Physio-biochemical responses and defining selection criteria for drought tolerance in *Sorghum bicolor*

Masood Qadir[1], Amir Bibi[1], Hafeez Ahmad Sadaqat[1], Faisal Saeed Awan[2]

[1] Department of Plant Breeding and Genetics, University of Agriculture, Faisalabad, Punjab, Pakistan. 38000

[2] Centre of Agricultural Biochemistry and Biotechnology, University of Agriculture, Faisalabad, Punjab, Pakistan. 38000

* Corresponding author: Masood Qadir

Department of Plant Breeding and Genetics, University of Agriculture, Faisalabad, Punjab, Pakistan. 38000

Cell # 00923017935415, E-mail: masoodpbg@yahoo.com

KeyWords *Cytoplasmic male sterile, traits, variability, multivariate, water potential and association*

Abstract

Forty cytoplasmic male sterile (CMS) sorghum lines were evaluated for different physiological and biochemical traits under drought stress. Considerable genetic variability was found among all physio-biochemical traits i.e. water potential (Ψ w), stomatal conductance (g_s), photosynthetic efficiency, acid detergent fiber (ADF), neutral detergent fiber (NDF), ash and sugar contents. Results indicated that ash contents were found to be most adversely affected by drought stress followed by sugar contents and stomatal conductance respectively. However, the values of crude protein, acid detergent fiber (ADF) and neutral detergent fiber (NDF) were observed to be increase under stress condition. Principal component analysis (PCA) was applied to recognize drought tolerant lines. Selection criteria was based upon findings of correlation analysis among all studied traits. The positive association of water potential, stomatal conductance and photosynthetic efficiency with desirable traits viz. ash and sugar contents; and NDF association with undesirable traits viz. ADF and NDF revealed a way forward to design future breeding programs of sorghum crop under the prevailing scenario of climate change.

Introduction

Climate anomalies such as biotic and abiotic stresses due to global warming adversely affects the yield and growth of agricultural crops (Atkinson et al., 2013; Suzuki et al., 2014; Pandey et al., 2015). Different abiotic stress conditions such as heat, cold, salinity and drought indirectly affect the crop plants by favoring the spread of insects, pathogens and weeds (McDonald et al., 2009; Peters et al., 2014). These stresses also directly affect the crop plants by decreasing photosynthesis and whole plant growth, stomatal closure and wilting (Sanchez et al., 2002). Additionally, abiotic stresses such as drought boost the weeds competition with crops as numerous weedy plants show enhanced water use efficiency than crop plants (Valerio et al., 2013).

Among these abiotic stresses, drought stress is one of the most damaging factors that causes significant loss of crop yield (Amelework et al., 2015; Boyer and Westgate, 2004). Drought is the most edaphic stress that damages cellular homeostasis and hinders the plant growth (Dai 2011; Pandey and Shukla, 2015). Water demand for irrigation is continuously increasing while there was a drastic reduction in the availability of water, this condition is more critical in semi-arid and arid conditions (Rostamza et al., 2011). Drought is the state of water shortage due to abnormal rainfall for a prolonged period of time. Agriculture drought is the lack of sufficient moisture essential for normal crop growth and development to complete life cycle. In general, drought stress at any growth stage showed detrimental and negative effects on development and growth of crop, depending upon the crop growth stage and severity of drought stress. Drought affects the biochemical, morphological and physiological processes in crop plants. Significant consequences of drought on crop include a reduction in cell expansion and division rate, impaired germination, reduction in leaf size, disturbed stomata oscillation, decreased chlorophyll contents.

The Earth is a water-scarce planet; feeding more people by using less water is the major goal (Foley et al., 2011). To cop this drought challenge, crops having high adaptability in drier regions should be used. Among these, sorghum is one of the best choices grown for feed, food, fuel and fiber (Paterson et al., 2009; Qadir et al., 2015).

Sorghum (*Sorghum bicolor* L.), also known as sorgo, chari or jawar, is an important summer forage crop of

Pakistan (Rooney et al., 2007). Sorghum represents an excellent choice for single cut system and has yield potential comparable to maize. Higher biomass with higher dry matter, wide adaptation to soil and climatic conditions (Dolciotti et al., 1998; Reddy and Reddy, 2003), quick growth, effective C4 photosynthesis (Reddy et al., 2004; Taylor et al., 2010) and drought tolerance capable sorghum as superior forage crop. Sorghum is suitable in dry areas; when properly managed it can provide super feed supplement during lean periods in the form of hay (Brouk and Bean, 2011) and silage (Zhang et al., 2016). It was previously reported that water stress negatively influenced the chemical characteristics by affecting crude fiber, sugar, total ash, nitrogen free extracts and protein (Bibi et al., 2012; Kuchenmeister et al., 2013). Plants facing drought stress accumulate more lignin and neutral detergent fiber (NDF) (Amaducci et al., 2000; Carmi et al., 2006). Drought stress also negatively affects the physiological traits (chlorophyll, osmotic potential, photosynthesis, relative water contents) of sorghum (Premachandra et al., 1995; Qadir et al., 2015; Inoue et al., 2013; Tsuji et al., 2003). So, the sorghum lines showing a better physiological and biochemical response against drought stress should be screened. Germplasm from different sources has been extensively used in breeding improvement program around the world. The genetic variation among these germplasm makes them as dynamic donor of different genes to develop desired variety. So, in this study selection criteria will be developed to screen sorghum lines for drought tolerance. These lines will be used in development of sorghum-sudangrass hybrids for cultivation in drought prone areas of Pakistan.

Materials and Methods

Germplasm comprising 40 cytoplasmic male sterile (CMS) accessions of sorghum were collected from Fodder Research Institute (FRI), Sargodha, Pakistan, Maize and Millet Research Institute (MMRI), Sahiwal, Pakistan, Jullundur Private Limited (JPL), Rahimyar Khan, Pakistan, United State Department of Agriculture (USDA), USA and Dryland Farming Institute (DFI), China. The experiment was performed during the growing seasons 2015 and 2016, in the field area of Department of Plant Breeding and Genetics, University of Agriculture, Faisalabad. Drought stress was created by omitting the irrigation i.e. the normal field was irrigated by two times i.e. 20 and 40 days after sowing while drought field was irrigated only one time (20 days after sowing). The sorghum lines were evaluated under normal irrigation and drought stress for evaluating physiological and biochemical traits, reported below.

Physiological Traits

Stomatal conductance (g_s)

Average counts of stomatal conductance for 10 randomly selected flag leaves of intact plants from each accession were recorded with the help of a porometer (Model MK-3, Delta-T Devices, 123, Burwell, Cambridge England).

Water Potential (Ψw)

Water potential (Ψw) was measured in units of pressure using a pressure chamber (Model OSK2710, OGAWA Seiki Japan).

Photosynthetic efficiency

The variable fluorescence (Fv) and maximum fluorescence (Fm) values were measured through chlorophyll fluorometer. The ratio of Fv over Fm were used to calculate the photosynthetic efficiency (Fv/Fm).

Biochemical Traits

Randomly selected plants of sorghum form field were oven dried at 80% for 72 hours. The oven dried plants were ground in powder form and were put into NIR (Near-Infrared Reflectance) integrating sphere of Agri-NIR system for measurement of crude protein, acid detergent fiber and neutral detergent fiber.

Ash contents (%)

Two gram of oven dried sample was placed in a clean dry previously weighed china dish. The sample was ignited in a furnace at 600 °C till white or grey ash was obtained. The residue was cooled in desiccators and weight was recorded.

Ash contents % = (Weight of ash / Weight of sample) ×100

Sugar Contents (Brix%)

Sugar contents (brix) were recorded through digital refractometer (MA871 Digital Brix Refractometer made in Hungary).

Statistical analysis

Analysis of variance was used to determine the genetic variability. The Principal component analysis (PCA) was performed to select the lines for drought tolerance. R program was used to find the genotypic and phenotypic association among physiological and biochemical traits

Table 1 - Mean Squares from Analysis of Variance for biochemical traits in sorghum

Source of variation	Degree of Freedom	Crude protein	Acid detergent fiber	Neutral detergent fiber	Ash contents	Sugar contents
Accessions (A)	39	7.37**	72.36**	105.94**	35.06**	16.97**
Treatments (T)	1	95.50**	42.76**	123.85**	399.41**	198.92**
Interaction A × T	39	0.55**	0.79**	1.39**	1.54**	0.99**

* Significant at 5% probability level, ** Significant at 1% probability level

Results

Genetic variability

Analysis of variance, respectively for biochemical and physiological traits, (Table 1 and Table 2) indicated the existence of significant genetic variability for all the studied traits under normal irrigation and drought stress. Interaction among genotypes and treatments was also significant and indicated that selection of treatments and genotypes was appropriate. So, variation of accessions over treatments could provide an opportunity for breeding of biochemical traits along with physiological traits under drought stress conditions.

Table 2: Mean Squares from Analysis of Variance for physiological traits in sorghum

Source of variation	Degree of Freedom	Stomatal conductance	Water potential	Photosynthetic efficiency
Accessions (A)	39	53.33**	2.38**	1.96**
Treatments (T)	1	1055.88**	60.33**	0.78**
Interaction A × T	39	5.92**	0.29**	0.20**

* Significant at 5% probability level, ** Significant at 1% probability level

Performance of physiological and biochemical traits

The mean performance expression of all traits except crude protein, acid detergent fiber and neutral detergent fiber showed decrease under drought stress as compared to normal irrigations during 2015 (Table 3). Highest reduction (-21.68%) was observed for stomatal conductance followed by ash contents (-18.50%) whereas maximum increase (14.45%) was observed in crude protein under drought stress during 2015. Similar trends were also observed in 2016 (Table 4); stomatal conductance, water potential, photosynthetic efficiency decreased while crude protein, neutral detergent fiber (NDF) and acid detergent fiber (ADF) increased under drought stress. More in detail, highest reduction (-20%) was observed for stomatal conductance followed by ash contents (-19.16%) while highest increase (13.25%) was observed in crude protein under drought stress during 2016.

Principal component analysis

Principal component analysis (Fig. 1) was performed to screen the sorghum lines. In PCA, lines FRI-A1,

Table 3: Range and average percentage increase or decrease under drought stress as compared to normal irrigation for physiological and biochemical traits in sorghum during 2015

Sr.	Traits	Conditions	Range	Average % decrease or increase in drought as compared to normal irrigation
1	Stomatal conductance (mol m-2 s-1)	Normal	11.8-23.3	-21.68
		Drought	7.7-19.90	
2	Water potential (MPa)	Normal	-4.88- -2.58	-14.20
		Drought	-5.60- -3.30	
3	Photosynthetic efficiency	Normal	0.31-0.66	-9.8
		Drought	0.19-0.62	
4	Protein (%)	Normal	5.2-9	14.45
		Drought	6.1-10.2	
5	Acid detergent fiber (%)	Normal	22.62-36.82	3.3
		Drought	25.24-37.4	
6	Neutral detergent fiber (%)	Normal	56.3-73.67	1.9
		Drought	57.17-74.2	
7	Sugar contents (%)	Normal	6.9-15.88	-15.7
		Drought	5.1-13.71	
8	Ash contents (%)	Normal	5.1-11.40	-18.5
		Drought	4.3-9.60	

Table 4: Range and percentage increase or decrease under drought stress as compared to normal irrigation for physiological and biochemical traits in sorghum during 2016

Sr.	Traits	Conditions	Range	Average % decrease or increase in drought as compared to normal irrigation
1	Stomatal conductance (mol m-2 s-1)	Normal	12.1-22.9	- 20%
		Drought	8.2-19.6	
2	Water potential (MPa)	Normal	-4.70- - 2.55	-13.1
		Drought	-5.47- - 3.20	
3	Photosynthetic efficiency	Normal	0.33-0.71	-10.4
		Drought	0.18-0.64	
4	Protein (%)	Normal	5.25-8.7	13.25
		Drought	6.9-11.8	
5	Acid detergent fiber (%)	Normal	23.45-35.75	2.5
		Drought	25.75-36.42	
6	Neutral detergent fiber (%)	Normal	54.6-72.9	2.1
		drought	56.45-74.99	
7	Sugar contents (%)	Normal	7-14.75	-13.8
		drought	5.2-12.76	
8	Ash contents (%)	Normal	6.2-11.15	-19.16
		drought	4.3-9.10	

FRI-A2, FRI-A5, FRI-A9, Red line, Y-1, Y-2, Y-6, PI 644512 and PI 569994 gained position in Quadrate I. Therefore, these sorghum lines are considered drought tolerant. While lines Y-3, Y-4, Y-7, PI 570821, PI 570821, PI 217799, PI 330036, Y-9 and FRI-A4, FRI-A3, FRI-A8, FRI-A12, FRI-A13 fell in Quadrate IV so these result the drought susceptible sorghum lines.

Association among physiological and biochemical traits

Association among the physiological and biochemical traits under drought conditions, during 2015-2016, (Table 5) showed that stomatal conductance has a significantly positive genotypic and phenotypic association with water potential, photosynthetic efficiency, sugar contents and ash contents while significantly negative association with crude protein. Water potential also has positive association with

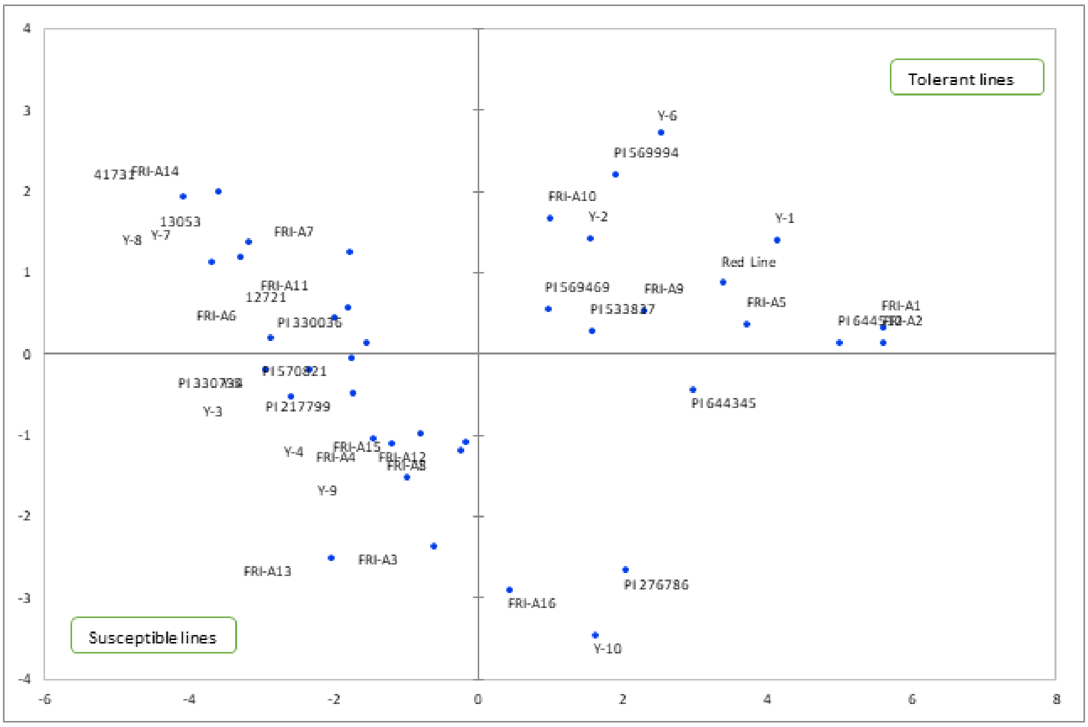

Fig. 1 -Principal Component Analysis (PCA) for 40 accessions of sorghum under drought stress.

Table 5: Genotypic and phenotypic association among physiological and biochemical traits in sorghum under drought stress during 2015-2016

		SC	WP	PE	Protein	ADF	NDF	Sugar contents	AC
SC	G	1	0.60**	0.52**	-0.40**	-0.11	-0.07	0.72**	0.32**
	P		0.57**	0.50**	-0.39**	-0.09	-0.06	0.70**	0.30**
WP	G		1	0.44**	-0.32**	-0.23*	-0.08	0.50**	0.42**
	P			0.41**	-0.32**	-0.21**	-0.05	0.48**	0.39**
PE	G			1	-0.39**	-0.18	-0.07	0.35**	0.27**
	P				-0.35**	-0.14	-0.05	0.33**	0.24*
Protein	G				1	0.25*	0.29*	-0.43**	-0.21**
	P					0.22*	0.27*	-0.40**	-0.18
ADF	G					1	0.69**	-0.11	-0.28**
	P						0.65**	-0.07	-0.24*
NDF	G						1	-0.08	-0.10
	P							-0.07	-0.08
AC	G							1	0.41**
	P								0.38**

G Genotypic association, **P** Phenotypic association, **SC** Stomatal conductance, **ADF** Acid detergent fiber, **WP** Water potential

NDF Neutral detergent fiber, **PE** Photosynthetic efficiency, **AC** Ash contents

photosynthetic efficiency, sugar contents and ash contents but negative correlation with protein, acid detergent fiber and neutral detergent fiber. Acid detergent fiber and neutral detergent fiber showed significantly negative genotypic and phenotypic association with all the traits except crude protein. Results showed that sugar contents and ash contents have a significantly positive genotypic and phenotypic association with all the physiological traits while negative association with all the biochemical traits. Table 5 also showed that values of phenotypic effects are lower than the those of genotypic effects.

Discussion

Genetic variability and mean performance of genotypes

Development of drought tolerant, high quality sorghum hybrids and sorghum-sudangrass hybrids is of prime importance for sorghum breeder. Effective screening of germplasm for biochemical and physiological traits especially under drought stress is a valuable way of selecting material for development of varieties and hybrids. The highly significant differences observed between the accessions for all the physiological and biochemical traits showed that the sorghum germplasm has high variability and could be effectively used for breeding purpose (Mwadzingeni et al., 2016).

Drought stress induced a negative effect on physiological and biochemical traits of sorghum (Bibi et al., 2010; Qadir et al., 2015); stomatal conductance, water potential and photosynthetic efficiency are important traits for drought tolerance. Exposure of plants to drought stress decreased the water potential (Siddique et al., 2001). Reduction in stomatal conductance under drought stress showed the partial closing of stomata to preserve water (Sumayao, 1977; Munamava and Riddoch, 2001). Reduction in stomatal conductance is due to accumulation of abundant abscisic acid (ABA) under water stress (Yang et al., 2011). Stomata are very sensitive to reduction in leaf water potential (Clark, 1982), however, sorghum has ability to keep stomata open even at very low water potential (Ackerson et al., 1977). Any reduction in stomatal conductance can limit the uptake of CO_2 which ultimately results in reduction of photosynthetic activity (Haworth et al., 2016). Photosynthetic efficiency was also reduced under drought stress due to reduction in leaf area (Munamava and Riddoch, 2001). Photosynthetic efficiency decreased under drought stress, as was also reported by Blanco et al. (2000) and Samarah and Alqudah (2011). Drought stress inhibits the photosynthetic efficiency by reducing chlorophyll contents and damaging the photosynthetic apparatus (Ormaetxe et al., 1998) and causing metabolic impairments or stomatal closure (Tezara et al., 1999). Water potential is reduced under water stress, as was also previously reported by Sgherri et al. (1995) and Pennypacker et al. (1990). The reduction in water potential may be due to change in osmotic pressure (Siddique et al., 2001).

Results of biochemical traits indicated that drought stress increased the crude protein, same results were also reported by Bibi et al. (2012); Kuchenmeister et al. (2013) and Qadir et al. (2015). While Liu et al. (2018) found a small reduction in crude protein under water stress. Carter and Sheaffer (1983) reported that crude protein remains unaffected in alfalfa during water stress. Hale and Orcutt (1987) observed that plant synthesize special high molecular proteins during water stress to assist them in resisting the effect of water stress. On the contrary, CP, NDF and ash contents were not affected and remain at the same level during water stress and normal irrigation (Dominguez et al., 1996). Peterson et al. (1992) reported inconsistency in crude protein concentration in forage legumes. Moreover, current study showed that sugar contents (brix value) were decreased under drought stress. Almodares et al. (2013) also found negative effects of drought on sugar production as well as sugar accumulation in sorghum. Post flowering drought also affect the sugar production in sorghum (Tovignan et al., 2016). Reduction in forage quality like sugar, crude fiber, nitrogen free extract (NFE) and protein by drought stress was also reported by Bibi et al. 2012 and Kuchenmeister et al. 2013. Ash contents were reduced with increasing water stress (Qadir et al., 2015). Bibi et al. (2012) and Kuchenmeister et al. (2013) also found a negative effect of drought on ash contents in sorghum. Results showed that drought did not affect significantly the neutral detergent fiber (NDF) and acid detergent fiber (ADF). There is little increase in ADF and NDF under drought. Udomprasert and Sawasdiphanich (1995) also found no effect of drought on ADF. While Seguin et al. (2002) found that drought increases ADF concentration but a little effect on NDF concentration was observed. On the other hand, drought decreases ADF and NDF content in alfalfa (Abid et al., 2016) and in forage legumes (Peterson et al., 1992). Halim et al. (1990) also reported an NDF reduction in both stem and leaves under drought. Contrary to these, Dominguez et al. (1996) observed no effect of drought on ADF and NDF which remained the same under normal as well as drought conditions.

Association Analysis

Association analysis provides the strength of relationship among two traits and allows finding direction and amount of association among them which is essential for creating an effective and efficient crop improvement procedure. For present study, correlation coefficients were figured among physiological and biochemical traits. During development of variety, the breeding could be very effective when there is positive association among the desired traits, but it could be very difficult if these traits are negatively associated (Nemati et al., 2009). The sugar contents and ash contents should be high under drought stress while NDF and ADF have negative impact so their content should be lower in plants under drought stress. The positive association of stomatal conductance, water potential and photosynthetic efficiency with sugar and ash contents and negative association with ADF and NDF showed that sorghum drought tolerant plants could be selected based on these physiological traits. The negative association of protein content with physiological traits is due to the increased protein production under drought stress (Dhindsa and Cleland, 1975; Cao et al., 2017). The lower association values of phenotypic traits than genotypic ones showed that these traits are less affected by the environments and traits are under genetic control.

References

Abid M, Mansour E, Yahia LB, Bachar K, Ben Khaled A, Ferchichi A, 2016. Alfalfa nutritive quality as influenced by drought in south-eastern oasis of tunisia. Italian J Animal Sci 15:334-42

Ackerson RC, Krieg DR, Sung FJM, 1980. Leaf conductance and osmoregulation of field-grown sorghum genotypes1. Crop Science 20:10-14

Almodares A, Hotjatabady R, Mirniam E, 2013. Effects of drought stress on biomass and carbohydrate contents of two sweet sorghum cultivars. J environ biol 34:585

Amaducci S, Amaducci M, Benati R, Venturi G, 2000. Crop yield and quality parameters of four annual fibre crops (hemp, kenaf, maize and sorghum) in the North of Italy. Industrial Crops Products 11:179-86

Amelework B, Shimelis H, Tongoona P, Laing M, 2015. Physiological mechanisms of drought tolerance in sorghum, genetic basis and breeding methods: a review. Afr J Agric Res 10:3029-40

Atkinson NJ, Lilley CJ, Urwin PE, 2013. Identification of genes involved in the response of Arabidopsis thaliana to simultaneous biotic and abiotic stresses. Plant physiol :pp. 162:228-2041

Bibi A, Sadaqat H, Tahir M, Usman BF, Ali M, 2012. Genetic analysis of forage quality traits in sorghum-sudangrass hybrids under water stress. J Animal Plant Sci 22:1092-100

Bibi A, Sadaqat HA, Akram HM, Mohammed MI, 2010. Physiological markers for screening sorghum (Sorghum bicolor) germplasm under water stress condition. Int J Agric Biol 12:451-5

Boyer J, Westgate M, 2004. Grain yields with limited water. J experimental Botany 55:2385-2394

Brouk M, Bean B, 2011. Sorghum in dairy cattle production feeding guide. United Sorghum Check off Program, Lubbock, TX, USA

Cao Y, Luo Q, Tian Y, Meng F, 2017. Physiological and proteomic analyses of the drought stress response in Amygdalus Mira (Koehne) Yü et Lu roots. BMC plant biol 17:1-16

Carmi A, Aharoni Y, Edelstein M, Umiel N, Hagiladi A, 2006. Effects of irrigation and plant density on yield, composition and in vitro digestibility of a new forage sorghum variety, Tal, at two maturity stages. Animal Feed Sci Technol 131:121-33

Carter P, Sheaffer C, 1983. Alfalfa Response to Soil Water Deficits. I. Growth, Forage Quality, Yield, Water Use, and Water-Use Efficiency 1. Crop Sci 23:669-75

Clark R, Mineral nutritional factors reducing sorghum yields: Micronutrients and acidity. Proc. ICRISAT, Sorghum in the Eighties: Proceedings of the international symposium on sorghum, 1982:179-90:

Dai A, 2011. Drought under global warming: a review. Wiley interdisciplinary reviews. Climate Change 2:45-65

Del Blanco I, Rajaram S, Kronstad W, Reynolds M, 2000. Physiological performance of synthetic hexaploid wheat-derived populations. Crop Sci 40:1257-63

Dhanda SS, Sethi GS, Behl RK, 2004. Indices of drought tolerance in wheat genotypes at early stages of plant growth. J Agronomy Crop Sci 190:6-12

Dhindsa RS, Cleland RE, 1975. Water stress and protein synthesis. i. differential inhibition of protein synthesis 55:778-81

Dolciotti I, Mambelli S, Grandi S, Venturi G, 1998. Comparison of two sorghum genotypes for sugar and fiber production. Industrial Crops Products 7:265-72

Foley JA, Ramankutty N, Brauman KA, Cassidy ES, Gerber JS, 2011. Solutions for a cultivated planet. Nature 478:337

Hale MG, Orcutt DM, 1987. The physiology of plants under stress. John Wiley & Sons

Halim R, Buxton D, Hattendorf M, Carlson R, 1990. Crop water stress index and forage quality relationships in alfalfa. Agronomy j 82:906-909

Haworth M, Killi D, Materasii A, Raschi A, Centritto M, 2016. Impaired stomatal control is associated with reduced photosynthetic physiology in crop species grown at elevated [CO2]. Front Plant Sci 7:e1568

Inoue T, Yamauchi Y, Eltayeb AH, Samejima H, Babiker AGT, Sugimoto Y, 2013. Gas exchange of root hemi-parasite Striga hermonthica and its host Sorghum bicolor under short-term soil water stress. Biologia Plantarum 57:773-777

Iturbe-Ormaetxe I, Escuredo PR, Arrese-Igor C, Becana M, 1998. Oxidative damage in pea plants exposed to water deficit or paraquat. Plant physiol 116:173-181

Kuchenmeister K, Küchenmeister F, Kayser M, Wrage-Mönnig N, Isselstein J, 2013. Influence of drought stress on nutritive value of perennial forage legumes. Int J Plant Prod 7:693-710

Liu Y, Wu Q, Ge G, Han G, Jia Y, 2018. Influence of drought stress on afalfa yields and nutritional composition. BMC plant biol 18:1-9

Dominguez LU, Maiti R, Francisco L, 1996. Nutritional quality of 15 glossy sorghum forages at different growth stages in irrigated and rainfed situations. International sorghum and millets newsletter (USA)

McDonald A, Riha S, DiTommaso A, DeGaetano A, 2009. Climate change and the geography of weed damage: analysis of US maize systems suggests the potential for significant range transformations. Agric ecosystems environment 130:131-140

Munamava M, Riddoch I, 2001. Response of three sorghum (Sorghum bicolor L. Moench) varieties to soil moisture stress at different developmental stages. South Afr J Plant Soil 18:75-79

Mwadzingeni L, Shimelis H, Tesfay S, Tsilo TJ, 2016. Screening of bread wheat genotypes for drought tolerance using phenotypic and proline analyses. Frontiers plant sci 7:1-12

Nemati A, Sedghi M, Sharifi RS, Seiedi MN, 2009. Investigation of correlation between traits and path analysis of corn (Zea mays L.) grain yield at the climate of Ardabil region (Northwest Iran). Notulae Botanicae Horti Agrobotanici Cluj-Napoca 37:194-198

Pandey P, Ramegowda V, Senthil-Kumar M, 2015. Shared and unique responses of plants to multiple individual stresses and stress combinations: physiological and molecular mechanisms. Frontiers in plant sci 6:1-14

Pandey V, Shukla A, 2015. Acclimation and tolerance strategies of rice under drought stress. Rice Sci 22:147-161

Paterson AH, Bowers JE, Bruggmann R, Dubchak I, Grimwood J, 2009. The Sorghum bicolor genome and the diversification of grasses. Nature 457:551

Pennypacker B, Leath K, Stout W, Hill R, 1990. Technique for simulating field drought stress in the greenhouse. Agronomy j 82:951-957

Peters K, Breitsameter L, Gerowitt B, 2014. Impact

of climate change on weeds in agriculture: a review. Agronomy Sustainable Devel 34:707-721
Peterson P, Sheaffer C, Hall M, 1992. Drought effects on perennial forage legume yield and quality. Agronomy J 84:774-779
Premachandra GS, Hahn DT, Rhodes D, Joly RJ, 1995. Leaf water relations and solute accumulation in two grain sorghum lines exhibiting contrasting drought tolerance. J Experimental Botany 46:1833-1841
Qadir M, Bibi A, Tahir MH, Saleem M, Sadaqat HA, 2015. Screening of sorghum (Sorghum bicolor L) genotypes under various levels of drought stress. Maydica 60:M35
Reddy B, Reddy PS, 2003. Sweet sorghum: characteristics and potential. Int Sorghum Millets Newsletter 44:26-28
Reddy BV, Ramesh S, Reddy PS, 2004. Sorghum breeding research at ICRISAT-goals, strategies, methods and accomplishments. Int Sorghum Millets Newsletter 45:5-12
Richards R, 1988. Breeding for drought resistance-physiological approaches
Rooney WL, Blumenthal J, Bean B, Mullet JE, 2007. Designing sorghum as a dedicated bioenergy feedstock. Biofuels Bioproducts Biorefining 1:147-157
Rostamza M, Chaichi M-R, Jahansooz MR, Mashhadi HR, Sharifi HR, 2011. Effects of water stress and nitrogen fertilizer on multi-cut forage pearl millet yield, nitrogen, and water use efficiency. Communications soil sci plant analysis 42:2427-2440
Samarah N, Alqudah A, 2011. Effects of late-terminal drought stress on seed germination and vigor of barley (Hordeum vulgare L.). Archives Agronomy Soil Sci 57:27-32
Sanchez A, Subudhi P, Rosenow D, Nguyen H, 2002. Mapping QTLs associated with drought resistance in sorghum (Sorghum bicolor L. Moench). Plant molecular biol 48:713-726
Seguin P, Mustafa A, Sheaffer C, 2002. Effects of soil moisture deficit on forage quality, digestibility, and protein fractionation of Kura clover. J Agronomy Crop Sci 188:260-266
Sgherri CLM, Navari-Izzo F, 1995. Sunflower seedlings subjected to increasing water deficit stress: oxidative stress and defence mechanisms. Physiologia Plantarum 93:25-30
Siddique KHM, Regan KL, Tennant D, Thomson BD, 2001. Water use and water use efficiency of cool season grain legumes in low rainfall Mediterranean-type environments. European J Agronomy 15:267-80
Sumayao CR, Kanemasu ET, Hodges T, 1977. Soil moisture effects on transpiration and net carbon dioxide exchange of sorghum. Agricultural Meteorology 18:401-408
Suzuki N, Rivero RM, Shulaev V, Blumwald E, Mittler R, 2014. Abiotic and biotic stress combinations. New Phytologist 203:32-43
Taylor SH, Hulme SP, Rees M, Ripley BS, Woodward FI, Osborne CP, 2010. Ecophysiological traits in C3 and C4 grasses: a phylogenetically controlled screening experiment. New Phytologist 185:780-791
Tezara W, Mitchell V, Driscoll S, Lawlor D, 1999. Water stress inhibits plant photosynthesis by decreasing coupling factor and ATP. Nature 401:914
Tovignan TK, Fonceka D, Ndoye I, Cisse N, Luquet D, 2016. The sowing date and post-flowering water status affect the sugar and grain production of photoperiodic, sweet sorghum through the regulation of sink size and leaf area dynamics. Field crops research 192:67-77
Tsuji W, Ali M, Inanaga S, Sugimoto Y, 2003. Growth and gas exchange of three sorghum cultivars under drought stress. Biologia Plantarum 46:583-587
Udomprasert N, Sawasdiphanich S, 1995. Effects of water stress on proline content and forage quality. Witthayasan Kasetsart (Sakha Witthayasat)
Valerio M, Lovelli S, Perniola M, Di Tommaso T, Ziska L, 2013. The role of water availability on weed–crop interactions in processing tomato for southern Italy. Acta Agriculturae Scandinavica, Section B–Soil Plant Sci 63:62-68
Yang S-D, Seo PJ, Yoon H-K, Park CM, 2011. The Arabidopsis NAC transcription factor VNI2 integrates abscisic acid signals into leaf senescence via the COR/RD genes. Plant Cell 23:2155-2168
Zhang Sj, Chaudhry AS, Ramdani D, Osman A, Guo Xf, 2016. Chemical composition and in vitro fermentation characteristics of high sugar forage sorghum as an alternative to forage maize for silage making in Tarim Basin, China. J Integrative Agriculture 15:175-82