time and intensity of its occurrence. An attempt to breed for improved adaptation to stress makes sense only if the stress is reasonably well defined. (Abuali2006) Drought (water stress and water defiency) is one of the most common environmental stresses that affect growth and development of plants through alterations in metabolism and gene expression (Leopold *et al.*, 1990). The effect of water stress on crop growth and yield depends upon the degree, duration of the stress and the developmental stage at which the stress occurs (Hasio *et al.*, 1976; Sullivan and Eastin, 1974

2.2 Mechanisms of water stress

Tuner and Begy,1981, stated that the withstand of the crops grown under unfavorable environment is escape through different modifications .Different methods of plant adaptation are divided into three major mechanisms, drought escape, drought avoidance. The first defined by

(Tunor 1986 and Ludlow and Muchor 1990) as strategy caused by crops in order to shorten the phonological development or late flowering ,this strategy is beneficial in escaping early from drought .The second is defined by (Blum ,1988) Defined as the ability of plants to attain a relatively high level of hydration under conditions of soil and atmospheric water stress (Blum, 1988). Plant can exhibit dehydration avoidance through increasing water uptake and reducing water loss by means of morphological or physiological modifications. The third one is defined by (Ugherughe ,1986) the ability of tissues of plant to tolerate drought Plants tolerate drought by ability of their tissues to withstand water stress. The mechanism of drought tolerance is maintenance of turgor through osmotic adjustment, increase in elasticity in cell and decrease in cell size and desiccation tolerance by protoplasmic resistance (Ugherughe *et al.*, 1986).

2.3 Effect of waterstress on growth and development of pearl millet

Most of the plant growth and development are sensitive to water stress (Turner and Kramer, 1980) and response of crop plants to drought periods is major factor influencing their adaptation to environments. Bunting and Kassam (1988) reported that during the growth of many plants, there are periods during which plants are susceptible to drought stress. Moreover, Bunting and kassam (1988) indicated that the time of transition from the vegetative to reproductive phase in cereals is the most sensitive to water deficit.

2.3.1 Improvement of tolerant to water stress in millet

Usually, development of drought – tolerant cultivars is hindered by poor understanding of the mechanisms of drought tolerance and by inadequate selection techniques (Bruckner and Frohberg, 1987; Richards, 1996). (Byrne *et al.*, 1995) reported that Strategies for improving drought tolerance in cearals and other crops include selection in low – stress environments, high stress environments and a combination of stress and no stress environments. Selection for high yield in an optimum environment is effective because

genetic variation is usually maximized and genotype – by – environment interactions is low (Richards, 1996). In addition to , selections is often complicated by low heritability of traits, non – uniform testing conditions and large genotype – by – environment interactions (Hamblin *et al.,* 1980; Smith *et al.*, 1990).

Responses of pearl millet genotypes to drought depends on growth period and differed from a period to another , during the period of seedling establishment to a point just prior to panicle initiation showed that drought has little effect on grain yield (Anon, 1984). However, Seetharama *et al. (*1984) reported that water stress during seedling stage resulted primarily in poor crop establishment, and consequently low grain yield. On the contrary, Farah *et al.* (1987) reported that stress during vegetative stage of sorghum growth affect yield through reduction in grain number. Many investigators e.g(Seetharama *et al.*, 1984; Mahalakshmi and Bidinger, 1988) stated that water stress during the panicle development stage was reported to have more severe effects on grain yield. They added the main effect on grain yield was through the number of grains per head and number of heads per unit area, but the loss

2.4. Effect of water stress on yield and yield components

(Mahalkshmi and Rao, 1990) and Abuali 2006 reported that The effect of water deficit on yield and yield components have been the subject of many investigations. Moisture deficit was found to account for 65% of variation in grain yield of sorghum and pearl millet

Timing of water supply generally has a larger effect on grain yield than total quantity of water for many crops (Shaw, 1988). Both pearl millet and grain sorghum are most sensitive to water stress during flowering and grain filling (Garrity *et al.*, 1983 and Hattendorf *et al.*, 1988).

Grain yield of pearl millet genotypes was found to be linear with severity of stress (Mahalakshmi *et al.*,1991). Grain number per unit area and grain size were reduced by severity of water deficit, where as grain yield and grain number, were affected by the time of the stress onset at all intensities. Grain number per panicle was found to be more affected by severe stress than panicle number.Also Abuali 2005 and Grant *et al.* (1989) showed that moisture stress occurred during early grain development significantly reduced kernal number per ear.

Field trails with pearl millet, irrigated and rainfed, showed significant differences between those two moisture regimes in grain yield, time to 50% flowering, time to maturity, number of heads per unit area, head mass and grain mass (Osmanzai, 1992) and(Abuali, 2006.)

These reports showed that plant height decreases with water deficit imposed at different stages of plant growth, except after anthesis. Eldichkery (1992) indicated that plant height was reduced as watering interval was increased. In field trails with rice, Gruz and Toole (1984) showed that water stress resulted in reduction of leaf area, plant height and number of tillers per plant. Number of tillers per plant was progressively increased with plant age where as it was decreased with water deficit. These results were in accordance with those reported by Conover *et al. (*1989) who found that panicle number and tiller number decreased by water stress in pearl millet. Similar findings were shown by Unger (1991) and Vanderlip. (1991), who reported substantial tillers production as a result of water stress. Leaf area index increased with plant age but was reduced with water stress (Payne *et al.*, 1991).

The shoot dry weight increased with plant age, where as it was declined with water regime. Mahalakshmi *et al.* (1991) found that biomass was reduced due to water deficit in two experiments under dry and rainy season conditions. Also Conover *et al.* (1989) reported that shoot dry weight decreased by water

stress in pearl millet. Muchow (1989) reported that the decrease in biomass of pearl millet in response to water deficit was associated more with reduction in radiation efficiency. Similarly, Ibrahim *et al. (*1985) found that dry weight was reduced significantly by water stress.

The effect of water stress on days to flowering reported by Anon (1984) suggested that flowering of pearl millet was delayed by water stress, with the effect being more pronounced in the tillers. On the contrary, Mahalakshmi *et al. (*1991) reported that there was no difference between two groups of tall and dwarf hybrid pearl millet growing in dry and rainy season in the time to flowering. A synchronous tillering habit in pearl millet was reported by some workers as an adaptive feature to water stress, allowing for development plasticity during the early stages of growth (Seetharama *et al.*, 1984).

2.4 Genetic variability

Genetic variability is essential to secure the success of any breeding programme. Selection is not effective unless considerable genetic variation is present in the population. Evidence for the existence of considerable amount of variability in pearl millet has been reported by investigators, and the germplasm resources are still largely un exploited (AbuElgasim, 1999). Pearl millet is grown in harsh environments and exposed to a variety of stresses such as drought, heat and low nutrient supply during the crop season. The cultivars targeted for these areas need to have a certain degree of adaptation to such stresses, and should have ability to take advantage of favorable growing seasons during better rainfall years (Yadav and Weltizein, 1997).

Genetic variability found in over 140 species of genus Pennisteum offers a vast potential for improvement through breeding and selection (Gill, 1991). Berwal and khairwal (1997) found highly significant differences in plant height, number of tillers, stem diameter and leaf area of pearl millet. They

predicated successful crosses between these accessions to improve each of these traits.

Berwal and Khairwal (1997), in their study of genetic divergence in pearl millet, where forty-two accessions were evaluated, indicated highly significant differences in plant height, number of tillers, stem diameter and leaf area. They predicted successful crosses between these accessions to improve each of these traits. Recently genetic variability in millet for different growth and yield characters was reported by many another (Abuali 2006;Subi 2012,Subi *et al* 2013 and Elsadig ,2016)

2.5 Molocular characterization

Previous generations of molecular markers were unable to detect enough genetic polymorphism among closely related millet cultivars to make them efficient tools for interpreting population structure. However, SSR markers are well suited to the task. In millet, the highly polymorphic nature of SSR motifs is coupled with a low level of homoplasy observed in millet cultivars (Chen *et al.*, 2002), providing an appropriate tool for population genetic studies. About 2240 micro satellite markers are now available through the published high-density linkage map (Moure *et al.*, 2001) or public database. The application of micro satellite markers in millet include characterization of the genetic structure of the cultivated millet, genetic diversity, determination of the purity of breeding material or seed stocks, prediction of hybrid performance and heterosis and the analysis and tagging of valuable quantitative trait loci (QTL) and genes (Weising *et al.,* 1995).

2.6 Quality parameters

Pearl millet is known for its culinary uses as well as health benefits. It is cultivated in countries of Africa and the Indian subcontinent since prehistoric times. Pearl times earlier known as bird food come in several delicious flavors what is unique about this cereal is that it may be as creamy as mashed

potatoes as fluffy as rice ,in India pearl millet are regarded as major sources of dietray energy and nutritional security for poor farmers and consumers, apoat. Lman alfering taste these millets contain essenlial minerd and nutnents uhich poruide the bodey uith avaiety of advantages In Ethuopia the center of diveisity for the crop it is used to make a flat bread known as injera in India flat bread called rote as bird seeds and livestock fodders in western Europe and north America but it has gained popularity as a delicious and nutrition benefits and gluten free status, Currently India is the leading commercial producer for pearl millet followed by China and Nigeria. Millets are a great source of starch making it is a high energy food it is also an excellent source of protein and fiber. It is said that amino acids in pearl millet are more easily digest able than the ones found in Wheat. Millet is a significant component of several necessary compounds including a denosine triphosphate (ATP) this element is also a crucial component of nucleic acids, which are the building blocks of the genetic code. Phosphorus is a constituent of lipid containing structures such as cell membranes and nervous system structure. Recent studies have proven that regular consumption of pearl millets help in preventing gallstone in women they contain insoluble fibers which not only speed up intestinal transit time but also reduce the secretion of bile acids. Pearl millet are known to increase insulin sensitivity and lower level of triglycerides, regular consumption of pearl millet breast cancer in pre menopausal women pearl millet contain phytonutrient lignin, which is very beneficial for the help of the human body with the help of natural flora lignin s get converted to mammalian lignins and they fight against hormone dependant cancers and reduces the cardial arrestes. Consumption of pearl millet helps in minimizing the risk of type2 diabetes, Being a good source of Mg and act as a co factor in a number of enzymatic reactions, pearl millet due to essential nutrients such as methionine an amino acids B complex vitamins niacin, thiamine and riboflavin folic acid lecithin, potassium

,magnesium and zinc. Millets are effective in several roles Niacin reduces cholesterol, while Mg is essential for maintaining good heart health, as it lowers blood pressure and reduces the risk of heart attacks.

The isolation procedure of protein, carbohydrates, minerals or vitamins from millet is highly needed among millet genotypes in order to base selection on the highly nutritive value genotype in addition to the ability of this selected genotype to tolerate drought. This procedure is different from that of other cereal crops, due to differences in protein and carbohydrates properties (Resurreccion *et al.*, 1993).

CHAPTER THREE

MATERIALS AND METHODS

3.1 The field experiments site

The field experiments were conducted during two successive summer seasons of 2012 and 2013., at the Experimental Farm of the College of Agricultural Studies ,Sudan University of Science and Technology at Shambat (32º32′E. longitude, 15º40′N latitude, and 380 meters above the sea level). The climate of Shambat is characterized short - humid air during the summer and cold - dry during the winter. The soil of Shambat is highly saline-sodic clay. The soil particles proportions follow the order: clay, silt and sand where the clay comprises the higher proportion. Monthly mean maximum and minimum temperature and total rainfall were recorded for the side (Appendix 2).

3.2 Grain millet genotypes used in the study

Thirty grain pearl millet (*Pennisetum glaucum* L.) genotypes used in this study and were shown in Table 3.1. Twenty of them were obtaining from Gene Bank, Plant Genetic Resources (PGR) Agricultural Research Corporation (ARC), Wad-Madani, Sudan and the other ten from different sites of main production areas of pearl millet at Darfour States, Sudan (Table 1).

Table3.1 Name and sources of 30 pearl millet genotypes used in the study

Millet Genotypes	Source
1. HSD7131	PGR Unit, Gene Bank, ARC, Sudan.
2. HSD7132	PGR Unit, Gene Bank, ARC, Sudan.
3. HSD7133	PGR Unit, Gene Bank, ARC, Sudan.
4. HSD7134	PGR Unit, Gene Bank, ARC, Sudan.
5. HSD7135	PGR Unit, Gene Bank, ARC), Sudan.
6. HSD10291	PGR Unit, Gene Bank, ARC), Sudan.
7. HSD10292	PGR Unit, Gene Bank, ARC, Sudan.
8. HSD10293	PGR Unit, Gene Bank,ARC), Sudan.
9. HSD10294	PGR Unit, Gene Bank, ARC, Sudan.
10. HSD10303	PGR Unit, Gene Bank, ARC, Sudan.
11. HSD10309	PGR Unit, Gene Bank, ARC, Sudan.
12. HSD10313	PGR Unit, Gene Bank, ARC, Sudan.
13. HSD10318	PGR Unit, Gene Bank, ARC, Sudan.
14. HSD10319	PGR Unit, Gene Bank, ARC, Sudan.
15. HSD10331	PGR Unit, Gene Bank, ARC, Sudan.
16. HSD10354	PGR Unit, Gene Bank, ARC, Sudan.
17. HSD10362	PGR Unit, Gene Bank, ARC, Sudan.
18. HSD10376	PGR Unit, Gene Bank, ARC, Sudan.
19. HSD10392	PGR Unit, Gene Bank, ARC, Sudan.
20. HSD55555	PGR Unit, Gene Bank, ARC, Sudan
21. Ashana	Released variety by ARC,Sudan.
22. SADC (Long)	Released variety by ARC,Sudan.
23. SADC (Togo)	Released variety by ARC,Sudan.
24. Ugandi	Released variety by ARC,Sudan.
25. Sudan II	Improved variety, ARC,Sudan.
26. MCNELC	Released variety by ARC,Sudan.
27. Dembi Millet	Local variety, E. Darfur State, Sudan.
28. Dembi Shangal Toby	Local variety, N. Darfur State Sudan.
29. Dembi Kabkabia	Local variety, N. Darfur State Sudan.
30. Dembi Sea	Local variety, N. Darfur State Sudan.

PGR = Plant Genetic Resources.
ARC = Agriculturai Research Corporation

3-3Watering treatment:

The whole experiment received equal quantities of water at **7** days interval for establishment, and then watering treatment was introduced four weeks after sowing. Water stress applied at different stages of growth as follows:

W_0: Watering every **7** days throughout the growing season (control).

W_1: Watering every **7** days till four weeks passed from sowing and then watering every **21** days till the grain reached physiological maturity.

3.4Design and layout of experiments

The field experiments were laid out in a randomized complete block design with four replications .The treatments were arranged in a Split – plot arrangements. The water intervals (7 days and 21 days) were assigned as main plots and the thirty grain millet genotypes as subplots. The experiment field was disc plowed, disc harrowed, leveled and ridged up north south, 70 cm apart.The land was divided into 2×3.5 m^2 plots, each composed of four ridges two meters long . Seeds were sown three seeds per hole spaced at 20 cm between holes Sowing date was the 6^{th} of July 2012 and 7^{th} of July 2013 for the two consecutive summer seasons. Seed rate applid was 2.5 kg/fed., Nitrogen fertilizer (urea 46% N) 80 Kg/F was applied in two equal doses after three and six weeks from sowing date, respectively. Hand weeding was conducted when needed. Thinning was carried out one week after sowing to raise two plants/hill.

3.5 Data collection

At each of the two seasons when the plants reached physiological maturity, five plants from the two inner ridges at each plot were randomly selected and tagged at each plot separately and from them data for the following growth

and yield characters except days to 50% flowering were collected as the following:

3.5.1 Growth characters

3.3.1.1 Plant height (cm)

The plant height was measured from the base of the main stem to the tip of panicle using meter tape.

3.5.2 Number of leaves/plant

The numbers of leaves/plant was counted for all tagged plants and the average of them was estimated.

3.5.3 Stem diameter (cm)

It was determined at maturity on the stalk at 10cm above the ground level.

3.5.4 Leaf area (cm^2)

It was calculated according to the following formula described by sticker (1974) method:

Leaf area (LA) = maximum length ×maximum width × 0.75

3.5.5 Days to 50% flowering

The days to 50% flowering were recorded from sowing date up the day when 50% of the plants had fully exerted heads.

3.3.2 Yield and its components

3.5.6 Number of panicles\plant

The number of the panicles of the five plants of each millet genotype at each plot was counted.

3.5.7 Panicle weight(g)

The weight of the panicles of five plant of each genotype at each plot was determined as average.

3.5.8 Number of grains/panicle

It was obtained by dividing the grain weight per main head by the corresponding weight of 1000-grain then multiplied by 1000.

3.5.9 The 1000 grain weight (g)

The weight of a random sample of 1000 grains taken from the grain yield of each ecch millet genotype.

3.5.10 Grain yield (t/ha)

The harvested heads from each genotype were air dried and threshed in bulk, then weighed and grain yield was calculated by the following Formula:

$$\text{Grain yield (t/ha)} = \frac{\text{grain weight (g) /plot} \times 10000\ (m^2)}{\text{Plot area } (m^2) \times 1000 \times 1000}$$

3.6.1 Statistical analysis

The collected data were subjected to different statistical analyses as follows:

3.6.2 Analysis of variance

3.6.3 Individual analysis of variance

It was carried out for all studied characters in each season separately according to the procedures described by Gomez and Gomez (1984) for split-plot (Table 2). 3.4.1.2 Combined analysis of variance

It was done for the characters in which the mean squares of errors were homogenous. It was carried out following the procedures described by Gomez and Gomez (1984) based on split plot design (Table 3).

3.4.1.3. Coefficient of variation (CV%)

It was determined for each character in both seasons using the formula

$$CV\% = \frac{\sqrt{Mean\ square\ of\ error}}{Grand\ mean} \times 100\%$$but we use the Mstat programme for computing this.

Table 2. Analysis of variance for different characters of 30 genotypes of pearlmillet, evaluated under tow water treatments, for each season separately, during the season (2012/13)

Source of variation	D.F	M.S	F	Expected mean squares
Replications	(r-1) =2	MQ1	MQ1 / MQ3	
Treatments	(t-1) = 1	MQ2	MQ2/MQ3	
Error (a)	(t-1) (r-1) = 2	MQ3	-	
Genotypes	(g-1) = 29	MQ4	MQ/MQ6	
Gen.× Treat.	(g-1) (t-1) = 29	MQ5	MQ/MQ6	
Error (b)	t (r-1) (g-1) = 116	MQ6	-	
Total	**(rtg-1) =179**			

r = Replications, t = Treatments (main factor), g = Genotypes (sub factor), MQ1, MQ6, Mean squares for replication, factor (A), error (a), factor (B), A×B interaction and error (b), respectively.

Table 3. Combined analysis of variance for characters of 30 genotypes of pearl millet, evaluated under tow water treatments, with three replications in two seasons under Shambat location

Source of variation	D.F	MS	F	Expected mean squares
seasons	(2-1) =1	MQ1	MQ1/ MQ5	
Replications /seasons	S(r-1) = 4	MQ2	-	
Treatments	(t-1) = 1	MQ3	MQ3 / MQ5	
Treat ×seasons	(t-1) (S-1) = 1	MQ4	MQ4 / MQ5	
Pooled error (a)	S(r-1) (t-1) =4	MQ5	-	
Genotypes	(g-1) = 29	MQ6	MQ6/MQ10	
Gen. × Treat	(g-1)(t-1) = 29	MQ7	MQ7 / MQ10	
Gen. ×S	(g-1) (2-1) = 29	MQ8	MQ8 / MQ10	
Gen.× Treat×S	(g-1) (t-1) (4-1) = 87	MQ9	MQ9 / MQ10	
Pooled error	Sr (g-1) (t-1) = 174	MQ10	-	
Total	**(Srtg-1) = 359**			

S= Season, r = replication, t = treatment, g = genotype

MQ1,…..MQ10 = mean squares for seasons, replications within seasons, factor (A), S × A interaction, error (a), factor (B), S×B , A×B, S×A×B interactions and Error (b), respectively.

3.6.2 Mean separation

3.6.3 Between water treatments

Means of water treatments were separated using Duncan's Multiple Range Test (DMRT) at 5% level of significance according to procedure described by Gomez and Gomez (1984) as follows:

3.6.4 Comparison between genotypes

The means were separated using the least significant difference (LSD) at 5% level of significant according to formula: $LSD = t\alpha x \sqrt{\frac{2\ EMS}{r}}$

Where

r = Number of replication's, EMS = mean squares.

α = level of significance for t – value (0.05) computing it using Mstat programme. 3.7 Molecular characterization of the 30 pearl millet genotypes

3.7.1 DNA extraction and PCR amplification:

Genomic DNA of each genotype was extracted by a sap-extraction method (CIMMYT, 2005) from 200 mg of fresh leaf tissues. Leaves of 2-week-old seedlings were placed in a 15ml falcon tubes and 5 ml of extraction buffer (50 mM Tris–HCl, 25 mM EDTA, 1 M NaCl, 1% CTAB, 1 mM 1, 10-phenathroline, and 0.15% 2-mercaptoethanol) was added. The contents of each tube were blended using a rod blender. The extract was incubated at 60°C for 1 h, and then mixed with equal volume of chloroform-isoamyl alcohol (24:1). After centrifuging at 12,000 rpm, the supernatant was transferred to a new tube and incubated with isopropanol for 30 minutes to precipitate the DNA in a pellet form. The pellet was dried and re-suspended in 200 µl of TE buffer (10 mM Tris–HCl, 0.1 mM EDTA, pH 8.0). The DNA solution was mixed with 200 µl of 8M ammonium acetate and 400 µl of cold absolute ethanol for 30min, centrifuged at 2000 rpm for 10min, the supernatant was decanted, DNA pellets were air- dried at room temperature. The DNA was then re-suspended in 300 ml of TE buffer and stored at -20°C till used.For genetic diversity studies, 10 RAPD primers (Operon Tech., NY, USA) were used to amplify genomic DNA of the 30 genotypes. Primers' codes and sequences are shown in Table 2. Each PCR amplification reaction was carried out in a total volume of 20 µl. The PCR mixtures contained (Final concentration): 5X FIRE Pol PCR Master Mix (Ready to load), 5 X reaction buffer (0.4 M Tris-HCl, 0.1 M (NH_4) SO_4, 0.1% w/v Tween 20), 12.5 mM dNTPs, 50ng of the primer under test, 1 U Taq polymerase and 2 µl template DNA. The amplification cycling procedure used consisted of one cycle at 94°C for 5min, followed by 35 cycles of initial denaturation at 94°C for 1min,

annealing at 32°C for 3min, extension at 72°C for 2min and a final extension step at 72°C for 10 min. PCR products were electrophoresed in 1.5% agarose gel (Sawada et al., 1993). and the gel was visualized under transillumination cabinet (Model TM-10E, Uvitec. Product).

3.7.2 Data statistical analysis

DNA fragments obtained by RAPD markers were scored as present (1) or absent (0). Polymorphism information content (PIC) values were calculated as, described by Anderson *et al*. (1993), as follows:

$$PIC= 1-\Sigma P_{ij}^2$$

Where, P_{ij} is the relative frequency of the j^{th} allele of the i^{th} locus, summed over all alleles for individual marker locus over all lines. A marker with a PIC value of more than 0.5 is considered as highly informative, between 0.25 and 0.5 as informative and less than 0.25 as slightly informative (Botstein *et al*., 1980). Similarity between the lines was analyzed on the basis of their scores. Data were used to create similarity matrices using the PAST 3.01 software package.(StatSoft, Inc., 2003).Dendrograms were constructed based on Jaccard's similarity coefficients (Jaccard, 1908).

Table 4. Primer names and sequences

Sr. No	Primer code	Primer sequence 5' - 3'
1	OPA9	GGGTAACGCC
2	OPA11	. CAATCGCCGT
3	OPY15	AGTCGCCCTT
4	OPY17	GGTGATTCGG
5	OPK15	ACCTGCCGTT
6	OPK16	CTGCGTGCTC
7	OPK17	CAGCGGTCAC
8	OPL16	AGGTTGCAGG
9	OPL17	AGCCTGAGCC
10	OPL18	ACCACCCACC

3.8 Laboratory experiments for Qualitytraits:

The quality laboratory experiments for 30 millet genotypes were carried out at the laboratory of National Food Research Center, Khartoum North, Shambat, Sudan. The seeds of the 30 millet genotypes were manually and separately cleaned to remove dust, broken seeds and other extraneous materials, then the dry samples were later milled and the processing of the samples was carried out in a randomized complete design (CRD) with 3 replicates for the following procedures:

3.8.1 Chemical composition analysis:

The chemical analyses were carried out according to methods described in AACC (2002). The moisture content at 105°C /12h, Crude protein was determined by the KjeldhalD s method (N x 5.95), as well as ash content at 550°C/5h, Crude fat in Soxhlet apparatus (solvent in above reference). Available carbohydrate was calculated by subtracting the sum of fat, protein, fiber and ash as a percentage from 100 as described by West *et al*. (1988).

3.8.2 Crude protein

Seeds were taken to the laboratory for crude protein determination and were analyzed by a modified Kjelkahl digestion method (Summerfield *et al.*, 1977). Samples were, weighed (about 0.5 g) and put into 25 × 200 mm pyrex test tubes with 1.0 g K_2SO_4, 0.1 g Na_2SeO_3, and 10 ml concentrated H_2SO_4. Samples were then digested at approximately 400 C on a block digester until they turned to a clear amber color. The tubes were removed and placed in test tube racks to cool. The digestate was then quantitatively transferred to a 100 ml volumetric flask along with 3g K_2SO_4. The sample was diluted to the mark with de-ionized water and transferred to polyethylene bottles.

Aliquat of the digest (0.4 ml) was injected into a Technicom Auto analyzer (manufactured by Technicom Industrial System, Terrytown, New York),

which quantitatively detected ammonia by indophenol-blue formation in the presence of sodium phenate and sodium hypochlorite. Different peaks for different samples were drawn and ppm of nitrogen were found from the peaks in reference to standards. Percent nitrogen was calculated be the equation:

$$\%N = \frac{(PPM)\ (100)\ (10)\ 100}{\text{Sample weight}}$$

Percent crude protein was obtained by multiplying the percent nitrogen by 6.25 based on the assumption that about 16% protein in nitrogen (Chapman and Pratt,1961).

3.8.2 Physical properties:

Include colour, Granule size (mm), 100 seeds weight, and Taste. The granules size of millet seeds were recorded using vernier calipers (model: E H B Stainless, Hardenend, Germany).

3.8.4 Mineral profile:

The mineral content included Ca, P, Zn, Mn and Cu the samples were extracted and determined by atomic absorption spectrophotometer (model: Instrument shimadzu - AA - 6800) according to method given in AOAC (2000).

3.8.5Statistical analysis of quality trials:

The statistical analysis of variance for the collected data of the chemical analysis, mineral contents and physical properties was carried out for a randomized complete design according to Gomez(1984). for CRD. The means were separated according to Duncan Multiple Range Test at 5% level of probability Duncan (1955).

CHAPTER FOUR

RESULTS

4.1 Phenotypic Variability

4.1.2 Plant height (cm)

The mean of plant height was highly significant ($P \leq 0.01$) affected by genotypes in season (2012), whereas, had nosignificant difference ($P \leq 0.05$) due to stress in the same Season Appendix (1). This character was none significant differences ($P \leq 0.05$) in second season by stress, genotypes and interaction stress x genotypes appendix (2). Similarly, as shown in Appendix (3), the combined analysis of variance revealed highly significant ($P \leq 0.01$) among genotypes, under the two level of water stress (7 days 21days) for this treatments, genotypes, seasons, and interactions (season x water; season x genotypes; water x genotypes and season × genotype × water), Appendix (3). The highest values of plant height (216) and (142.72) were regarded by genotype HSD7132 in (7 days and 21days) respectively and the lowest values (33) and (33.7) were regarded by the genotype HSD10392 and genotype HSD10291 under 7 days and 21days, respectively (Table 5).

4.1.2 Number of leaves/plant

Highly significant differences ($P \leq 0.01$) were shown for the 30 millet genotypes for number of leaves at season 2012 and 2013 appendices (1 and 2), this character showed significant ($P \leq 0.05$) with gentypes in season (2012) appendix (1). The results of seaon2013 showed highly significant differences ($P \leq 0.01$) in genotypesxstress Appendix (2).The means of number of leaves due genotypexstress to season2 results showed that the highest values (11.6) and (11.53) regarded by the genotype HSD7132and HSD7133 in (7 days) and the lowest values (4) and (4.3) were regarded by the genotypes HSD10313 and 10319 in (21 days), (Table,6).

Table 5.Effects of water stress and millet genotypes on plant height (cm) during 2012 and 2013 seasons and combined analyses.

Season	2012			2013			Combined(2012-2013)		
Treatments	**7 days**	**21 days**	**Mean**	**7 days**	**21 days**	**Mean**	**(7 days)**	**21 days)**	**Mean**
HSD7131	61.00	85.50	73.25	172.33	153.00	162.65	116.62	119.25	7.94
HSD7132	59.20	47.83	53.52	216.67	179.00	197.84	137.94	113.41	125.68
HSD7133	79.40	95.50	87.45	202.67	169.50	186.09	141.04	130.00	135.57
HSD7134	62.00	85.00	73.50	185.500	157.00	171.25	123.75	121.00	122.38
HSD7135	102.50	97.00	99.75	182.33	159.00	170.65	142.45	128.00	135.23
HSD10291	96.30	33.70	65.00	197.17	165.00	181.85	146.74	99.35	123.05
HSD10292	122.20	49.00	85.60	169.50	163.00	166.25	145.85	106.00	125.64
HSD10293	67.30	45.80	56.55	184.50	151.00	167.75	125.90	98.40	112.15
HSD10294	132.20	45.33	88.77	183.67	161.83	172.75	157.94	103.58	130.76
HSD10303	98.30	80.02	89.61	197.17	165.50	181.30	147.74	122.76	135.25
HSD10309	185.3	62.00	123.65	182.83	157.67	170.25	184.07	109.84	146.96
HSD10312	154.2	38.67	96.44	184.33	167.67	176.00	169.27	103.17	136.22
HSD10313	181.3	49.00	115.15	185.00	147.67	166.30	183.15	98.34	140.75
HSD10318	181.7	34.67	108.19	187.50	150.33	168.92	184.60	92.50	138.55
HSD10319	187.2	64.20	125.70	193.67	165.00	173.67	190.44	114.60	152.52
HSD10331	142.3	145.67	143.99	199.50	150.00	174.75	170.90	140.00	155.45
HSD10354	176.7	119.80	148.25	193.67	153.67	173.67	185.19	136.74	160.97
HSD10362	182.00	90.00	135.50	185.50	173.67	179.59	183.75	131.84	157.80
HSD10376	187.00	126.20	156.60	197.00	173.17	185.09	192.00	149.69	170.85
HSD10392	171.00	28.80	99.90	180.33	155.00	94.00	125.67	91.90	154.74
Ashana	78.20	187.50	132.85	185.17	162.33	173.75	131.69	174.92	153.31
SADC (Long	61.00	181.67	121.34	206.17	142.03	174.10	133.59	161.85	147.72
SADC (Togo	59.20	168.50	113.85	191.83	142.17	166.99	125.52	155.34	140.43
Ugandi	79.40	165.67	122.50	197.17	172.33	184.75	139.29	169.00	154.15
Sudan II	62.00	132.20	97.10	197.33	162.83	180.07	129.67	147.52	138.60
MCNELC	102.50	189.80	146.15	188.00	149.80	168.90	145.25	170.00	138.60
DembiShangal Toby	96.30	172.50	134.40	185.00	167.00	176.00	140.65	169.75	157.63
Dembi Kabkabia	122.20	205.20	163.70	181.00	157.17	169.09	155.60	181.19	155.20
Dembi Sea	67.30	193.30	130.30	205.00	168.83	186.93	136.15	181.07	158.61
Means	115.77	104.14	109.95	190.26	160.83	172.46	151.46	131.76	138.37
LSD (S)	44.92								
LSD (G)	3.41								
LSD c (S x G)	1.25								

Table 6. Effects of water stress and millet genotypes on number of leaves during 2012 and 2013 seasons and combined analyses

Season	2012			2013			Combined(2012-2013)		
Treatments	**7 days**	**21 days**	**Mean**	**7 days**	**21 days**	**Mean**	**7 days**	**21 days**	**Mean**
HSD7131	7.95	5.80	11.33	8.57	6.00	9.50	8.57	7.25	7.95
HSD7132	9.55	8.20	15.00	11.60	5.20	10.67	11.60	7.95	9.55
HSD7133	9.77	6.70	14.33	11.53	6.20	9.83	11.53	8.02	9.77
HSD7134	8.26	6.30	10.33	8.32	5.00	11.50	8.32	8.25	8.26
HSD7135	8.10	7.80	10.17	8.48	5.00	10.33	8.48	7.65	8.10
HSD10291	8.10	7.30	9.67	8.48	4.70	10.00	8.48	7.65	8.10
HSD10292	7.46	6.50	9.33	7.42	5.30	9.67	7.42	7.50	7.46
HSD10293	7.77	6.00	10.83	8.42	4.70	9.50	8.42	7.10	7.77
HSD10294	8.02	6.50	10.83	8.72	4.80	9.83	8.72	7.32	8.02
HSD10303	8.00	7.16	9.33	8.25	5.20	10.33	8.25	7.75	8.00
HSD10309	8.13	5.20	10.83	8.02	4.50	12.00	8.02	8.25	8.13
HSD10312	9.45	7.80	11.50	9.65	4.00	14.33	9.65	920	9.45
HSD10313	9.00	5.80	11.50	8.95	4.30	13.33	8.95	9.30	9.00
HSD10318	8.95	7.50	12.00	8.75	4.00	14.33	8.75	9.20	8.95
HSD10319	9.25	8.20	10.50	9.35	4.80	13.33	9.35	9.15	9.25
HSD10331	9.50	5.20	12.500	8.35	6.30	15.00	8.35	10.65	9.50
HSD10354	8.44	6.20	9.50	7.35	5.80	13.17	7.35	9.50	8.44
HSD10362	7.80	4.70	11.50	8.10	5.70	9.83	8.10	7.50	7.80
HSD10376	9.20	7.00	11.50	9.25	7.20	10.83	9.25	9.15	9.20
HSD10392	8.15	6.80	9.50	8.15	5.20	10.83	8.15	8.15	8.15
Ashana	9.70	8.70	10.33	9.02	9.00	11.33	9.02	10.17	9.70
SADC (Long	9.07	8.50	10.00	9.25	8.30	9.50	9.25	8.90	9.07
SADC (Togo	9.00	7.70	9.00	8.35	8.30	9.83	8.35	9.65	9.00
Ugandi	8.90	8.30	10.00	9.15	7.30	10.00	9.15	8.65	8.90
Sudan II	9.80	9.25	9.50	9.65	8.20	9.50	9.65	8.85	9.25
MCNELC	9.36	7.30	12.00	9.65	7.80	10.33	9.65	9.07	9.36
DembiShangal Toby	9.47	8.50	11.50	10.00	7.50	10.33	10.00	8.94	9.47
Dembi Kabkabia	9.75	8.30	11.50	9.90	7.70	11.50	9.90	9.60	9.75
Dembi Sea	9.13	8.20	10.50	9.35	7.80	10.00	9.35	8.90	9.13
Means	8.79	7.15	10.91	8.97	6.06	11.05	8.97	8.59	8.78
LSD S	1.5								
LSD G	1.90								
LSD S x G	3.10								

4.1.3 Stem diameter (cm)2

The analysis of variance indicated that the mean of stem diameter was highly significantly ($P \leq 0.01$) affected by genotypes in the two seasons whereas, only significant ($P \leq 0.01$) by stress and stress genotypes in the second season Appendix (1, 2). The combined analysis regarded, high significant difference ($P \leq 0.01$) between genotypes, seasons, and the interaction between all treatments except stress × seasons, which was significant ($P \leq 0.05$), (Appendix 3). The mean for stem diameter showed that heights values of the stem diameter (51.3) and (50) were obtained by the genotype HSD10291for (7days and 21 days) consecutively whereas lowest value (21.5) and (24.4) were revealed by the genotype ugandi for (7days), and the genotype HSD10376 for (21days), (Table 7).

4.1.4 Leaf area (cm)

The study showed significant differences ($P \leq 0.05$) for this character among genotypes in the seasons 2012 whereas significant difference ($P \leq 0.05$) was shown for interaction between stressxgenotype in seasons 2013 appendices (1 and 2). The combined analysis also showed highly significant difference ($P \leq 0.01$) among season and interaction (genotype x season and genotypes x season x stress) due to combined analysis appendix (3).For the means of leaf area register, the highest values due to combined analysis where (323) and (264) and 267.6 regarded by the genotypes SADC TOGOand UGANDI and MCLECN under water regimes (7days and 21 days) successively and the lowest values were regarded by the genotypes HSD10291 and HSD7135was (99.9) and (142), respectively (Table 8).

Table 7. Effects of water stress and millet genotypes on stemdiameter (mm) during 2012 and 2013 seasons and combined analyses

Season	2012			2013			Combined(2012-2013)		
Treatments	**7 days**	**21 days**	**Mean**	**7 days**	**21 days**	**Mean**	**7 days**	**21 days**	**Mean**
HSD7131	34.00	25.00	29.50	43.33	33.40	22.56	23.70	23.50	23.07
HSD7132	38.00	26.20	32.00	40.00	32.60	27.65	25.83	23.50	25.60
HSD7133	39.67	27.50	33.30	42.00	33.60	22.65	25.35	25.11	24.38
HSD7134	41.00	27.50	34.00	45.67	36.10	25.50	28.60	25.20	25.35
HSD7135	31.00	22.70	26.50	46.33	33.90	30.70	25.60	26.50	28.60
HSD10291	39.33	27.40	33.30	51.30	50.33	29.75	26.47	21.46	25.60
HSD10292	43.67	29.30	36.50	47.00	39.90	26.65	32.45	26.30	26.47
HSD10293	34.67	24.70	36.50	46.00	34.40	33.20	26.60	32.70	32.45
HSD10294	44.67	30.20	29.60	44.00	36.00	30.40	28.00	22.70	26.60
HSD10303	42.00	28.10	37.40	45.33	36.60	29.00	27.80	28.00	28.50
HSD10309	36.00	25.90	35.50	46.33	35.00	32.70	23.65	27.80	30.25
HSD10312	40.00	27.20	31.00	37.67	31.90	28.20	26.11	23.65	26.00
HSD10313	42.67	27.40	34.00	43.33	35.30	28.80	27.30	26.11	27.45
HSD10318	34.67	24.50	29.10	45.33	34.40	30.31	23.50	27.30	28.80
HSD10319	36.33	25.80	30.65	47.00	35.30	32.25	23.50	23.50	27.35
HSD10331	39.33	25.70	32.5	49.67	37.30	*29.65*	25.01	23.50	26.55
HSD10354	38.33	23.80	31.00	49.33	47.00	29.80	26.00	25.01	27.40
HSD10362	35.33	23.80	29.75	33.00	28.20	28.80	32.26	30.00	31.13
HSD10376	30.00	21.50`	25.75	32.00	26.20	28.80	20.40	23.26	26.50
HSD10392	44.00	28.70	36.35	34.00	30.70	29.12	27.36	20.40	24.80
Ashana	43.33	27.30	35.36	47.00	36.80	29.83	26.65	27.36	28.60
SADC (Long	40.33	25.50	34.91	49.67	37.20	24.45	24.70	26.65	25.55
SADC (Togo	37.33	24.60	31.00	49.33	36.80	27.00	24.70	24.32	26.00
Ugandi	38.00	25.70	32.00	24.41	2400	24.21	24.80	24.32	24.56
Sudan II	37.00	24.20	30.60	32.00	28.00	28.05	24.83	24.02	26.44
MCNELC	40.33	26.00	33.16	36.00	26.80	26.40	26.16	24.02	17.52
DembiShangal Toby	35.67	24.30	30.00	47.00	37.20	42.10	27.26	26.17	19.80
Dembi Kabkabia	36.00	24.11	30.50	49.67	38-70	44.00	27.26	27.26	18.88
Dembi Sea	46.00	44.67	35.34	49.33	40.90	45.11	27.65	27.65	23.44
Means	38.57	26.71	32.31	43.21	34.85	29.72	26.19	25.42	25.99
LSD S	4.36								
LSD G	2.13								
LSD S x G =	1.38								

Table 8. Effects of water stress and millet genotypes on leaf area (cm2) during

Season	2012			2013			Combined(2012-2013)		
Treatments	**7 days**	**21 days**	**Mean**	**7 days**	**21 days**	**Mean**	**7 days**	**21 days**	**Mean**
HSD7131	213.17	196.500	204.34	184.700	119.88	152.29	197.44	158.19	177.82
HSD7132	238.80	210.17	224.49	199.633	164.33	181.98	209.23	201.75	205.40
HSD7133	218.83	126.167	172.50	202.00	168.967	185.50	193.90	164.08	178.99
HSD7134	187.00	177.933	183.49	270.433	147.67	209.05	228.67	162.80	195.74
HSD7135	193.433	142.87	168.20	221.733	181.00	201.40	187.30	182.22	184.76
HSD10291	221.60	99.900	160.80	199.33	146.967	173.21	184.29	145.12	185.76
HSD10292	223.33	175.100	199.20	196.100	190.33	193.21	209.72	182.72	196.22
HSD10293	194.33	167.133	180.70	221.767	204.67	213.20	208.05	185.90	196.98
HSD10294	198.367	158.00	178.20	209.367	204.33	206.84	201.53	183.79	192.52
HSD10303	205.67	142.233	174.00	221.567	150.33	185.90	213.62	146.12	179.87
HSD10309	175.33	173.100	174.70	241.233	181.67	211.50	206.78	178.30	192.59
HSD10312	217.00	201.867	209.43	264.000	211.33	237.70	240.50	206.60	222.60
HSD10313	215.00	161.333	188.00	191.367	248.00	219.70	204.18	203.67	203.93
HSD10318	202.67	106.467	154.54	201.800	170.67	186.20	202.24	138.57	170.38
HSD10319	204.167	173.20	188.70	232.000	213.67	222.80	208.60	202.92	205.76
HSD10331	228.33	173.333	200.83	188.067	182.00	185.00	208.20	177.67	192.49
HSD10354	210.00	145.867	177.93	231.133	190.33	210.73	220.57	168.10	194.34
HSD10362	184.138	145.67	146.90	194.33	140.933	167.60	189.23	143.32	166.28
HSD10376	171.00	161.739	166.40	235.367	195.50	215.43	203.18	178.62	190.90
HSD10392	244.00	140.910	192.50	214,333	147.33	180.90	229.17	144.12	186.65
Ashana	296.611	201.33	248.97	254.600	188.00	221.30	242.30	227.97	235.14
SADC (Long	255.467	192.67	224.60	218.200	182.90	200.60	319.18	205.44	262.321
SADC (Togo	323.867	206.00	264.90	305.657	247.23	276.40	314.67	226.62	270.65
Ugandi	269.067	166.33	217.70	259.33	246.700	253.01	264.20	206.52	225.37
Sudan II	258.133	218.00	238.00	257.233	194.33	225.00	237.62	225.20	231.45
MCNELC	258.200	318.00	288.06	235.167	192.33	213.70	276.58	225.72	250.93
DembiShangal Toby	242.933	218.67	230.80	211.367	195.00	203.20	218.97	206.97	216.85
Dembi Kabkabia	276.900	172.00	224.45	254.067	178.33	216.20	227.72	213.03	220.38
Dembi Sea	168.967	147.567	202.00	218.83	210.42	214.65	168.96	126.16	147.56
Means	224.01	173.01	199.9	225.33	186.04	205.66	221.26	183.39	202.28
LSD S	20.11								
LSD G	112.45								
LSD S x G	98.07								

4.1.5 Days to 50% flowering

The means for days to 50% flowering was highly significant, in seasons (2012) and season 2013 (Appendices 1 and 2). Whereas, highly significant differences ($P \leq 0.01$) due to genotype and stress and interaction between genotypes and stress appendix (2 appendix (1),and the combined showed highly significant ($P \leq 0.01$) in season, and the interaction between all treatments Appendix (3). The means separation due to combined analysis regarded that the highest values (63) and (62) were shown by the genotype HSD10376 and HSD10312 and HSD713 and DembiShangal Toby in 7day, 21days water regime respectively, (Table, 5) where lowest value (45.0) and (45.5) in two water regime (7days 21days), consecutively, were regarded by the genotype HSD10293 and HSD10293 (Table, 5).

4.2 Yield and its components

4.2.1 Number of panicle \plant

The study showed thatnumber of panicle/plant in season (2012), was highly significant differences ($P \leq 0.01$) genotype, and none significant in stress and interaction (stress × genotype), Appendix (1), whereas the individual analyses in season (2013), revealed that number of panicle\plant was highly significant differences ($P \leq 0.01$) in genotype and none significant in stress and interaction between (stress x genotype) appendix (3)between (season x stress), and highly significant in interaction between (season x stress x genotype), Appendix (3). This character regarded highest values (30.83) and (19.83) were shown by the genotype HSD10319 and HSD7134for two watering (7days and 21days) consecutively, and lowest values (7.83) and (8.08) were showed by the genotypes HSD10312and Ugandi and Sudan 2 for the two watering (7days and 21days) respectively (Table 9)

Table 9. Effects of water stress and millet genotypes on days to 50% flowering during 2012 and 2013 seasons and combined analyses

Season	2012			2013			Combined(2012-2013		
Treatment	7 days	21 days	Mean	7 days	21 days	Mean	7 days	21 days	Mean
HSD7131	59.33	54.67	57.00	60.33	59.00	59.88	57.00	59.50	58.25
HSD7132	60.00	48.33	54.20	56.00	59.00	57.00	56.84	59.83	58.34
HSD7133	59.00	54.33	56.70	57.67	58.33	58.00	53.67	58.00	55.84
HSD7134	57.00	51.00	54.00	57.00	59.33	58.20	56.33	58.33	57.33
HSD7135	59.33	59.33	59.33	56.00	59.33	57.20	55.17	57.00	56.30
HSD10291	57.33	45.00	51.20	55.67	61.67	58.67	59.33	57.67	58.50
HSD10292	56.67	50.67	53.67	56.67	60.67	58.67	53.34	56.50	54.92
HSD10293	56.33	45.33	59.88	55.00	60.67	57.34	55.677	56.67	65.17
HSD10294	60.67	45.00	52.34	57.33	61.67	59.50	53.00	55.65	54.33
HSD10303	55.00	59.33	57.20	55.00	60.00	57.50	53.34	59.00	56.17
HSD10309	58.33	58.67	58.67	59.00	59.00	59.00	59.67	55.00	57.34
HSD10312	57.67	52.00	54.84	60.67	63.00	61.88	58.84	58.65	58.75
HSD10313	58.33	47.00	52.88	60.67	58.33	59.50	57.50	59.17	58.34
HSD10318	58.33	55.00	56.88	57.67	59.33	58.50	52.67	59.50	56.07
HSD10319	60.00	59.00	59.50	57.67	61.67	59.00	57.17	58.00	57.67
HSD10331	58.00	60.00	59.00	59.33	59.00	59.00	60.34	58.88	59.62
HSD10354	56.00	57.67	56.84	56.67	62.33	59.50	59.50	58.65	59.06
HSD10362	60.00	61.67	60.84	62.33	62.33	62.33	60.00	56.34	58.17
HSD10376	56.67	60.00	58.34	58.00	63.00	60.50	62.00	61.16	61.66
HSD10392	58.33	60.67	59.50	56.67	55.00	55.88	61.50	57.34	59.17
Ashana	48.33	50.00	59.20	55.00	55.00	55.00	57.84	57.50	57.77
SADC (Long	52.33	47.33	49.88	55.00	55.00	55.00	52.50	51.67	52.30
SADC (Togo	48.33	47.33	47.88	56.67	56.67	56.67	51.17	53.67	52.37
Ugandi	48.33	47.33	47.88	55.00	57.00	56.00	52.00	52.50	52.30
Sudan II	48.33	47.33	47.88	55.00	62.67	58.88	52.17	51.67	51.77
MCNELC	61.33	48.67	55.00	59.67	60.33	60.00	55.00	51.67	53.34
DembiShangal Toby	60.00	46.67	48.34	59.33	62.67	61.00	54.50	50.00	52.50
Dembi Kabkabia	57.67	48.33	53.00	57.00	61.67	59.34	54.67	59.67	57.17
Dembi Sea	59.00	48.33	53.,88	57.67	64. 7	60.20	55.00	57.28	56.14
Means	56.76	52.28	55.07	57.44	59.72	58.59	56.77	56.78	56.78
LSD S	3.15								
LSD G	3.83								
LSD S x G	5.14								

Table 10.Effects of water stress and millet genotypes on number of panicles per plant during 2012 and 2013 seasons and combined analyses

Season	2012			2013			Combined(2012-2013)		
Treatments	7 days	21 days	Mean	7 days	21 days	Mean	7 days	21 days	Mean
HSD7131	15.33	7.55	11.44	18.50	8.00	13.30	13.03	11.67	12.35
HSD7132	16.00	7.83	11.14	19.33	8.17	13.75	13.58	12.90	13.44
HSD7133	15.67	7.50	11.58	23.17	7.33	15.30	15.34	11.50	13.42
HSDS7134	19.00	6.33	12.66	24.07	6.50	15.30	15.20	12.75	14.00
HSD7135	15.33	7.83	11.58	24.83	5.00	14.50	16.33	10.17	13.25
HSD10291	17.17	5.83	11.50	19.17	8.83	14.00	12.50	13.00	12.75
HSD10292	16.00	7.33	11.35	27.40	9.50	18.45	17.34	12.75	15.48
HSD10293	14.00	6.500	10.25	25.73	6.33	16.30	16.53	10.17	13.35
HSD10294	15.07	*7.67*	11.37	14.73	8.33	11.50	11.20	11.70	11.45
HSD10303	17.33	12.33	614.88	24.00	9.5	16.75	18.60	13.42	16.10
HSD10309	14.67	4.67	9.67	20.17	6.33	13.25	12.40	17.84	15.02
HSD10312	7.83	6.200	7.00	14.67	8.33	11.50	10.44	8.08	9.27
HSD10313	11.83	10.17	11.00	15.67	11.33	13.50	12.92	11.58	12.25
HSD10318	9.00	7.00	8.00	15.33	9.00	12.20	11.16	9.00	10.80
HSD10319	12.23	8.83	11.53	30.83	7.17	19.00	19.83	9.70	13.75
HSD10331	19.67	8.20	13.91	16.17	11.67	13.92	12.17	15.67	13.92
HSD10354	11.17	10.17	10.67	21.33	8.17	15.00	15.75	9.67	12.71
HSD10362	10.73	7.33	9.03	16.00	6.83	12.41	11.16	8.78	9.00
HSD10376	14.07	10.50	14.28	16.00	6.83	11.41	12.75	10.45	11.60
HSD10392	14.57	9.50	12.35	11.67	7.50	9.58	10.58	11.04	10.30
Ashana	13.83	7.33	10.68	12.33	7.33	9.88	9.88	10.58	10.23
SADC (Long	10.17	8.17	9.17	10.50	9.00	9.75	10.33	8.59	9.46
SADC (Togo	14.00	11.33	12.66	12.33	11.83	12.80	11.88	12.92	12.40
Ugandi	15.33	8.00	11.16	11.33	9.00	10.16	9.65	12.17	10.01
Sudan II	19.57	7.50	13.85	12.17	7.83	10.00	9.65	13.70	11.75
MCNELC	20.83	7.50	14.16	17.33	12.00	14.66	12.94	16.46	14.70
DembiShangal Toby	13.00	10.50	11.75	13.67	7.33	10.50	12.85	10.17	11.51
Dembi Kabkabia	10.73	9.67	10.17	10.17	8.50	6.90	9.92	9.62	9.77
Dembi Sea	16.40	9.17	12.83	11.90	8.33	10.11	10.53	8.20	9.36
Means	14.50	8.22	11.13	17.60	8.34	12.95	12.22	11.53	12.19
LSD s	2.76								
LSD G	3.14								
Lsd G× s	4.87								

4.2.2 Panicle weight

Analysis of variance indicated that Panicle weight was highly significant ($P \leq 0.01$) affected by stress, genotype interaction between genotype in season (2012) Appendix (1), whereas, in season (2013) highly significant differences ($P \leq 0.01$) were shown by genotype and stress and interaction between genotype and stress Appendix (2), The individual analyses showed that combined analyses was only significant ($P \leq 0.05$) due to interaction between (stress x season), but highly significant ($P \leq 0.01$) by interaction between (season × stress × genotype), appendix (3).The mean separation registered by combined highest values (24.72g) and (23.7g) showed by the genotypes Dembi Shangal Toby and Dembi Millet in the two water regimes (7days and 21 days) in succession, and the lowest values (4.12 g) and (4 g) were reviled by the genotypes HSD10362 and HSD10312 in the two watering (7 days and 21 days), respectively (Table 11).

4.2.3 Number of grains/pancile

The analysis of variance showed highly significant differences ($P \leq 0.01$) were shown by the stress, genotype, and interaction (stress × genotype), Appendix (1). Whereas, in season (2013) significant differences ($P \leq 0.05$) showed by stress, and highly significant differences ($P \leq 0.01$) showed by the genotype and interaction (stress × genotype) Appendix (2). The individual analyses showed that no significant due to combined Appendix (3). Separation of means due to combined analysis reveled highest values (883.84) and (639.67) reveled by the Genotype Dembi Kabkabia and HSD10354, while the lowest was obtained by genotype. Ashana and HSD10309 and was (180.87) and (41), respectively (Table 12).

Table 11. Effects of water stress and millet genotypes on panicle weight (g) during 2012 and 2013 seasons and combined analyses

Season	2012			2013			Combined(2012-2013)		
Treatments	**7 days**	**21 days**	**Mean**	**7 days**	**21 days**	**Mean**	**7 days**	**21 days**	**Mean**
HSD7131	14.64	11.78	13.26	1057	8.57	8.57	11.75	10.20	10.98
HSD7132	14.322	9.25	11.88	13.00	13.00	13.00	13.66	11.15	12.40
HSD7133	8.024	8.61	88.32	17.10	16.10	17.10	12.55	11.35	11.95
HSD7134	17.24	7.30	11.80	15.10	13.10	15.10	21.50	12.17	16.21
HSD7135	25.55	4.50	15.00	16.40	14.40	15.40	22.00	10.45	16.22
HSD10291	13.60	6.88	10.20	10.50	9.50	9.50	11.80	8.38	9.88
HSD10292	10.58	8.56	9.60	23.40	22.40	23.40	16.70	15.93	16.32
HSD10293	6.977	8.00	7.50	14.97	12.97	13.97	13.00	12.00	12.50
HSD10294	17.5	9.29	13.40	15.43	13.43	14.43	16.50	12.36	14.43
HSD10303	12.42	11.42	11.76	19.47	17.47	18.47	15.94	15.00	15.47
HSD10309	8.66	8.26	7.96	6.83	4.83	5.83	7.61	7.24	7.43
HSD10312	10.06	4.20	7.10	11.27	9.27	10.27	10.63	7.72	9.17
HSD10313	6.546	7.07	6.71.88	13.60	11.60	12.60	10.30	10.00	10.15
HSD10318	11.39	5.73	9.50	13.47	11.47	12.47	12.25	9.55	11.90
HSD10319	17.24	8.75	13.49	11.30	10.30	10.80	14.37	10.00	12.18
HSD10331	11.10	5.41	8.20	13.57	12.51	13.04	12.33	8.50	10.44
HSD10354	7.66	5.12	6.44	17.93	15.91	16.92	13.28	11.50	12.39
HSD10362	10.06	4.12	7.90	14.83	12.83	13.83	17.44	9.47	13.45
HSD10376	6.54	7.02	6.78	17.27	15.29	16.28	12.95	11.13	12.40
HSD10392	11.39	6.24	8.81	9.27	7.29	8.28	9.33	6.75	8.04
Ashana	17.21	6.04	11.23	21.10	23.16	22.13	19.15	13.77	16.46
SADC (Long	18.07	17.50	17.78	13.36	11.38	12.37	15.77	15.40	15.58
SADC (Togo	10.68	10.57	10.61	14.80	12.80	13.80	12.74	12.65	12.69
Ugandi	6.64	10.00	8.32	28.70	24.70	26.70	19.67	17.35	18.60
Sudan II	9.66	9.438	9.50	28.57	26.57	27.57	19.13	19.00	19.06
MCNELC	18.33	8.68	13.20	29.80	26.80	27.80	19.60	18.26	18.93
Dembi Millet	26.35	6.55	16.45	11.07	9.07	10.07	23.71	9.86	16.79
DembiShangal Toby	13.75	8.33	11.20	42.70	38.70	40.70	27.72	24.00	25.86
Dembi Kabkabia	11.90	11.77	11.88	27.20	25.20	26.20	19.50	19.45	19.5
Dembi Sea	9.99	10.56	10.20	25.57	23.57	24.57	19.77	18.38	19.25
Means	12.80	8.23	13.42	17.61	15.81	16.71	15.76	12.63	14.22
LSD s	1.30								
LSD G	0.48								
LSD S x G	0.67								

Table 12. Effects of water stress and millet genotypes on number of grains per panicle during 2012 and 2013 seasons and combined analyses

Season	2012			2013			Combined (2012-2013)		
Treatments	**7 days**	**21 days**	**Mean**	**7 days**	**21 days**	**Mean**	**7 days**	**21 days**	**Mean**
HSD7131	444.00	206.33	468.50	325.19	444.00	206.33	253.29	325.19	289.34
HSD7132	658.00	263.00	611.67	460.50	658.00	263.00	77.8.83	460.50	614.40
HSD7133	520.67	382.00	625.67	451.34	520.67	382.00	786.33	451.34	618.84
HSD7134	735.00	278.33	555.17	506.67	735.00	278.33	433.10	506.67	469.89
HSD7135	305.67	260.67	433.67	283.17	305.67	260.67	710.50	283.17	406.84
HSD10291	196.200	303.33	639.67	249.77	196.200	303.33	557.45	249.77	403.61
HSD10292	165.400	442.67	401.84	304.04	165.400	442.67	800.42	304.04	552.23
HSD10293	441.33	514.00	453.67	477.67	441.33	514.00	694.34	477.67	586.0
HSD10294	*595.663*	219.00	627.24	407.33	*595.663*	219.00	621.17	307.33	514.25
HSD10303	684.67	339.33	180.87	512.00	684.67	339.33	814.67	512.00	663.34
HSD10309	660.00	175.00	391.84	41.00	660.00	175.00	784.50	413.00	598.75
HSD10312	712.00	225.00	591.00	468.50	712.00	225.00	354.10	468.50	411.30
HSD10313	848.33	375.33	502.00	611.67	848.33	375.33	435.84	611.67	532.76
HSD10318	718.33	533.00	437.72	625.67	718.33	533.00	418.17	555.17	521.92
HSD10319	633.00	477.33	500.33	555.17	633.00	477.33	445.60	433.67	500.39
HSD10331	603.33	264.00	397.65	433.67	603.33	264.00	511.00	639.67	472.34
HSD10354	804.33	475.00	361.00	639.67	804.33	475.00	705.33	401.84	672.47
HSD10362	596.67	207.00	402.09	401.84	596.67	207.00	718.84	453.67	560.34
HSD10376	518.67	388.67	468.50	453.67	518.67	388.67	427.50	627.24	440.59
HSD10392	747.67	506.80	611.67	627.24	747.67	506.80	315.67	627.24	471.46
Ashana	159.73	202	625.67	180.87	159.73	202	613.67	180.87	397.27
SADC (Long	424.67	359.00	555.17	391.84	424.67	359.00	398.34	391.84	395.09
SADC (Togo	649.00	533.00	433.67	591.00	649.00	533.00	531.17	591.00	561.09
Ugandi	433.33	571.00	639.67	502.00	433.33	571.00	645.00	502.00	573.50
Sudan II	236.43	639.00	401.84	437.72	236.43	639.00	582.44	437.72	510.08
MCNELC	812.33	188.33	453.67	500.33	812.33	188.33	692.32	500.33	592.33
DembiShangal Toby	667.3	182.00	627.24	397.65	667.3	182.00	327.72	397.65	362.96
Dembi Kabkabia	209.00	513.00	180.87	361.00	209.00	513.00	883.84	361.00	622.42
Dembi Sea	571.83	412.33	391.84	402.09	571.83	412.33	671.83	402.09	536.96
Means	543.19	359.84	481.77	434.49	543.19	359.84	576.22	443.93	512.16
LSD s	187.0								
LSD G	264								
LSD S×G	235								

4.2.5 1000 seed weight (g)

Analysis of variance indicated that thousand seed weight was highly significant ($P \leq 0.01$) affected by genotype and interaction between stressx genotype in season (2012) Appendix (1), whereas in season (2013), and highly significant differences ($P \leq 0.01$) were shown by genotype stress, and interaction between genotype xstress Appendix (2). The individual analyses showed that combined analyses was only significant ($P \leq 0.05$) due to interaction between (stress × season), but highly significant ($P \leq 0.01$) by interaction between (season × stress × genotype), Appendix (3).The mean separation registered by combined highest values (18.3g) and (28g) showed by the genotypes HSD1062 and MCNELC in the two water regimes (7days and 21days) in succession, and the lowest values (11.6) and (7.44g) were reviled by the genotype HSD10331 and HSD10376 in the two watering (7days and 21 days), respectively (Table 9).

4.2.6 Grain yield ton/ha

The means yield ton/ha was significantly affected by all treatments in both seasons, in season (2012) the analyses of variance showed highly significant differences ($P \leq 0.01$) were shown by the stress, genotype, and interaction (stress × genotype), Appendix (1). Whereas in season (2013) significant differences ($P \leq 0.05$) showed by stress, interaction (stress × genotype), and high significant differences ($P \leq 0.01$) only showed by the genotypes Appendix (2). The individual analyses showed that no significant due to combined Appendix (3). Separation of means due to combined analysis revealed highest values (2.3, 2.1, 2.07, 2.06, 2.01, 2.01, 2 ton/ha) and revealed by the genotypes HSD10294, HSD7133, HSD7135, HSD10303, HSD10319, Ashana, ugandi, sudan1, and the low yield was 1.06 ton/ha and gaind by genotype MCNELC. The yield was ranged (2.3-1.06) with mean 1.68 ton/ha.

Table 13. Effects of water stress and millet genotypes grains yield (ton/ha) during 2012 and 2013 seasons and combined analyses

Season	2012			2013			Combined(2012-2013)		
Treatments	**7 days**	**21 days**	**Mean**	**7 days**	**21 days**	**Mean**	**7 days**	**21 days**	**Mean**
HSD7131	1.51	1.50	1.60	1.50	1.30	1.60	1.52	1.47	1.56
HSD7132	1.10	120	1.65	1.40	0.93	1.90	1.42	1.40	1.43
HSD7133	2.01	1.42	1.42	2.00	1.43	2.52	1.41	1.41	2.77
HSD7134	1.90	2.00	2.00	1.93	2.20	1.70	1.82	1.82	1.86
HSD7135	2.40	2.32	2.32	1.91	2.40	1.45	2.27	2.27	1.48
HSD10291	1.80	1.72	1.73	2.10	1.92	2.30	1.54	1.54	1.45
HSD10292	1.64	1.50	1.50	2.20	1.90	2.44	1.10	1.10	1.22
HSD10293	1.92	1.82	1.82	2.50	2.20	2.80	1.73	1.73	1.24
HSD10294	2.01	2.33	2.33	2.10	1.40	2.94	*1.71*	1.71	2.04
HSD10303	2.01	2.60	2.60	3.30	2.60	4.03	1.11	1.11	1,62
HSD10309	1.60	1.71	1.71	2.50	2.81	2.15	0.61	0.61	0.73
HSD10312	1.80	1.80	1.80	2.54	2.80	2.31	0.84	0.84	1.29
HSD10313	1.61	1.10	1.10	2.40	1.50	3.30	0.74	0.74	0.93
HSD10318	1.80	1.41	1.41	2.20	1.70	2.64	1.14	1.14	1.60
HSD10319	2.30	2.50	2.50	2.83	2.50	3.20	2.51	2.51	2.90
HSD10331	1.70	1.92	1.92	2.60	2.10	3.04	0.81	0.81	0.85
HSD10354	1.82	1.44	1.44	2.70	1.70	3.61	1.20	1.20	0.78
HSD10362	1.55	1.51	1.51	2.41	2.34	2.48	0.70	0.67	0.70
HSD10376	120	1.31	1.31	1.72	1.70	2.80	1.00	1.00	1.04
HSD10392	2.00	1.01	1.60	2.31	2.40	2.30	0.83	0.83	1.05
Ashana	2.10	2.03	2.20	1.60	1.43	1.77	2.64	2.64	2.57
SADC (Long	1.40	1.40	2.00	1.60	1.90	2.22	1.32	0.90	1.80
SADC (Togo	180	1.60	2.03	2.30	2.00	2.60	1.60	1.20	2.11
Ugandi	2.91	1.90	2.13	2.74	2.80	2.70	1.30	1.00	1.60
Sudan II	2.10	2.00	2.14	1.83	2.30	1.40	2.64	1.70	3.60
MCNELC	1.10	0.80	1.90	1.45	1.54	1.34	2.02	1.60	2.43
DembiShangal Toby	180	1.64	1.82	2.40	2.30	2.53	1.10	1.00	1.10
Dembi Kabkabia	1.51	1.50	1.60	1.50	1.43	1.60	1.52	1.50	1.60
Dembi Sea	2.00	2.00	2.00	1.80	1.90	1.82	1.94	1.10	1.94
Means	1.81	1.69	1.83	2.15	1.98	2.40	1.45	1.33	1.63
LSD S	0.77								
LSD G	0.66								
LSD S×G	0.86								

4.3 Chemical analysis (Proximate composition analysis):

The Chemical analyses in this study Included moisture content%, Crude protein%, Crude fiber%, Crude fats%, ash content%, carbohydrates%, they were carried out according to the official methods described in [6].

B. The mineral contents:

The mineral contents (in mg/kg) in the seedsof the 30 pearl millet genotypes included Calcium (Ca), Phosphor (P), Iron (Fe), Potassium (K) Magnesium (Mg), Sodium (Na) and Zinc (Zn). These minerals were determined according to the official method of [6].

C. Physical properties:

The physical properties in this study included seed colour, granule (seed) size (mm), 100 seed weight (g) and Taste. The granules size of the 30 pearl millet seeds were recorded using Vernier Caliper.

Analysis of Variance for Physico-chemical showed that there were high statistically significant differences between the moisture, protein, Fiber, fats, Ash, and Carbohydrate according to M-stat programme Table 15.

The least significant difference test at 5% level showed that the local genotypes gave the highest level more than introduced genotypes. .

All The genotypes of ICRISAT and ARC gave the lowest level in #physico-chemical parameters.

Local genotypes are indicated the highest level of Protein Fiber, Ash, and Carbohydrate respectively.

ARC genotypes gave the highest level of Moisture, and Fats respectively~ table (4). This result disagree with the result by

Physical properties:

There was a statistically significant different it the 5% level in the 100 seed weight table (1).

Dembi Kabkabia and HSD10354 gave the highest weight in 100 seed weight table (3) while SADC (Togo) and HSD7132 gave the lowest weight which effected the Yield of millet

HSD10294, HSD10319, HSD10331 HSD10392, HSD55555, Ashana, SADC (Long), SADC (Togo), Ugandi Sudan II, Dembi Millet and Dembi Shangal

gave the most desirable taste, while HSD7131,HSD7132, HSD7133,HSD7134, HSD10292, HSD10293,HSD10313, HSD10318, HSD10354, HSD10376, Dembi Kabkabia,Dembi Sea, MCNELC

HSD10291and HSD10303 Gave the normal taste. Genotypes HSD10309, HSD7135and HSD10362 are off taste table (3).

Miniral Content:

There were high statistically significant differences among the mineral content in the millet genotypes table (2).

HSD10291 had the highest content of Ca and Mg, HSD10292 had the highest content of Na,Mg and Zn chart (), MCNELC,SADC LONG,Dembi kabkabia and Ugandi were the highest content of p, Znand Mg respectively chart (5). Shanglitoby and HSD76134 had the highest value of Fe,

While the genotype HSD10309 gave the lowest value in P,genotype Dembi Kabkabia gave the lowest value in Fe, genotype.HSD10294 gave the lowest value in K, genotype.HSD10318 gave the lowest value in Mg genotypes.HSD10318, HSD10303 Ashana and SADC (Togo) gave the lowest value in Na. genotype. Sudan II gave the lowest value in Zn table (16)

Table (15) Quality character chemical Properties(Proximate composition analysis) for the mean of the different genotype (2013)

variety	Protien	Carbo	Fiber	Moisture	Fats	Ash
1. HSD7131	10.517^{p}	6.8400	15.213^{jk}	68.240^{cd}	1.4833^{hi}	5700fg
2. HSD7132	67.913^{fg}	1.4600^{ij}	4.4867^{i}	11.353^{k}	6.9233^{cde}	15.120^{mn}
3. HSD7134	66.967^{l}	1.5533^{e}	4.5067^{hi}	11.973^{g}	6.8667^{ghi}	15.000^{Opq}
4. HSD7135	68.210^{cd}	1.6833^{ab}	4.5100^{hi}	10.507^{p}	6.9267^{cd}	15.090^{no}
5. HSD10291	67.967^{efg}	1.5067^{gh}	4.6000^{f}	11.007^{m}	6.6667^{jk}	14.920^{rs}
6. HSD10292	68.283^{bc}	1.6167^{c}	4.6533^{e}	10.297^{q}	6.8900 ^{efgh}	15.150^{lm}
7. HSD10293	67.440^{jk}	1.5500^{e}	4.5667^{fg}	11.317^{k}	6.8867^{fgh}	15.127L^{m} n
8. HSD10294	68.463^{ab}	1.7033^{a}	5.2100^{a}	9.630^{u}	6.8700^{gh}	14.993^{q}
9. HSD10303	67.640^{hij}	1.4833^{hi}	4.9533^{b}	10.867^{n}	6.8467^{i}	15.057^{no}
10. HSD10309	67.903^{fg}	1.5200^{fg}	4.4833^{i}	11.137^{l}	6.9400^{c}	14.957^{qr}
11. HSD10313	67.790^{ghi}	1.5167^{fg}	4.7867^{c}	10.657^{o}	6.8733 ^{ghi}	15.250^{ij}
12. HSD10318	66.160^{n}	1.7033^{a}	4.6600^{e}	12.303^{e}	6.8400^{i}	15.173^{kl}
13. HSD10319	66.970^{l}	1.7000^{a}	4.3167^{l}	11.970^{g}	6.8933^{defgh}	15.043^{op}
14. HSD10331	66.560^{m}	1.5333^{efg}	4.4933^{i}	11.817^{h}	6.9967^{b}	15.597^{ef}
15. HSD10354	67.820^{gh}	1.5367^{ef}	4.7700^{cd}	10.987^{m}	6.8633^{hi}	14.887^{s}
16. HSD10362	68.127^{cde}	1.5333^{efg}	4.7433d	10.013s	6.9900b	15.583fg
17. HSD10376	67.407^{k}	1.2533^{m}	4.6467^{e}	11.507^{j}	6.9433^{c}	15.000^{pq}
18. HSD10392	67.930^{efg}	1.4433^{jk}	4.5200^{hi}	10.517^{p}	6.9000 ^{defg}	15.587^{fg}
19. HSD55555	68.627^{a}	1.6667^{b}	4.5067^{hi}	10.103^{r}	7.0533^{a}	15.130^{lm}
20. Ashana	67.633^{hig}	1.5433^{ef}	4.5367^{gh}	11.717^{i}	6.6867^{j}	14.570^{u}
21. SADC (Long)	67.803^{ghi}	1.4633^{ig}	4.6467^{e}	10.543^{p}	6.6667^{jk}	15.543^{g}
22. SADC (Togo)	66.247^{n}	1.4833^{hij}	4.7767^{cd}	12.227^{f}	6.5900^{m}	15.267^{l}
23. Ugandi	65.597^{o}	1.3500^{l}	4.2367^{m}	12.927^{d}	6.8933 ^{defghg}	15.940^{c}

24. Sudan II	62.527^{q}	1.6567^{b}	4.3433^{kl}	15.370^{a}	6.6433^{kl}	16.103^{b}
25. MCNELC	66.290^{n}	1.4167^{k}	4.4033^{j}	13.243^{c}	6.8933^{defgh}	14.647^{t}
26. Dembi Millet	64.427^{p}	1.5300^{efg}	4.5133^{hl}	13.317^{b}	6.1633^{n}	16.213 ^{a}
27. DembiShangal Toby	67.603^{ijk}	1.6033^{cd}	4.5200^{hi}	10.633^{o}	6.1633^{n}	15.640^{e}
28. Dembi Kabkabia	68.553^{a}	1.4733^{i}	4.3733^{jk}	9.763^{t}	6.9167 ^{cdef}	15.747^{d}
29. Dembi Sea	68.063^{def}	1.5833^{d}	4.3633^{jk}	10.513^{p}	6.8733^{ghi}	15.477^{h}
30. HSD7133	66.900^{l}	1.4867^{hi}	4.6500^{e}	11.813^{h}	6.8733^{ghi}	15.150^{lm}

For each character (nutrient), different letters indicate means are significantly different (P <0.05).

Table (16) Quality character Physical Properties (minerals) for the mean of the different genotype (2013)

Variety	MeanCa	Mean P	MeanFe	Mean K	Mean Mg	MeanNa	Mean Zn
1. HSD7131	70.10^{b}	654.00^{j}	70.36^{q}	3525.00^{x}	1312.30^{defghij}	19.00^{o}	25.66^{h}
2. HSD7132	70.30^{b}	651.33^{jk}	69.93s	3608.00^{v}	1181.70^{hilk}	29.33^{ij}	50.33^{a}
3. HSD7133	74.00^{b}	624.67^{n}	80.53	3579.70^{w}	1358.00 ^{cdefghi}	40.00^{d}	26.66^{h}
4. HSD7134	72.00^{b}	652.00^{jk}	100.08^{b}	4517.30^{j}	1354.00^{cdefghi}	41.66^{bcd}	24.66^{hi}
5. HSD7135	72.40^{b}	653.33^{j}	80.6i	3812.30^{r}	1365.30^{cdefgh}	27.00^{jk}	30.66^{g}
6. HSD10291	2429.00^{a}	753.33^{h}	60.50^{x}	3763.00^{s}	1452.00^{abcde}	33.00^{gh}	24.33^{hi}
7. HSD10292	60.70^{b}	760.67^{g}	75.12^{n}	3471.00^{z}	1478.70 ^{abcd}	45.00^{a}	45.66^{bc}
8. HSD10293	74.50^{b}	853.33^{e}	80.10^{m}	3688.30^{t}	1253.70^{fghijk}	40.66^{cd}	39.33^{d}
9. HSD10294	49.60^{b}	649.33^{k}	71.42^{p}	3212.00^{a}	1364.30^{cdefgh}	44.00^{ab}	46.66^{b}
10. HSD10303	71.60^{b}	755.67^{h}	90.53^{g}	3661.70^{u}	1174.70^{ijk}	15.00^{p}	44.66^{bc}
11. HSD10309	85.10^{b}	529.33^{p}	90.83^{f}	4513.00^{j}	1523.00^{abc}	25.66^{kl}	32.00^{fg}
12. HSD10313	71.40^{b}	637.33^{m}	70.15^{r}	3614.00^{v}	1268.70 ^{EFGHIJK}	32.33^{gh}	36.66^{de}
13. HSD10318	91.50^{b}	749.00^{l}	65.68^{v}	4761.30^{g}	375.30^{l}	19.00^{o}	24.00^{hi}
14. HSD10319	84.10^{b}	651.00^{jk}	81.22^{k}	4470.70 ^{k}	1361.30^{cdefghi}	26.00^{kl}	34.33^{ef}
15. HSD10331	85.00^{b}	651.33^{jk}	60.85^{w}	4615.30^{i}	1118.30^{k}	20.33^{no}	36.00 ^{e}
16. HSD10354	83.50^{b}	551.67^{o}	66.58^{u}	4356.70^{i}	1346.30^{cdefghi}	15.00^{p}	50.66^{a}
17. HSD10362	92.30^{b}	764.33	84.79^{i}	4269.70^{n}	1416.70^{abcdef}	20.66^{no}	37.33^{de}
18. HSD10376	74.30^{b}	651.67^{jk}	81.14^{k}	3488.30 ^{y}	1588.30^{a}	22.66^{mn}	32.00^{fg}
19. HSD10392	83.10^{b}	643.67^{l}	73.13^{o}	4221.70^{o}	1144.70^{ijk}	40.66^{cd}	43.00^{c}
20. HSD55555	94.60^{b}	651.67^{jk}	68.86^{t}	5194.70^{c}	1481.30^{abcd}	42.66^{abc}	44.00^{bc}
21. Ashana	115.20^{b}	955.67^{b}	90.55^{g}	5264.30^{b}	1431.70^{abcdef}	15.00^{p}	45.00^{bc}
22. SADC (Long)	96.10^{b}	972.33^{a}	97.92^{c}	4876.30^{f}	1228.30^{ghijk}	44.66^{a}	25.33^{h}
23. SADC (Togo)	100.50^{b}	972.00^{a}	83.23^{j}	4981.70^{e}	1365.30^{cdefgh}	13.66^{p}	27.00^{h}
24. Ugandi	120.50^{b}	972.00^{a}	86.21^{h}	4631.30^{h}	1561.30^{ab}	18.66^{o}	30.66^{g}
25. Sudan II	95.30^{b}	873.67c	90.92^{ef}	4154.70^{p}	1449.30^{abcde}	40.66^{cd}	21.66^{i}
26. MCNELC	110.90^{b}	957.67b	91.02^{e}	5013.00^{d}	1243.70^{fghijk}	34.33^{fg}	45.66^{bc}
27. Dembi Millet	120.00^{b}	759.67g	91.50^{d}	5742.30^{k}	1384.70 ^{bcdefg}	37.00^{e}	39.33^{d}
28. DembiShangal Toby	94.10^{b}	873.67	100.82^{a}	4332.00^{m}	1102.70^{k}	31.00^{hi}	34.66^{ef}
29. Dembi Kabkabia	115.50^{b}	861.33^{d}	10.23^{z}	4215.70^{o}	1431.30^{abcdef}	35.66^{ef}	46.66^{b}
30. Dembi Sea	125.70^{b}	864.00^{d}	11.09^{y}	3952.3^{q}	1328.70^{defghij}	24.00^{lm}	32.66^{fg}

Mean with the same letter for parameters are not significant at 5% level (LSD

Table 14. Physical properties of the 30 pearl millet genotypes

Genotype	Colour	Granule size* (mm)	Weight of 100 seeds (g)	Taste**
1..HSD7131	Yellowish black	3.4x2.0x1.5	0.94	4
2.HSD7132	Dark yellow	3.2x2.0x1.0	0.56	4
3.HSD7133	Bright Yellow	3.4x2.2x1.0	1.23	4
4.HSD7134	Yellow	2.8x2.0x1.8	0.66	4
5.HSD7135	Yellow	3.0x2.0x0.8	0.56	1
6.HSD10291	Bright Yellow	3.2x2.0x1.4	1.19	3
7.HSD10292	Yellow	3.0x2.0x1.0	0.95	4
8.HSD10293	Dark yellow	3.0x1.8x1.2	0.93	4
9.HSD10294	Yellow	3.2x2.0x1.5	1.37	5
10.HSD10303	Dark yellow	3.0x2.0x1.0	0.94	3
11.HSD10309	Yellow	3.0x1.8x1.0	0.52	1
12.HSD10313	Black yellow	3.2x2.0x1.2	0.92	4
13.HSD10318	Dark yellow	3.0x1.8x1.0	0.68	4
14.HSD10319	Yellow	3.2x2.0x1.0	0.69	5
15.HSD10331	Dark yellow	3.4x2.0x1.2	1.02	5
16.HSD10354	Yellow	3.0x2.2x1.8	0.86	4
17.HSD10362	Yellow	3.0x2.0x1.2	0.48	1
18.HSD10376	Yellow	2.8x1.8x1.0	0.68	4
19.HSD10392	Brown	3.0x1.8x1.2	1.02	5
20.HSD55555	White	3.2x2.0x1.	0.76	5
21.Ashana	Greenish yellow	3.4x2.2x1.4	1.05	5
22.SADC (Long)	Bright yellow	3.2x2.0x1.2	0.71	5
23.SADC (Togo)	White	2.6x1.8x0.8	0.65	5
24.Ugandi	Greenish yellow	3.0x2.2x1.2	0.93	5
25.Sudan II	Greenish yellow	3.4x2.4x1.0	1.27	5
26.MCNELC	Yellowish black	3.2x2.2x1.2	0.82	4
27.Dembi Millet	Yellowish black	3.0x2.2x1.2	0.98	5
28.Dembi Shangal Toby	Yellowish brown	3.2x2.4x1.4	1.13	5
29.Dembi Kabkabia	Yellowish black	3.4x2.2x1.6	1.25	4
30.Dembi Sea	Bright yellow	3.4x2.2x1.4	0.70	4

* LengthXwidthXthickness
** 5: Desirable, 4-3: Normal, 2-1: bitterness(Off taste).

The 30 Millet genotypes were amplified using 10 different Operon RAPD primers. Eight of the 10 primers gave amplification products while the remaining two (OPA9 and OPK15) didn't. Of the eight primers, five (OPY15, OPK16, OPK17, OPL16 and OPL18) were reproducible; exceptions were OPY17, OPA11 and OPL17. A total of 753 fragments were detected for the 30 genotypes representing 51 different loci with 96.1% polymorphism. The most relative genotypes were HSD10313 and HSD10318 with 92% similarity; while the most distant were Dembi millet and Dembi Shangal Toby with similarity percentage of 17%. According to the similarity indices, the 30 genotypes were grouped into 11 clusters (Fig 1). Distribution pattern of all the genotypes into various clusters indicates the presence of considerable genetic divergence among the 30 pearl millet genotypes.

A dendrogram was constructed with the 30 varieties using Squared Euclidean distances (Table 4) by ST A TISTICA software to analyze the genetic distances. It indicated that all 30 millet varieties could be distinguished by RAPD markers. Cluster analysis indicated eleven main clusters at a similarity level of 17% (Fig. 2). Distribution pattern of all the genotypes into various clusters showed the presence of considerable genetic divergence among the genotypes.

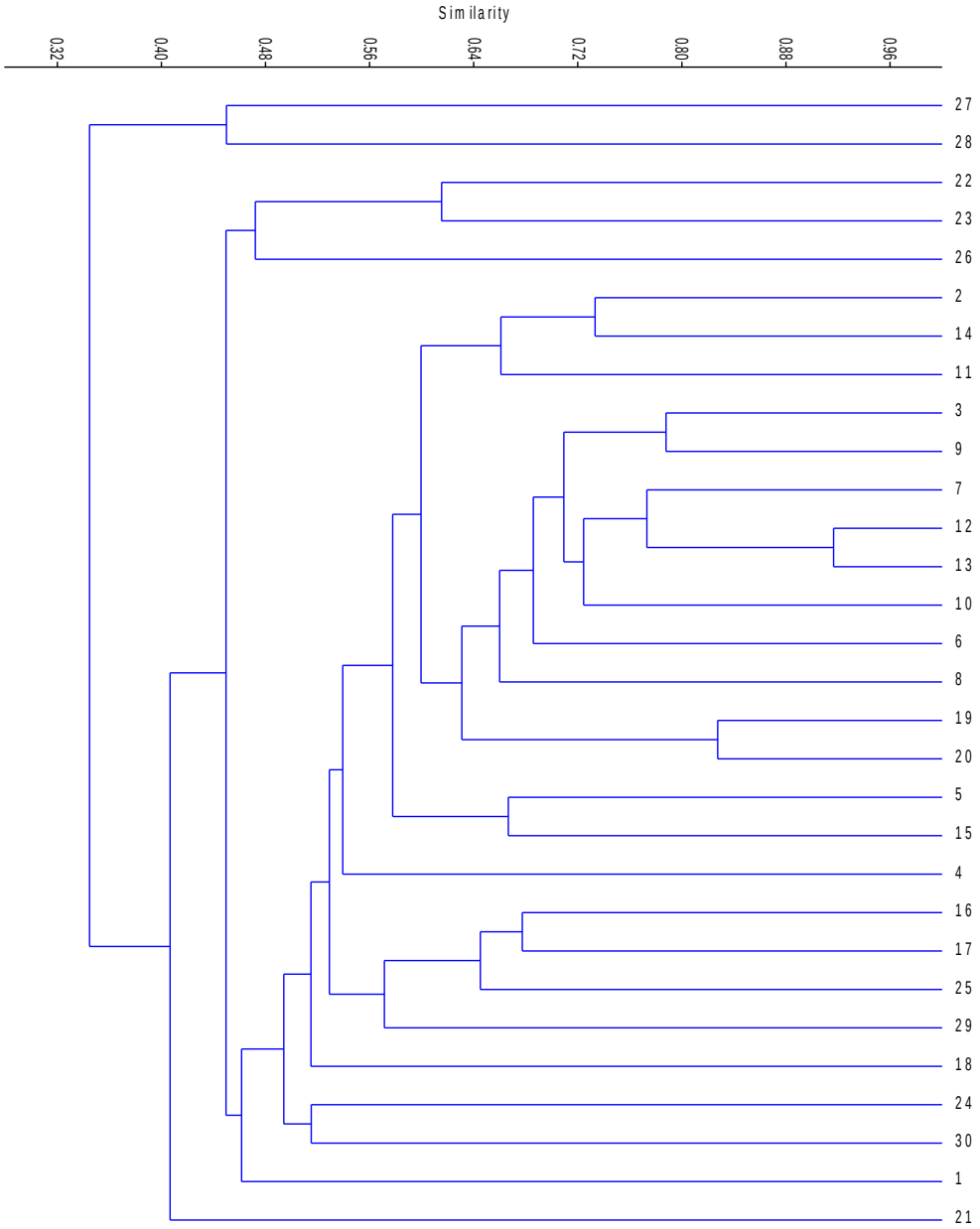

Fig.1. Dendrogram showing grouping of 30 varieties of millet from ICRISAT and Sudan based on the genetic distance derived from RAPD markers using UPGMA analysis

Table 18. Genetic distance estimated between 30 varieties of millet

	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20	21	22	23	24	25	26	27	28	29	30
1	1.00																													
2	0.47	1.00																												
3	0.54	0.73	1.00																											
4	0.38	0.52	0.69	1.00																										
5	0.47	0.56	0.68	0.42	1.00																									
6	0.54	0.63	0.73	0.54	0.68	1.00																								
7	0.46	0.63	0.73	0.54	0.54	0.68	1.00																							
8	0.45	0.55	0.71	0.55	0.46	0.53	0.66	1.00																						
9	0.40	0.68	0.79	0.63	0.58	0.69	0.64	0.61	1.00																					
10	0.46	0.69	0.74	0.50	0.64	0.65	0.69	0.67	0.60	1.00																				
11	0.45	0.66	0.62	0.41	0.60	0.57	0.57	0.53	0.56	0.63	1.00																			
12	0.45	0.66	0.76	0.57	0.53	0.67	0.76	0.74	0.71	0.72	0.60	1.00																		
13	0.53	0.65	0.79	0.53	0.56	0.70	0.79	0.72	0.70	0.76	0.55	0.92	1.00																	
14	0.53	0.73	0.64	0.44	0.53	0.59	0.55	0.47	0.54	0.60	0.67	0.58	0.62	1.00																
15	0.42	0.61	0.78	0.62	0.67	0.63	0.58	0.50	0.58	0.64	0.50	0.57	0.61	0.49	1.00															
16	0.50	0.42	0.61	0.55	0.42	0.53	0.66	0.58	0.47	0.53	0.44	0.59	0.68	0.39	0.59	1.00														
17	0.41	0.50	0.61	0.50	0.38	0.53	0.66	0.63	0.56	0.45	0.44	0.59	0.63	0.43	0.46	0.68	1.00													
18	0.45	0.55	0.53	0.37	0.50	0.61	0.53	0.53	0.51	0.62	0.53	0.69	0.63	0.51	0.38	0.44	0.49	1.00												
19	0.44	0.63	0.74	0.58	0.53	0.69	0.69	0.51	0.64	0.56	0.56	0.67	0.66	0.59	0.58	0.56	0.66	0.47	1.00											
20	0.41	0.65	0.71	0.55	0.50	0.61	0.71	0.49	0.61	0.57	0.53	0.64	0.63	0.61	0.59	0.53	0.63	0.49	0.83	1.00										
21	0.33	0.43	0.43	0.43	0.39	0.47	0.43	0.38	0.41	0.34	0.37	0.42	0.42	0.41	0.48	0.42	0.47	0.38	0.45	0.47	1.00									
22	0.39	0.48	0.51	0.39	0.39	0.43	0.47	0.34	0.41	0.47	0.38	0.46	0.50	0.45	0.48	0.47	0.47	0.42	0.45	0.57	0.50	1.00								
23	0.43	0.53	0.61	0.43	0.48	0.56	0.51	0.42	0.45	0.52	0.52	0.42	0.46	0.45	0.64	0.57	0.47	0.38	0.50	0.52	0.39	0.62	1.00							
24	0.41	0.46	0.57	0.37	0.55	0.57	0.57	0.44	0.61	0.49	0.53	0.55	0.59	0.51	0.46	0.49	0.41	0.49	0.47	0.49	0.38	0.42	0.52	1.00						
25	0.47	0.43	0.63	0.42	0.47	0.54	0.54	0.55	0.53	0.54	0.45	0.57	0.65	0.41	0.56	0.65	0.65	0.46	0.58	0.59	0.48	0.59	0.53	0.50	1.00					
26	0.38	0.26	0.47	0.43	0.34	0.35	0.52	0.52	0.41	0.42	0.28	0.42	0.46	0.25	0.39	0.57	0.47	0.33	0.36	0.42	0.33	0.50	0.44	0.42	0.48	1.00				
27	0.29	0.22	0.39	0.33	0.39	0.31	0.35	0.38	0.32	0.43	0.28	0.34	0.35	0.17	0.39	0.43	0.33	0.33	0.32	0.29	0.33	0.35	0.35	0.29	0.44	0.45	1.00			
28	0.37	0.29	0.42	0.32	0.33	0.34	0.31	0.28	0.35	0.29	0.36	0.33	0.38	0.31	0.33	0.46	0.37	0.32	0.45	0.32	0.27	0.44	0.44	0.37	0.48	0.38	0.45	1.00		
29	0.48	0.48	0.56	0.53	0.36	0.47	0.65	0.56	0.55	0.56	0.42	0.58	0.62	0.50	0.44	0.67	0.56	0.47	0.46	0.47	0.24	0.41	0.45	0.47	0.48	0.45	0.27	0.30	1.00	
30	0.48	0.40	0.47	0.39	0.48	0.56	0.65	0.43	0.46	0.43	0.42	0.50	0.58	0.46	0.40	0.56	0.52	0.43	0.50	0.52	0.35	0.36	0.41	0.52	0.48	0.45	0.23	0.30	0.55	1.00

CHAPTER FIVE

DISCUSSION

5.1 Variability due to water stress on growth and yield characters

5.1.1 Water stress effect on growth characters

Most of the growth characters were sensitive to water stress, plant height, leaf area, stem diameter, number of leaves, 50% days to flowering. Moreover, water stress was highly significant and reduced plant height in the two seasons among all genotypes. Similar finding were shown by Rauf (2008); Khayatnezhad *et al.* (2010) who found that effect of stress coincided with various growth stages such as germination; seedling; shoot length; and flowering. On the other hand stem diameter, leaf area and number of leaves also were highly significant and decrease due to stress, generally all of this characters were highest in (7days) watering and lowest in (21days) watering.

5.1.2 Water stress effect on yield and yield components

Drought had highly significant effect on yield and yield component of all the thirty genotypes of millet used in this study. Yield ton/ha showed high value in (7 days) 1.37 ton/ha in both season among all genotypes. Whereas, (21days) regime revealed small value 1.36 among all genotypes. Similar results showed by Al-karaki, and Clark, 1998, who found that sorghum (as another cereal crop) differed in their responses to deficit irrigation. Under full irrigation millet yields was good. However, irrigation deficit reduced growth character and yield in millet, giving higher yields for millet under moderate or severe water deficit treatments. Under water limited conditions; soil water extraction was more important component in millet yield. Thousand seed weight as one of the yield component was affected by drought stress (7days) watering register 10.63g which was high than (21days) value10.5g. The

reduction of thousand seed weight due to drought stress was reported by ELdikhary, 1992 and Osmanzai, 1992. Grain yield ton/ha was highly significantly affected by drought stress and high values were reported by HSD7133, HSD10354, HSD10318, HSD10313, Ashana, Sudan11, Ugandi in (7days) were 2.6, 2.19, 2.17, 2.13, 2.09 ton/ha compared with (21days) reported by HSD10303, HSD10319, HSD10294, HSD7135, Ashanaand Sudan11 and were gained 2.57, 2.49, 2.32, 2.33, 2.03 and 2.00) ton/ha respectively This result matched the one reported by Vanderlip 1991. In this study HSD7135 HSD10319 HSD10303 HSD7133 HSD10292, Ashana, Ugandi, Sudan11scored high yield under stress condition and could be used in drought tolerance breeding program.

5.1.3 Phenotypic and genotypicVariability

The results of this study revealed variability for most of the traits of the thirty pearl millet genotypes under study normal and water stress condition variation can be attributed to phenotypic as well as genotypic variability. Similar conclusions were detected by others in millet under different environments Khalafalla, 1993 and Abuelgusim, 1989. Variability in millet genotypes was reported by many investigators Abuali et al 2012; Subi, 2012; Subi et al , 2013.

5.2 Molecular characterization using RAPD Markers

The results of genetic diversity by using RAPD Markers of this study revealed that the most relative genotypes in the studied thirty pearl millet genotypes were HSD10313 and HSD10318 with 92% similarity; while the most distant were Dembi millet and Dembi Shangal Toby with similarity percentage of 17% and the 30 genotypes were grouped into 11 clusters according to this similarity indices. Distribution pattern of all the genotypes into various clusters indicates the presence of considerable genetic divergence

among the 30 pearl millet genotypes.Characterization of diversity has long been based mainly on morphological traits. Molecular tools provide valuable data on diversity through their ability to detect variation at the DNA level and a number of different techniques are available for identifying genetic differences between organisms Somasundaram and Kalaiselvam (2011).The high level of polymorphism (96%) presented in this study is in accordance to the results of Kaleand Munjal (2005). and Jaya Prakash et al, (2006). The dendrogram obtained clearly demonstrates that genetic diversity exists among the 30 studied genotypes. Similarly, Govindarajet al (2009) reported genetic diversity analysis in some pearl millet accessions using molecular markers. The results from Sudan and ICRISAT depict a clear separation between improved cultivars and gene bank accessions. The clustering of ICRISAT genotypes in the same main cluster reflects pedigree relationship as well as geographical origin (ICRISAT). Results of this study will help the use of genotypes from different clustering groups for any breeding program aiming to develop suitable varieties or hybrids with specific characters. Selections based on genotypes identified from genetic diversity studies using molecular markers will greatly increase breeding efficiency Irada and Samira (2010). The clustering of gene bank accessions (Sudan collection) together is an evidence for geographical origin, as these accessions were collected from millet grown areas in the Sudan, there is a chance of gene flow. Concordant with our results, Matsuoka *et al. (*2002) found that dendograms based on RAPDs in *Zea mays* were in good agreement to expect genetic relationships based on pedigree information. Similar results were found by Ordon *et al. (*1997) analyzing genetic Cluster because they came from the same origin. These results suggested that the dendrogram based on the estimated genetic similarity reflects morphological relationship as reported by Hormaza, (2002) on apricot (*Prunus armeniaca L.)* accessions. Yana *et al. (*2000) reported a weak differentiation of Ethiopian and Eritrian sorghum according to common

agro-ecological adaptation zones and regions of origin. Characterization of diversity has long been based mainly on morphological traits. Molecular tools provide valuable data on diversity through their ability to detect variation at the DNA level and a number of different techniques are available for identifying genetic differences between organisms Somasundaram, and Kalaiselvam,(2010). The high level of polymorphism (96%) presented in this study is in accordance to the results of Kale and Munjal, 2005 and Jaya *et al.*,(2006). The dendrogram obtained clearly demonstrates that genetic diversity exists among the 30 studied genotypes. Similarly, Govindaraj*et al*., (2009) reported genetic diversity analysis in some pearl millet accessions using molecular markers. The results from Sudan and ICRISAT depict a clear separation between improved cultivars and gene bank accessions. The clustering of ICRISAT genotypes in the same main cluster reflects pedigree relationship as well as geographical origin (ICRISAT). The clustering of gene bank accessions (Sudan collection) together in the same cluster gives an evidence for geographical origin, as these accessions were collected from millet grown areas in the Sudan. Results of this study will help the use of genotypes from different clustering groups for any breeding program aiming to develop suitable varieties or hybrids with specific characters. Selections based on genotypes identified from genetic diversity studies using molecular markers will greatly increase breeding efficiency Irada and Samira, 2010.

5.3 Quality traits

Variability between millet genotypes in quality traits could also be attributed to the effect of different environmental factors as reported by Irada and Samira, (2010), StatSoft, (2003) and Yu, *et al*., (1996). Variability between millet genotypes in quality characters was reported by many investigators, Jaccard, (1908),Duncan, 1955, Gomez, (1984), Salih, (2005), Sawada, (1993).

5.3.1Chemical composition of the 30 pearl millet genotypes

5.3.1.1 Protein content

The means of protein percentage in the 30 pearl millets ranged from14.57% to 16.21%, obtained by the genotypes Ashana and Dembi millet, respectively. These results reflect the high percentage of protein in these genotypes and indicate the high nutritive value of them. Similar findings were also revealed by CIMMYT, (2005), AOAD, (2008) and Obilana, (2013) in their studies in different millet genotypes. The Bibliography Govindaraj,*et al*., (2009) found that millet has a higher protein content (8.8 to 20.9%) than other cereals grown under similar conditions. The results of Nwasike, (1979) and Promeranz, (1980) indicated that millet protein was similar to corn (*Zea mays* L.) rather than that of grain sorghum in the distribution and lycine content. In general, millet is low in lysine, tryptophan, threonine and sulfur containing amino acids, than other cereal crops Promeranz, (1980) and Nambiar, *et al*(2011). Recently, high nutritive value and several health benefits of millet grains were reported by many authors. Johnson, *et al*(1955), Nambiar, *et* al (2011).,Somasundaram, and Kalaiselvam, (2011), Sarwar, *et al*., (2013) and Thapliyal and Singh, (2015).

5.3.1.2Carbohydrates and Fiber contents

The means of carbohydrates contents in the 30 pearl millet genotypes ranged from 62.53 to 68.63 obtained by the genotypes Sudan II and HSD55555, respectively. The means of the fiber contents ranged from 9.63 to 15.37 obtained by the genotypes HSD10294 and Sudan II, respectively. The high levels of carbohydrates and fiber obtained in the studied 30 grain millet genotypes could be of a great nutritive value and human health benefits. These results agreed with Jaya, *et al*., (2006) who found that the fiber content of pearl millet was12-13 % and it helps the body to get rid of stomach fats, supply the human body with sufficient energy and protect it from heart attack. The Bibliography Thapliyal and Singh, (2015)reported that millet contains

(18%) dietary fiber. In addition Sarwar, *et al*., (2013) reported that, millet is considered as a great source of starch, carbohydrates and fibers making it a high-energy food.

5.3.1.3 Moisture content, Fats content and Ash content:

In the 30 millet genotypes, the moisture content means ranged from 6.16 obtained for Dembi Millet to 7.05 obtained by the genotypes HSD55555 and Dembi Shangal Toby. Fats content means ranged from 4.24 to 5.21 obtained by the genotypes Ugandi and HSD10294, respectively. Ash content means ranged from 1.25 obtained for HSD10376 to 1.70 obtained for HSD10294 and HSD10318, respectively. Similar findings for Ash and Fats in different pearl millet genotypes were reported by Gomez,(1984) and AOAD,(2008). Nambiar, *et al*.,(2011) reported that the energy of millet is greater than sorghum and nearly equal to that of brown rice. In addition Subi, and Idris, (2013) reported that millet exceeded wheat, brown rice, maize and sorghum in total fats.

5.3.2Minerals contents:

The Ca ranged from 60.7 to 125.7 obtained by HSD10292 and Dembi Sea, respectively. The P ranged from 529.33 to 972.33 obtained by HSD10309 and SADC (Long), respectively. The Fe ranged from 60.50 to 100.82 obtained by HSD10291and Dembi Shangal Toby, respectively. The K ranged from 3212.0 to 5742.3 obtained by HSD10294 and Dembi Millet, respectively. The Mg ranged from 375.3 to 1588.3 obtained by HSD10309 and HSD10293, respectively. The Na ranged from 13.67 to 45.00 obtained by SADC (Togo) and HSD10292, respectively. The Zn ranged from 21.68 to 50.68 obtained by Sudan II and HSD10354, respectively. These results illustrate that three millet genotypes from Darfur states (Dembi Sea, Dembi Shangal Toby and Dembi Millet) scored the highest values of Ca, P and K, respectively. In addition, the

released genotype SADC (Long) scored the highest value of P, these genotypes could be of a great nutritive value for the consumers of millet at the productive areas of Darfur and Kordofan states, Sudan. Similar findings for Ca, Mg, Fe, Na, and K were observed in eight finger millet genotypes as explained by (Shashi, *et al*., 2007)Shashi, *et al* (2007). The Bibliography (AOAD, 2008) and (Somasundaram and Kalaiselvam, 2011) found that millet is rich of macro-minerals specially P and trace elements specially Fe. Many investigators e.g. (McKevith, 2004) explained that pearl millet exceeded wheat, sorghum and rice in total contents of minerals, fibers and calcium.

5.3.3.Physical properties of the 30 millet genotype:

5.3.3.1Granule size

The granule size of the 30 pearl millet genotypes ranged between 3.7 to 11.9 obtainedby SADAC Togo and Dembi kabkabia, respectively. In addition to millet granules high nutritive value for human and animals, small millet granules could be of a benefit for birds feeding especially ornamental birds. The variation in millet granule size was studied by many authors, e.g. (Subi, 2012) and (McDonough, *et al.*, 1986)

5.3.3.2Grain colour of the 30 millet genotypes

Among the 30 millet genotypes, 9 genotypes shown yellow grain colour, 5 genotypes shown dark yellow grain colour, 4 genotypes shown yellowish black grain colour, 4 genotypes shown bright yellow grain colour, 3 genotypes shown greenish yellow grain colour, 2 genotypes shown white grain colour and the three remaining genotypes shown brown, black yellow and yellowish brown grain colour. The Bibliography (Subi, 2012) indicated the yellow colour is favourite for the millet farmers and consumers in the Sudan. In India (PCU, 2014) explained that various colours were existed in different millet types, e.g. light brown, brown copper and purple colours in

finger millet; brown and golden colours in Kodo millet; little grey, dark grey, brown and dark brown colours in little millet.

5.3.3.3Weight of 100 seeds (gm)

The weight of 100 seeds of the 30 millet genotypes ranged from 0.48 to 1.36 gm obtained by the genotypes HSD10362 and HSD10294, respectively. Among yield components of pearl millet, 100 seed weight is considered as the most important yield component (Abuelgasim, 1999) and (Subi, 2012). Similar findings in different pearl millet genotypes were reported by (Subi and Idris, 2013).

5.3.3.4Taste estimation

The taste estimation of the 30 millet genotypes seeds was divided in this study to three ranges of numbers: 5 represent desirable taste, 3-4 represent normal taste and 1-2 represent bitterness taste. 12 millet genotypes included 7 local, released and improved varieties exhibited desirable taste, 15 millet genotypes exhibited normal taste and the remaining 3 millet genotypes exhibited bitterness taste. In the Sudan, the consumption of millet depends principally on its taste but few studies were conducted to estimate the taste of pearl millet genotypes (Subi, 2012).

CHAPTER SIX

CONCLUSIONS and recommendations

Based on the results obtained from this study, it could be concluded that:

1. Significant and considerable amount variability was detected among the 30 Sudanese pearl millet genotypes for growth, yield chemical compositions, mineral contents and physical properties, this variability would be useful in any millet breeding program aiming for improving growth, yield andquality traits in millet.

2. High level of polymorphism among genotypes was detected by using RAPD markers, indicating that, the technique was efficient in determining the genetic diversity among the thirty pearl millet genotypes, and thus could be exploited further to establish consistent heterotic groups between pearl millet genotypes.

3- The millet genotypes scored high values of yieldquality traits couldbeused by Sudanese millet consumers and/or millet breeders by selection orhybridization in order to produce improved millet genotypes or hybrids characterized with high and good yield quality traits.

Based on the results obtained from this study, it could be recommended that:

1- variety HSD7135, HSD10319, HSD10303, HSD7133, HSD10292, Ashana, Ugandiand Sudan11scored high yield under stress condition and could be used in drought tolerance breeding program.

2- Variety Dembi millet scored high protein ,V.HSD55555 scoredhigh Carbohydrates, V. Sudan II scored high Fiber,V. HSD55555 and V. Dembi Shangal Toby scored highMoisture,V.HSD10294 scoredhighFats,HSD10294 and HSD10318scored highAsh and could be used in quality improved breeding program.

6- Variety Dembi Sea scored High Ca, Variety SADC Long scored High P ,Dembi Shangal Toby scored High Fe,Dembi Millet scored High K, HSD10293scored High Mg, HSD10354 scored High Zn,HSD10292 scored High Na.

7- The Similarity indices were calculated using Jaccard's coefficient.- The most relative lines were .HSD10318and HSD10313 with 92% similarity; while the most distant were Dembi Millet and HSD10319 with similarity percentage of 17%.

According to the similarity indices, the 30 lines were grouped into 11 clusters.

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