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PHENOTYPIC AND PHYSIOLOGICAL ASPECTS RELATED TO DROUGHT TOLERANCE IN SORGHUM

J. MUTISYA, J.K. SITIENEY and S.T. GICHUKI
Kenya Agricultural Research Institute, Biotechnology Centre, P. O. Box 14733 00800, Nairobi, Kenya
Corresponding author: jmjoel2002@yahoo.com

ABSTRACT

Drought is one of the major limitations to crop productivity worldwide. Identifying suitable screening tools and quantifiable traits would facilitate the crop improvement process for drought tolerance in sorghum. This study evaluated phenotypic characteristics and physiological parameters determine which cultivars are more drought tolerant. Signs of drought intolerance in sorghum include leaf rolling, death of lower leaves, stunted growth and low yields. Experiments were conducted using 8 and 25 sorghum accessions planted at two sites in Kenya, namely; Biotechnology Centre and Kiboko Research site, respectively, for evaluation and seeds maintenance. Based on phenotypic characteristic of the 25 cultivars evaluated, the best drought tolerant cultivars were, IS.13615, KAK1950, KBM078, E-36.1, B-35, KBM-003 and IE SV 92036. These observations were specifically deduced from their performance, root characteristics, tillering ability and leaf parameters as drought tolerance indicators. B 35 and E-36 ranked the highest relative water content in leaves, hence more drought tolerant.

Key Words: Pre-anthsis, post-anthesis, *Sorghum bicolor*

RÉSUMÉ

La sécheresse est l'une des contraintes majeures à la productivité des cultures dans le monde. L'identification des outils d'étude et des caractères quantifiables pourrait faciliter le processus d'amélioration de la culture de sorgho pour la tolérance à la sécheresse. Cette étude avait pour but l'évaluation des caractéristiques phénotypiques et des paramètres physiologiques pour déterminer les cultivars les plus tolérants à la sécheresse. Les signes de l'intolérance à la sécheresse englobent l'enroulement de la feuille, la mort des feuilles les plus basses, croissance rabougrie et réduction de rendement. Les essais étaient conduits en utilisant 8 et 25 accessions de sorgho plantées dans deux sites au Kenya, à savoir le centre de Biotechnologie et le site de recherche de kiboko, respectivement, pour l'évaluation et la maintenance des semences. Basé sur les caractéristiques phénotypiques de 25 cultivars évalués, les meilleurs cultivars en terme de tolérance à la sécheresse étaient*:* IS.13615, KAK1950, KBM078, E-36.1, B-35, KBM-003 et IE SV 92036. Ces observations étaient spécifiquement déduites de leur performance, caractéristiques des raciness, la capacité de tallage et les paramètres des feuilles comme indicateurs de tolérance à la sécheresse. B 35 et E-36 avaient une teneur relative plus élevée en eau et par conséquent les plus tolérants à la sécheresse.

Mots Cles: Pré-anthésis, post-anthésis, *Sorghum bicolor*

INTRODUCTION

Sorghum (*Sorghum bicolor* (L.) Moench) like rice, wheat, maize, rye, barley, oat and millets is a grass belonging to the *Poaceae* family and is classified as an important agricultural and economic cereal (Buchanan *et al.,* 2005). It is ranked fifth most planted cereal crop in the world (Zhao, 2007), and an important staple food crop in many parts of Africa, Asia and the semi-arid tropics worldwide (Oria *et al.,* 1995; Duodu *et al.,* 2003; O'Kennedy *et al.,* 2006). In Africa, it is an indigenous cereal adapted to semi-arid and sub-tropical agronomic conditions, representing the only viable food grain (Zhao, 2007; Belton and Taylor, 2004). Consumers of sorghum-based diets depend on the available protein and energy from the grain (Oria *et al.*, 1995) thus, is one of the most versatile crops in terms of its utility. It continues to be an important food grain for farmers in the dry regions of the semi-arid topics (Rai *et al*., 1999).

In the Eastern Africa, sorghum is the second most important cereal crop after maize. In this region, it is grown on approximately of 7 million hectares per year (FAO, 2010). It is mostly cultivated in the semi-arid and arid areas that span from Northern Ethiopia, through North-eastern Kenya, Northern Uganda, and Central and Southern Tanzania.

Drought contributes heavily to the constant food insecurity and rampant poverty characteristic of these zones. Drought stressed plants produce inferior grain, low yields or no grain yield at all. Evolution of sorghum under pressures of drought has resulted in favourable physiological properties of the crop such as metabolic suppression and structural adjustment.

Water stress is known to alter a variety of phenotypic and physiological processes in crops. However, there is limited knowledge on the extent of genotypic adaptation to drought among sorghum cultivars in relation to yield and grain quality. Stay green is one of the traits largely associated with drought tolerance in sorghum. This trait is the ability of the plant to retain greenness during grain ripening under water limited conditions (Walulu *et al*., 1994; Borrell *et al*., 2000; Xu *et al.*, 2000). This trait is also reported to be associated with increased cytokinin concentration (McBee, 1984). This phenomenon enables the plant to exhibit drought tolerance and resistance to stalk lodging (Woodfin *et al*., 1998) and charcoal rot (Rosenow, 1983). Other traits related to drought tolerance in sorghum include early maturity and increased root density. Attempts to exploit these genetic variation for drought tolerance in sorghum through conventional plant breeding methods have been slow and arduous.

Thus, understanding and characterising the traits associated with drought in sorghum forms a major prerequisite for the development of a wide range of varieties as a feasible solution to climate change adaptation strategy.

This study aimed at carrying out phenotypic and physiological measurements of sorghum germplasm from different agro-ecosystems to establish the genetic factors associated with drought tolerance. The objective of this study was therefore; to determine phenotypic and physiological parameters exhibited by drought tolerant sorghum..

MATERIALS AND METHODS

Plant materials. Sorghum plants were grown in the growing seasons of 2007 and 2008 at the Biotechnology Centre and Kiboko Research Station. In these two sites, 25 sorghum accessions were planted at a spacing of 30 cm by 100 cm. A selection of other 12 varieties was grown in the greenhouse as described elsewhere (Mutisya *et al.*, 2003). Fertiliser application, watering and weeding were done as described by Borrell *et al*. (2000).

Phenotypic measurements in sorghum. We examined 10 phenotypic characteristics in selected sorghum accessions to establish which cultivars showed more drought tolerance. These were: green colouration, disease levels, tillering ability, plant size, leaf rolling, leaf drying, root weight, root length, root thickness and yield. On green colouration, disease levels and tillering ability we used a scale of 1 to 5 representing poor to best drought tolerance. Some of the measurements were repeated in both locations to ascertain the results.

Plant physiological measurements. This study was conducted at National Research Laboratories at Kabete in Nairobi, Kenya. Leaf disks, 1.3 cm in diameter, of the 8 varieties were collected with a cock borer during the 2007 and 2008 growing seasons. These varieties were Othuwa, B-35, E-1291, Is- 21146, Is-33461, B-36, Ochuti, and KAK-7801. Forty leave disks per plant were collected, immediately sealed in glass vials and transported to the laboratory in an ice-cooled box to determine leave relative water content (RWC) following the method of Martin *et al*. (1989). fresh weights of the disks were weighed within 2 hours after excision. The turgid weights were obtained by rehydration in deionised water for 24 hours at room temperature. After re-hydration, leaves were left for 48 hours to dry at room temperature, whereby leaves were quickly and carefully blotted dry with tissue paper before determining turgid weight. Dry weights were determined after 48 hours.

A similar study was conducted on plants in the experimental fields at Kiboko and Biotechnology Centre to measure relative water content (RWC) accumulated on the leaves. Initial measurements were taken in February 2008 on plants grown at Biotechnology Centre. Between 8 and 25 sorghum accessions were analysed for water content. A 20 leaf disks sample was collected using a leaf punch from a leaf from each plant.

Each leaf disk was approximately 1.3 cm in diameter. The samples were weighed immediately within 2 hours after excision and either socked in water for 24 hours or dried for 3 days before weighing to determine water loss. Data were subjected to a T-test or analysis of variance as appropriate.

RESULTS AND DISCUSSIONS

Phenotypic data. Ten leaf, root and yield phenotypic characteristics in selected sorghum accessions were examined to establish which cultivars showed more drought tolerance than others. Data taken on leaf parameters indicated that B35, E36-1, Livowya, macia, KAK 1950, KAK 7801 and KAK 7837 had the highest leaf retention level (Table1). Among those cultivars, the most drought tolerant varieties were KAK1950, E-36.1 and B-35 based on combination of phenotypic factors measured (Table 1).

Based on root weight and length, B35, E36-1, KBM003, KBM097, KAK1950, IESV92036 and IS8193 showed highest root length and weight implying that they are more drought tolerant

TABLE 1. Leaf and yield characteristics of 25 sorghum at Kiboko in Kenya

Cultivar	Score on stay green(1-5)	Level of disease resistance (1-5)	Tillering ability (1-5)	Plant size (S,M,T)	Yield levels (H,M,L)
Macia	4	2	3	s	M
Gopan	2	5	3	m	M
Gataraga	1	3	1	m	M
Sudan	3	2	1	m	M
Othuwa	3	4	3	s	L
Livowya	4	3	2	t	M
Essuti	3	3	2	t	M
IS 33461	2	3	2	s	H
B-35	5	3	5	s	S
E-36-1	4	4	3	m	M
KARI Mtama1	3	3	2	m	M
Ochuti	3	1	2	m	L
KBM 003	3	1	2	t	L
KBM 022	2	4	2	t	L
KBM 078	3	5	5	m	M
KBM 097	3	3	3	m	M
KAK 1950	4	4	5	m	M
KAK 7801	4	2	2	s	L
KAK 7837	4	3	4	m	M
(12X46)-1	3	2	3	s	L
IS 21055	4	3	3	t	M
IESV 92036	3	3	2	t	M
IS 8193	3	2	4	s	L
IS 21146	4	3	4	m	L
IS 13615	5	4	3	m	M

1 – 5 increase from poor to best, s-small, m-medium, t-tall, l-low, h-high

among 25 accessions (Table 2). A recent study has shown that high root weight and length are associated with drought tolerance (Wataru *et al*., 2005) implying maximisation of water absorption by tolerant sorghum cultivars. However, the above cultivars were not necessarily high in yields. The lack of direct correlation between yield and drought tolerance has previously been reported (Wright and Smith, 1983).

Tillering ability is commonly associated with plants such as sorghum that grow in regions with limited rainfall. We observed variations in tillering ability among the cultivars (Table 1 and 3); however, the occurrence was not consistent within cultivars at different sampling periods. Studies done by Lafarge *et al*. (2002) could not associate tillers with either yield of drought tolerance. However, it is likely that emergence of tillers is genetically controlled and partly serve as a survival mechanism in stress conditions.

In this study, the variation in phenotypic data observed was expected because some cultivars could have other mechanisms like drought escape as the major strategy in tolerating water stress. Visual rating scale has been used to evaluate stay green characteristics in sorghum. Normally, there is a linear relationship between green leaf area retention and drought tolerance (Wanous *et al*., 1991).

Physiological measurements related to drought. Measurements of relative water content taken on plants grown at the Biotechnology Centre and Kiboko were analysed to establish variations in water retention, after they were subjected to drought conditions. RWC ranged from 0.0535 to 0.0886 g on leaf disks collected from different plants. The highest and the lowest leaf dry weights measured were 0.04 and 0.03 g. Thus, B 35 and E-36 had the highest RWC, while KAK

TABLE 2. Mean measurements on leaf, root and yield data collected at Kiboko on second ratoon crop

Cultivar	Leaf rolling score	Leaf drying score	Root thickness (mm)	Root length (cm)	Root weight (g)	Shoot weight (g)	Shoot: root weight ratio	Yield weight (g)
Macia	2	3	5	23	35	200	5.714	65
Gopan	2	3	5	20	34	250	7.353	25
Gataraga	3	2	5	16	25	245	9.8	35
Sudan	2	3	4	22	25	150	6.0	40
Othuwa	3	2	3	20	22	120	5.455	45
Livowya	4	3	5	24	40	400	10.0	75
Essuti	4	4	5	24	35	330	9.429	75
IS 33461	2	4	5	20	20	120	6.0	35
B-35	4	1	3	16	20	105	5.25	50
E-36-1	3	2	5	22	41	195	4.756	55
KARI Mtama1	2	3	4	20	20	220	11.0	45
Ochuti	2	3	4	17	20	185	9.25	30
KBM 003	1	3	5	25	45	260	5.778	60
KBM 022	2	4	2	21	20	255	12.25	45
KBM 078	2	4	2	30	22	280	12.727	75
KBM 097	1	3	4	37	23	360	15.652	50
KAK 1950	1	3	2	18	18	170	9.444	45
KAK 7801	2	3	2	16	15	45	3.0	55
KAK 7837	2	3	2	17	14	105	7.5	35
(12X46)-1	2	5	4	27	19	110	5.789	40
IS 21055	2	3	2	16	18	150	8.333	35
IESV 92036	1	3	3	22	30	200	6.667	60
IS 8193	1	3	2	12	18	80	4.444	50
IS 21146	2	4	2	18	12	60	5.0	50
IS 13615	2	4	4	20	24	300	12.5	45

TABLE 3. Mean measurements on plant size, leaf and panicle morphological characteristics on plants grown at KARI biotechnology

Cultivar	Leaf					Panicle			
	Total plant height	Total node height	No (cm)	Length (cm)	Width (cm)	Length (cm)	Width (cm)	No. of tillers	Stem sizes
Macia	-	-	-	-	-	-	-	-	65
Gopan	51	30	6	19.5	6	5.5	2	10	25
Gataraga	81	49	7	26.5	3	8	3	11	35
Sudan	-	-	-	-	-	-	-	-	40
Othuwa	40	18	11	19	2.3	6	2	4	45
Livowya	85	54	9	28	3.5	9.5	6	18	75
Essuti	70.5	40	9	21	3	5	3.5	4	75
IS 33461	75	48	8	25	2.8	8	2	3	35
B-35	-	-	-	-	-	-	-	-	50
E-36-1	-	-	-	-	-	-	-	-	55
KARI Mtama1	-	-	-	-	-	-	-	-	45
Ochuti	53	26	5	22	2.5	8	1.5	16	30
KBM 003	76	47	8	25	3	9	4.5	7	60
KBM 022	74	47	8	22.5	2.5	8.5	2	15	45
KBM 078	66	46	9	20.5	3.3	5.5	3	8	75
KBM 097	60	40	8	16.5	2.5	5	2.7	5	50
KAK 1950	56.3	36	9	17.5	1.8	6	2	1	45
KAK 7801	45	23	6	11	1.5	7	1.5	33	55
KAK 7837	55	31	8	11	1.5	3.5	1.5	14	35
(12X46)-1	45	24	5	16	2	7	2	-	40
IS 21055	51	29	7	17	2	7	3	2	35
IESV 92036	64.5	35	6	20	3	9.5	1.5	11	60
IS 8193	58.2	35	7	14.5	2	7.2	2.5	10	50
IS 21146	40	14	4	18	2	7	2	9	50
IS 13615	50	20	4	19	2	8.2	1.5	20	45

7801 had the least water content (Fig. 1). On materials planted at Kiboko, Macia and Gataraga were among those cultivars that could be classified drought tolerant based on their high water content. However, the cultivars KAK 7801 and Othuwa were considered drought susceptible because of their low leaf water content and these measurements were repeated to confirm the results.

According to Silva *et al.* (2007), plants that can hold high amounts of leaf water are presumed more drought tolerant. This is also in agreement with reports by Araus *et al.* (1998), O'Neill *et al*. (2006) and Rong-hua *et al.* (2006) that worked on drought in wheat, corn and barley, respectively. These results demonstrated that phenotypic and physiological characteristics might be used as a selection criterion for yield performance in sorghum under drought stress.

Water shortage is one of the major limitations to productivity worldwide, and a feasible solution is to improve the drought tolerance of crop varieties through breeding. Water deficit stress is known to alter a variety of physiological processes such as leaf temperature LT, stomatal conductance, transpiration, photosynthesis and respiration which ultimately determines yield. The amount of water used by a crop is closely associated with photosynthetic activity, dry matter and yield in many species (Tollenaar and Aguillera, 1992). However, the maximum photosynthetic potential of crop is seldom reached due to unfavorable environmental factors including drought.

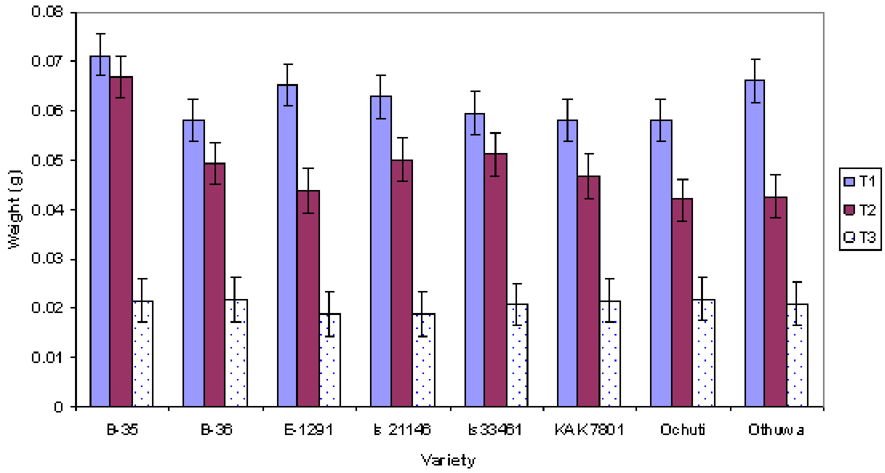

Figure1. Leaf weights measurements taken immediately after excision and after 2 drying periods (24 hours and 3 days after excision) at KARI Biotechnology Centre. Each sample was 20 sorghum leaf disks.

The degree of limitation of yield by environmental stresses varies even among genotypes within a species (Aguillera et *al.*, 1999). Therefore, the ability to maintain key physiological processes, such as photosynthesis during moderate drought stress, is indicative of the potential to sustain productivity under water shortage. To achieve this goal, a set of reliable parameters that can be rapidly and relatively inexpensive for screening is needed. Although not all phenotypic traits evaluated in this study were reliable in distinguishing between tolerant and susceptible sorghum cultivars additional physiological parameters may be more supportive in rapid screening for drought tolerance in sorghum.

CONCLUSION

Phenotypic and physiological factors in sorghum can be used to determine which cultivars are more resistant to drought than others. Based on phenotypic data, it is clear that five cultivars namely B35, E-36.1, KBM-003, IE SV 92036 and IS 13615 are the most drought tolerant. Data on RWC also support the observation that B35 and E-36.1 are the most drought tolerant cultivars among those evaluated. Measurements of both phenotypic and physiological are more reliable in determining drought tolerance in sorghum cultivars and in other cereals.

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