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Growth response and plant water status in forage sorghum [*Sorghum bicolor* (L.) Moench] cultivars subjected to decreasing levels of soil moisture

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Abstract

A pot experiment was conducted to determine the response of two recent forage sorghum cultivars (JS-2002 and Chakwal Sorghum) and an old one (JS-263) to three levels of soil moisture (30%, 50% and 70% field capacity). Several traits were assessed addressing plant morphology, functional growth, leaf water status, biomass yield and water use efficiency (WUE). Soil moisture variation greatly affected all traits, while cultivars significantly differed in the response to drought. At low moisture the three genotypes showed similar net assimilation rate, specific leaf area, root and shoot dry weight. Conversely, at high moisture JS-2002 exhibited a higher potential than Chakwal Sorghum, in turn passing JS-263. As it concerns plant height, leaf area, leaf water potential (LWP) and relative water content (RWC), the three cultivars consistently behaved across moisture levels maintaining the same ranking between best (JS-2002) and worst performer (JS-263). Especially in LWP and RWC the gap between JS-263 and JS-2002 (LWP, -1.84 vs. -1.55 MPa; RWC, 71 vs. 78% in the two respective cultivars) points out the old genotype inadequacy to face drought. WUE outlined an increasing difference between most (S-2002) and least efficient cultivar (JS-263) at rising moisture. JS-263 also showed a higher yield response factor to water supply, meaning a stronger yield decrease under water deficit. The resilience to drought shown by recent varieties (JS-2002 and Chakwal Sorghum) is a good premise for their use in areas subjected to dry spells. Further research at field plot scale is nevertheless needed to assess actual gains in varying moisture conditions.

Keywords: Biomass yield; Drought; Leaf water potential; Relative water content; Net assimilation rate; Water use efficiency.
Abbreviations: a.s.l.- above sea level; CV- cultivar; DAS- day(s) after seeding; FC- field capacity; Ky, yield response factor to water supply; LWP- leaf water potential; LS- leaf succulence; ML- moisture level; NAR- net assimilation rate; R:S- root to shoot ratio; RDW- root dry weight; RWC- relative water content; SDW- shoot dry weight; SLA- specific leaf area; TDW- total dry weight; WUE- water use efficiency.

Introduction

Sorghum ranks fifth among world cereals after wheat, rice, maize and barley (Sato et al., 2004; Khalil, 2008). The crop is primarily grown in the warm dry climates of Africa, India, Pakistan, China and the Southern United States, to be used as food and fodder (Alagarswamy and Chandra, 1998). Sorghum grain is also used as poultry feed; the stems for sugar extraction; the whole biomass for bio-fuels, fibre extraction and feed for animals during periods of fodder scarcity (Doggett, 1988). The crop is adapted to the arid and semi-arid tropics and dry-temperate areas of the world (Kidambi et al., 1990; Blum, 2004). Sorghum is better suited to bio-chemically and physiologically withstand high temperatures and low moisture conditions than C_3 cereals (Downes, 1972) and maize (Farré and Faci, 2006). In Pakistan, forage sorghum is grown on an area of 221 thousand hectares, 80% of which is located in the central province of Punjab where it averages 29.1 t ha^{-1} of fresh forage. Sorghum contributes 30% to the country's total fodder production (NARC, 2008). Drought is a multidimensional stress affecting plants at various levels of their organization (Blum, 1996). It is generally accepted as the most widespread abiotic stress (Quarrie et al., 1999) as well as a major crop limiting factor in many areas of the world. Even intermittent water deficit at critical stages of cereal crops may reduce yield (Ludlow and Muchow, 1990).
Water deficit affects nearly all growth processes; however, the stress response depends upon the intensity, rate, duration of exposure and stage of plant development (Brar et al., 1990; Sinaki et al., 2007). In sorghum, water stress occurring between pre- and post-flowering decreases seed filling duration, seed size and number, thus leading to strong yield reduction or even total crop loss (Mkhabela, 1995; Tuinstra et al., 1997). Sorghum avoids dehydration by enhanced water uptake through its deep and extensive root system, and tolerates dehydration by osmotic regulation (Wright and Smith, 1983; Singh, 1989). The high tillering of forage sorghum provides compensation when the main stem is damaged by water stress, fostering yield stability in rainfed areas (Richards, 1987; Mahalakshmi and Bidinger, 1986). In addition to this, sorghum restrains transpirational loss of water through upright leaf habit (Begg, 1980). Soil moisture deficiency may also affect the growth of the root apparatus, which is responsible for establishing the soil-plant-atmosphere continuum in the flow of water (Kuchenbuch et al., 2006). Previous studies in sorghum have shown that total leaf area and specific leaf area decrease under water stress, while the root to shoot ratio increases (Munamava et al.,

2001). Leaf area reduction in response to water stress occurs either through hastened leaf senescence or decline in leaf expansion; the extent of the reduction depends on the degree of tolerance possessed by different sorghum varieties (Stout et al., 1980; Krieg, 1994; Ashraf and Ahmad, 1998). In view of the above mentioned divergent results, the present study was conducted to evaluate the response of forage sorghum cultivars to a wide range of soil moisture conditions, in order to assess the advantages and disadvantages of recent varieties vs. an old variety in terms of morphology, biomass and traits associated with plant moisture status.

Results

Morphological traits

Plant height significantly varied among cultivars and moisture levels in the three growth stages (Table 1): the cultivar JS-2002 was the tallest, followed by Chakwal Sorghum, which in turn was taller than JS-263. Likewise, 70% FC showed higher plants than 50% FC, which in turn was taller than 30% FC. In both factors, the intermediate treatment (Chakwal Sorghum and 50% FC) was a little closer to the worst (JS-263 and 30% FC) than to the best treatment (JS-2002 and 70% FC). Leaf number and area at harvest outlined a pattern similar to plant height (Table 1): the three cultivars and moisture levels determined the same ranking with no significant interaction. In general, JS-2002 had about 15% more leaves, 10% more expanded than JS-263, while 70% FC determined an approximate 30% and 10% gain in the two respective traits, compared to 30% FC.

Growth and leaf water traits

Moisture exerted a stronger influence on functional growth and leaf water status, compared to genotypes. The two factors significantly interacted in three out of five traits (Table 2).

Net assimilation rates increased 40% between JS-263 and JS-2002; more than four times between 30% and 70% FC (Table 2). The interaction showed a variable behaviour of the intermediate cultivar (Chakwal Sorghum) with respect to the other two (Fig. 1.a). Specific leaf area increased an approximate 50% between JS-263 and JS-2002; more than three times between 30% and 70% FC (Table 2). The interaction showed a steeper SLA increase in JS-2002 than in the other two cultivars (Fig. 1.b). Leaf water potential exhibited the highest level, i.e. the least negative one, in Chakwal Sorghum; JS-2002 closely followed (-5%), whereas JS-263 lagged behind (-25%) (Table 2). Water regimes depicted a consistent LWP decrease between 70% and 30% FC (-30%). Therefore, soil moisture determined about the same LWP range as cultivars (~0.35 MPa). The insignificant interaction indicates a consistent behaviour of genotypes across the three water regimes. Relative water content displayed only a 10% difference between top (JS-2002) and bottom ranking cultivar (JS-263), compared to a 30% variation between adequate (70% FC) and poor water availability (30% FC) (Table 2). Also in RWC, the insignificant interaction means consistent behaviour of genotypes across water regimes. Moisture exerted a stronger effect (two-fold increase) than genotypes (50% increase) on leaf succulence (Table 2). The interaction outlined a steady rise of JS-2002 across water regimes; a steep increase of Chakwal Sorghum until 50% FC; a slow rise of JS-263 up to 70% FC (Fig. 1.c).

Plant biomass, water effects and trait inter-relations

Biomass traits and WUE were more influenced by water regimes than cultivars. Moreover, the significant interactions in all traits indicate specific genotype behaviour in response to soil moisture variations (Table 3). Relative dry weight exhibited a 20% increase between JS-263 and JS-2002, compared to an almost three-fold increase between 30% and 70% FC (Table 3). The interaction showed a progressive distancing of JS-263 from the other two varieties (Fig. 2.a). Stem dry weight underwent relevant increases in response to cultivars (+55% between JS-263 and JS-2002) and especially moisture levels (five-fold increase between 30% and 70% FC). The interaction showed enhanced genotype differences at increasing water regime (Fig. 2.b). Total dry weight reflected the combined variations of the two previous traits, although the influence of SDW was more noticeable (Table 3; interaction not shown). Root to shoot ratio reflected the relative variations of RDW and SDW (Table 3): the ratio decreased by one third between JS-263 and JS-2002; by 50% between 30% and 70% FC. The interaction showed a steady, parallel decrease in JS-2002 and Chakwal Sorghum, whereas in JS-263 the R:S ratio only dropped to 70% FC (Fig. 2.c). Water use efficiency was also significantly influenced by cultivars (50% increase between JS-263 and JS-2002) and especially moisture levels (two-fold increase between 70% FC and 30% FC) (Table 3). In both factors, the intermediate treatment (Chakwal Sorghum; 50% FC) was a little closer to the best (JS-2002 and 70% FC) than to the worst treatment (Js-263 and 30% FC). The interaction showed a consistent WUE rise of JS-2002 at increasing moisture, while the other two cultivars lagged behind to a greater (JS-263) or lesser extent (Chakwal Sorghum) (Fig. 2.d). JS-263 also showed a higher yield response factor (Ky) to water (Table 3), meaning a stronger reduction in SDW than in ET when diminishing the amount of water supply.

Significant correlations were observed among functional, leaf water and biomass traits (Table 4). Only the R:S ratio featured negative correlations with the other traits, the rest of them being positively inter-related. More specifically, NAR showed r values between 0.810 and 0.914 with the four leaf traits (SLA, LWP, RWC and LS), and was also well correlated with plant biomass (r between 0.933 and 0.997 with RDW, SDW and TDW) and WUE (r = 0.979). The four leaf traits showed somewhat weaker inter-relations (r between 0.714 and 0.838); they were better associated with RDW, SDW and TDW (average r = 0.868), although LWP proved a little weaker than the other three leaf traits in these correlations. Relative dry weight was well related to SDW and TDW (r = 0.924 and 0.937, respectively). A very good correlation (r = 0.998) was also observed between SDW and TDW, which is consistent with the fact that SDW accounted for an average 87% of TDW. Last, RDW, SDW and TDW showed good correlations with WUE (r between 0.939 and 0.984), indicating a straight relationship between consumptive water use and biomass output.

Discussion

Plant morphology

The good correlation between plant height and SDW (r between 0.827** and 0.844** in the three growth stages; Table 4) makes it possible to predict the final output of biomass (DAS 60) at about mid growth (DAS 25). It there-

Table 1. Effects of cultivars and soil moisture levels on the morphology of forage sorghum cultivars grown in a controlled environment.

Source	Plant height (m)			Leaves pl.$^{-1}$	Leaf area (m^2)
	5th leaf	pre-booting	50% heading	50% heading	50% heading
Cultivar (CV)					
JS-2002	0.96 a	1.05 a	1.18 a	5.9 a	0.028 a
Chakwal S.	0.87 b	0.98 b	1.11 b	5.6 ab	0.027 b
JS-263	0.83 c	0.92 c	1.04 c	5.1 b	0.025 c
P	<0.001**	<0.001**	<0.001**	<0.001**	<0.001**
Moisture level (ML)					
30% FC	0.81 c	0.90 c	1.02 c	4.7 c	0.026 c
50% FC	0.87 b	0.98 b	1.10 b	5.6 b	0.027 b
70% FC	0.99 a	1.08 a	1.21 a	6.3 a	0.028 a
P	<0.001**	<0.001**	<0.001**	<0.001**	<0.001**
CV × ML					
P	0.306 ns	0.484 ns	0.911 ns	0.489 ns	0.976 ns
C.V. (%)	4.2	4.7	4.8	12.5	3.1

ns, * and ** mean non-significant, significant at $P \leq 0.05$ and $P \leq 0.01$, respectively. Different letters indicate significantly-different means (SNK test; $P \leq 0.05$). FC, field capacity.

fore appears that, under steady irrigation treatments as in our experiment, early sorghum behaviour is consistently maintained up to the end. This circumstance is seldom addressed in the literature, despite the possibility of using early trait assessment in yield forecasts. This perspective is supported by a field experiment with Chakwal Sorghum (Sher et al., 2011), where a good correlation was observed between height at mid growth (DAS 30) and SDW at DAS 60 (r = 0.740**). The trends observed by Saeed and El-Nadi (1998) further support our hypothesis.

Functional growth and leaf water status

In general, low NAR values were recorded in our experiment, although the comparison is often made with sorghum growing in field plots instead of pots. Under field conditions, Amal et al. (2010) observed an approximate 5% variation in NAR between two grain sorghum cultivars, whereas skipping one irrigation out of five plunged NAR from about 50 to 30 g m^{-2} d^{-1}. In our experiment, the variations determined by cultivars and moisture levels may be compared with the cited paper in relative terms, although the average NAR was only 5 g m^{-2} d^{-1}. However, in sweet and fibre sorghum a similar NAR (~7 g m^{-2} d^{-1}) was registered over a 130-day growth in well watered field plots (Dolciotti et al., 1998), while in grain sorghum a significant response to drought was observed despite a very low NAR (average, 1.5 g m^{-2} d^{-1}) (Younis et al., 2000). Therefore it appears that trait variations within each experiment are meaningful despite large differences among study cases. Specific leaf area was reduced by 66% when shifting from 70% to 50% FC (Table 2), whereas in the literature the decrease was less than 20% when skipping one irrigation out of five (Amal et al., 2010) and even negligible when cutting water supply by one third (Bullock et al., 1991). In our experiment, NAR and SLA proved to be good indicators of genotype performance also at low moisture, deserving to be employed in programmes of sorghum breeding for a wide range of environmental conditions. Leaf water status outlined a consistent picture among LWP, RWC and LS. Leaf succulence is associated with forage palatability (Marten, 1978): a threshold of 10 g water mm^{-1}, which is desirable to boost forage intake, was easily passed by JS-2002 and Chakwal Sorghum but barely attained by JS-263 (Fig. 1.c). This proves the inadequacy of an old cultivar to face drought also in terms of forage quality. Leaf water potential and relative water content are often addressed in the

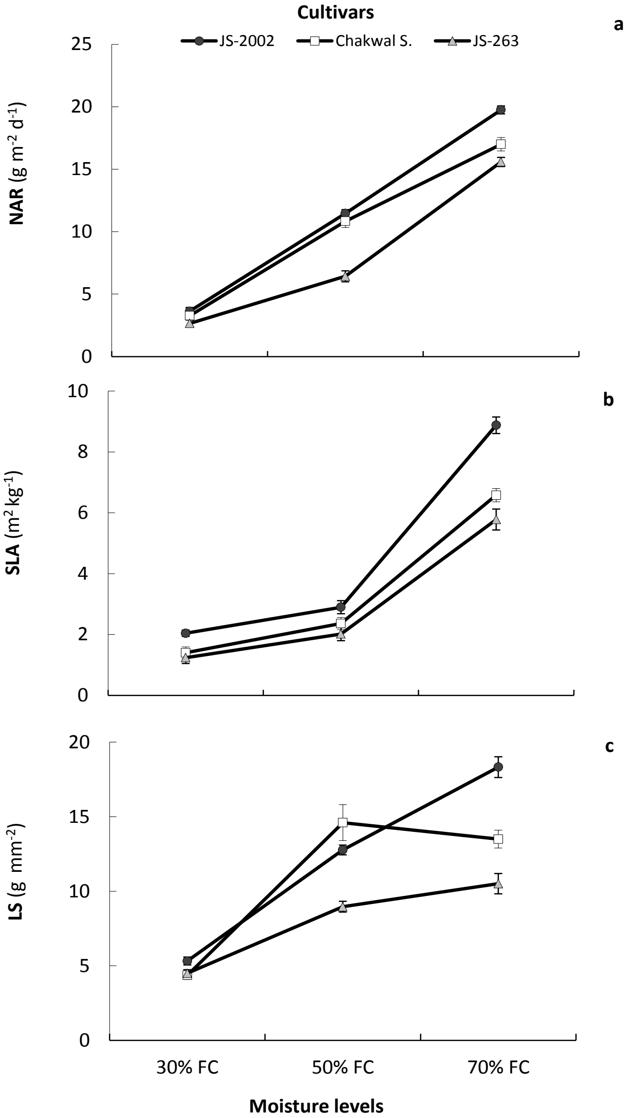

Fig 1. Significant Cultivar × Moisture interactions in functional and leaf water traits of forage sorghum grown in a controlled environment: a, Net assimilation rate (NAR); b, Specific leaf area (SLA); c, Leaf succulence (LS). Error bars represent the standard error (n = 5). FC, field capacity.

Table 2. Effects of cultivars and soil moisture levels on functional traits and leaf water status of forage sorghum grown in a controlled environment.

Source	NAR (g m^{-2} d^{-1})	SLA (m^{2} kg^{-1})	LWP (MPa)	RWC (%)	LS (g mm^{-2})
Cultivar (CV)					
JS-2002	5.8 a	4.6 a	-1.55 b	78 a	12.1 a
Chakwal S.	5.2 b	3.4 b	-1.47 a	74 b	10.8 b
JS-263	4.1 c	3.0 c	-1.84 c	71 c	8.0 c
P	<0.001**	<0.001**	<0.001**	<0.001**	<0.001**
Moisture level (ML)					
30% FC	1.6 c	1.6 c	-1.84 c	62 c	4.7 c
50% FC	4.8 b	2.4 b	-1.61 b	74 b	12.1 b
70% FC	8.7 a	7.1 a	-1.40 a	87 a	14.1 a
P	<0.001**	<0.001**	<0.001**	<0.001**	<0.001**
CV × ML					
P	<0.001**	<0.001**	0.466 ns	0.521 ns	<0.001**
C.V. (%)	8.6	13.7	4.2	5.7	12.9

ns, * and ** mean non-significant, significant at $P \leq 0.05$ and $P \leq 0.01$, respectively. Different letters indicate significantly-different means (SNK test; $P \leq 0.05$). FC, field capacity; LS, leaf succulence; LWP, leaf water potential; NAR, net assimilation rate; RWC, relative water content; SLA, specific leaf area.

literature: in grain sorghum at DAS 61, a treatment at -50% water supply determined LWP and RWC decreases analogous to those observed in our experiment between 70% and 30% FC (Tsuji et al., 2003). A similar decline in LWP was recorded in another grain sorghum experiment after two months at -51% available moisture (Singh and Singh, 1995). In contrast to this, in sweet sorghum only a modest variation in LWP and RWC occurred at 30% vs. 70% FC at DAS 60 (Tingting et al., 2010). This may be explained by the fact that the plant was still in the vegetative stage, involving a certain resilience to drought. Stronger decreases in either LWP or RWC were seldom observed in the literature: in a field test among 10 sorghum genotypes, JS-2002 scored a RWC of only 40% (Ali et al., 2009). In another experiment dealing with grain sorghum, LWP dropped from about -1 MPa to less than -2 MPa after 60 days at -33% water supply (Berenguer and Faci, 2001). It thus appears that in our experiment both LWP and RWC were very responsive to genotypes and water, providing a sensitive indication of plant moisture status.

Plant biomass and water effects

In the literature, the interaction between sorghum cultivars and moisture levels describes a variable effect on biomass traits. Varieties performing better at high water availability were often surpassed by other genotypes at low water availability (Younis et al., 2000; Aishah et al., 2011). Conversely, in our experiment the gain of the two modern varieties at high moisture was only diminished but not reversed at low moisture (Fig. 2.b). Younis et al. (2000) also found that RDW was less affected by drought than SDW, leading to a significant increase in the R:S ratio as in our experiment. This is consistent with the enhanced role of the root apparatus under moisture or nutrient deficiency. Water use efficiency revealed a very modest output of biomass in exchange for the consumptive amount of water (Table 3). In forage sorghum, WUE levels up to 7 and 8.5 g l^{-1} were recorded by Aishah et al. (2011) and Saeed and El-Nadi (1998), respectively; however, levels below 2 g l^{-1} were observed by Singh and Singh (1995). In grain sorghum, WUE levels of the sole grain portion between 0.2 and 1.5 g l^{-1} were recorded by Farré and Faci (2006); between 1 and 1.9 g l^{-1} by Abdel-Motagally (2010). Therefore it appears that a large inter-specific variation affects this trait. Moreover, all the

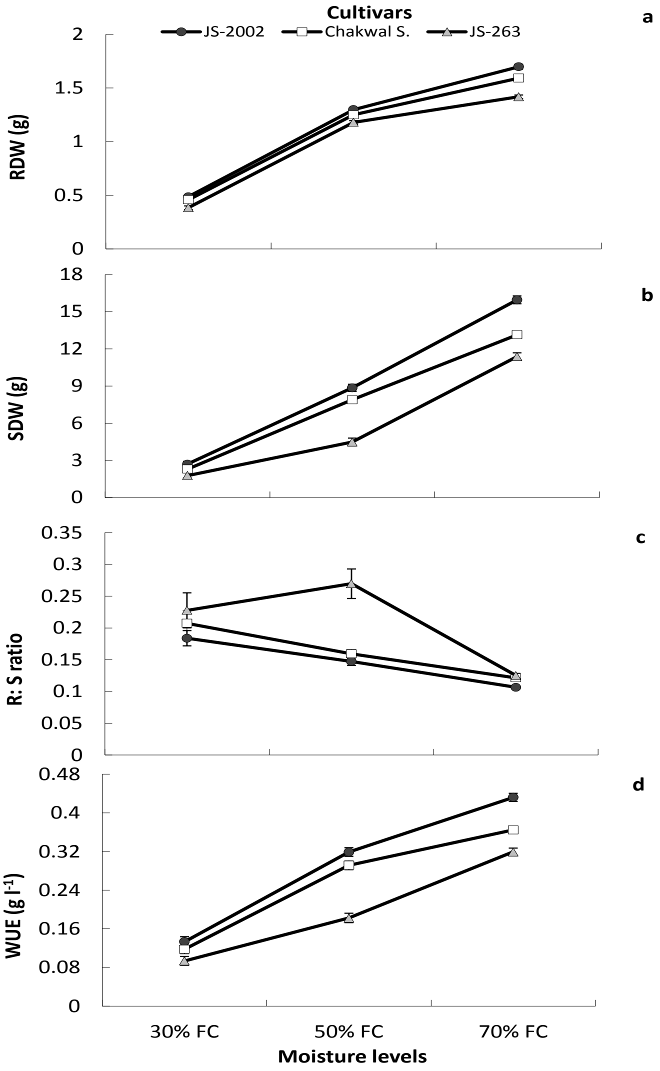

Fig 2. Significant Cultivar × Moisture interactions in biomass traits of forage sorghum grown in a controlled environment: a, Root dry weight (RDW); b, Shoot dry weight (SDW); c, Root to shoot (R:S) ratio; d, Water use efficiency (WUE). Error bars represent the standard error (n = 5). FC, field capacity.

Table 3. Effects of cultivars and soil moisture levels on biomass traits and water effects of forage sorghum grown in a controlled environment.

Source	RDW (g pot^{-1})	SDW (g pot^{-1})	TDW (g pot^{-1})	R:S (g g^{-1})	WUE (g l^{-1})	Ky
Cultivar (CV)						
JS-2002	1.16 a	9.17 a	10.33 a	0.15 b	0.29 a	1.98 b
Chakwal S.	1.09 b	7.77 b	8.87 b	0.16 b	0.26 b	1.84 b
JS-263	0.99 c	5.88 c	6.87 c	0.21 a	0.20 c	2.30 a
P	<0.001**	<0.001**	<0.001**	<0.001**	<0.001**	<0.001**
Moisture level (ML)						
30% FC	0.44 c	2.26 c	2.70 c	0.21 a	0.11 c	
50% FC	1.24 b	7.07 b	8.32 b	0.19 a	0.26 b	
70% FC	1.57 a	13.49 a	15.06 a	0.12 b	0.37 a	
P	<0.001**	<0.001**	<0.001**	<0.001**	<0.001**	
CV × ML						
P	<0.001**	<0.001**	<0.001**	<0.004*	<0.001**	
C.V. (%)	5.0	8.3	7.3	19.3	8.1	12.8

ns, * and ** mean non-significant, significant at $P \leq 0.05$ and $P \leq 0.01$, respectively. Different letters indicate significantly-different means (SNK test; $P \leq 0.05$). FC, field capacity; Ky, yield response factor to water supply; RDW, root dry weight; R:S, root to shoot ratio; SDW, shoot dry weight; TDW, total dry weight; WUE, water use efficiency.

Table 4. Correlations (r) among functional, leaf water and biomass traits of forage sorghum cultivars grown at variable soil moisture in a controlled environment.

	NAR	SLA	LWP	RWC	LS	RDW	SDW	TDW	R:S
NAR	1								
SLA	0.911	1							
LWP	0.810	0.714	1						
RWC	0.914	0.838	0.782	1					
LS	0.879	0.722	0.781	0.806	1				
RDW	0.933	0.797	0.778	0.890	0.888	1			
SDW	0.996	0.926	0.818	0.908	0.876	0.924	1		
TDW	0.997	0.920	0.819	0.912	0.882	0.937	0.999	1	
R:S	-0.768	-0.684	-0.682	-0.663	-0.602	-0.563	-0.771	-0.757	1
WUE	0.979	0.853	0.841	0.889	0.917	0.939	0.982	0.984	-0.779

Correlation between RDW and R:S, significant at $P \leq 0.05$; all the other correlations, significant at $P \leq 0.01$; n = 45.
LS, leaf succulence; LWP, leaf water potential; NAR, net assimilation rate; RDW, root dry weight; R:S, root to shoot ratio; SDW, shoot dry weight; SLA, specific leaf area; TDW, total dry weight; WUE, water use efficiency.

cited experiments were carried out in field plots, which may at least partially explain the low WUE observed in our pot experiment. Water use efficiency is another point of concern: in sorghum, WUE is either seen to increase at decreasing moisture (Singh and Singh, 1995; Abdel-Motagally, 2010; Aishah et al., 2011), or the opposite (Saeed and El-Nadi, 1998; Farré and Faci, 2006; Ahmed et al., 2007) as in our experiment. Singh and Singh (1995) reported the oddest case, as WUE rose up to a 43% decrease in available moisture, then fell at a 51% decrease. There is no apparent clue in the cited papers as to why WUE either rises or falls. However, it may be speculated that until the restriction in water supply is severe, sorghum can make a more efficient use of moisture and restrain the loss of yield, enhancing WUE. This could explain the contrasting result of Singh and Singh (1995), who observed an increase in the trait as long as grain yield smoothly decreased, followed by a sudden WUE loss when yield sharply fell. This explanation holds true for all the cited papers except one (Farré and Faci, 2006) and fits our data. We recorded the most relevant yield loss (-83%) among the cited papers, in exchange for a less than proportional restraint in water supply (-43%). In our experiment biomass accumulation was therefore a somewhat stronger determinant of WUE (r = 0.982** with SDW) than water use (r = 0.916**; not shown), in agreement with the findings of Xin et al. (2009). Reference Ky in grain sorghum is set at 0.9 (Doorenbos and Kassam, 1979), meaning that relative yield decrease is expected to be 10% less than relative evapotranspiration deficit. Values between 0.9 and 1 are reported in the literature for sweet sorghum (Perniola et al., 1992; Curt et al., 1995). In our experiment, we found much higher levels (average, 2.04), which is consistent with the low WUE levels. However, the ranking of the three cultivars is consistent with their overall behaviour and Ky may represent a comprehensive indicator of their fitness to face drought.

Materials and methods

Site and conditions

The study was conducted in earthen pots in a controlled environment at the Department of Agronomy, Pir Mehr Ali Shah - Arid Agriculture University of Rawalpindi (33° 6’ N; 73° 07’ E; 488 m a.s.l.), Pakistan, in summer 2009. The soil was brought from the University Research Farm, air dried, ground and subjected to the determination of field capacity (FC): the saturated soil paste was prepared and transferred to a porous Buckner funnel for the removal of surplus water. The residual amount of water, representing FC, was determined by the gravimetric method (48 h at 105 °C) (Anderson and Ingram, 1993). In our soil, FC was 408 ml kg^{-1} dry soil. On June 27, 2009, each pot was filled with 10 kg of dry soil and seeded (10 seeds pot^{-1}). Fifteen days after seeding (DAS), the seedlings were thinned to three plants per pot. The pot surface was covered with aluminium foil to prevent soil temperature increases. During the experiment, the pots were kept in a laterally open polyethylene tunnel; the air could freely circulate to keep the temperature at par inside and outside the tunnel. During the experiment (60 days), minima and maxima daily temperatures consistently averaged 23.8 ± 2.7 °C and 36.5 ± 3.4 °C, respectively.

Experimental treatments and design

Treatments consisted of three levels of soil moisture (30%, 50% and 70% FC) in a cross combination with three cultivars of forage sorghum approved by the Punjab Seed Council: one old variety (JS-263, approved in 1968) which has become very popular among farmers and is commonly known as Local Sorghum; two modern varieties (JS-2002 and Chakwal Sorghum, approved in 2002 and 2008, respectively). JS-263 (Ayub Agricultural Research Institute, Faisalabad, Pakistan) is a medium cycle, medium high variety, semi-resistant to drought, with loose panicle and bold, whitish grain. JS-2002 (Fodder Research Institute, Sargodha, Pakistan) is a late variety remaining green for a long period, with small/compact panicle and yellow grain. Chakwal Sorghum (Barani Agricultural Research Institute, Chakwal, Pakistan) is a medium cycle variety resistant to drought with loose panicle and whitish grain. The three moisture levels were monitored every other day through the gravimetric method (~10 g soil samples taken from the pots, oven dried at 105 °C). During the experiment, an average of 24.7, 33.5 and 43.3 l of water pot^{-1} were supplied in the 30%, 50% and 70% FC treatments, respectively. The nine treatment combinations (3 cultivars x 3 moisture levels) were arranged in a completely randomized factorial design with 5 replicates, totalling 45 pots. Recommended doses of nitrogen and phosphorus fertilizers were applied during pot filling at the equivalent rates of 60 and 30 kg ha^{-1} of N and P_2O_5, respectively.

Data collection

Plant height (m) was measured on one tagged plant per pot from the base to the tip of the highest fully expanded leaf at three growth stages: 5th leaf (DAS 25), pre-booting (DAS 40) and 50% heading (DAS 60). At harvest (DAS 60), the number of leaves plant^{-1} (Leaves pl.$^{-1}$) was counted on the tagged plant, and green leaf area (LA; m^2) was determined on the tagged plant by means of a leaf area meter (CI-202L, Forestry Suppliers, Jackson, MS, USA). Two functional traits of plant growth were assessed at harvest (DAS 60) according to Hunt (1978): i) net assimilation rate (NAR; g m^{-2} d^{-1}), expressing the efficiency in dry biomass accumulation per unit leaf surface per day of growth; ii) specific leaf area (SLA; m^2 kg^{-1}), indicating leaf expansion per unit dry weight. Leaf water potential (LWP; MPa) was measured at DAS 60 on a fully expanded leaf from each pot, using the pressure bomb apparatus (Decagon Devices, Pullman, WA, USA). Relative water content (RWC; %) was determined on the same leaf used to measure LWP: a fresh leaf sample was cut into a small disc and the fresh leaf weight (FW) was measured. The leaf sample was then soaked in distilled water (15 ml tube) for 4 hours in the dark; thereafter, the turgid leaf weight (TW) was measured. Finally, leaf dry weight (DW) was obtained after drying the leaf at 80 °C for 24 hours. RWC was calculated according to Smart and Bingham (1974):

$$\mathrm{RWC} = \frac{\mathrm{FW} - \mathrm{DW}}{\mathrm{TW} - \mathrm{DW}} \times 100$$

Based on previous data, leaf succulence (LS; g mm^{-2}) was calculated as: (FW – DW)/LA.

Plants from each pot were harvested at 50% heading (DAS 60). Fresh weight of roots and shoots was assessed. The root and shoot material was then dried at 65 °C for 72 hours and the dry weight of roots (RDW), shoots (SDW) and their total (TDW) was recorded; the root to shoot (R:S) ratio was calculated on a dry weight basis. Water use efficiency (WUE; g l^{-1}) was assessed as the ratio between SDW and the total supply of water (Passioura, 1977). The yield response factor to water (Ky) was calculated according to Doorenbos and Kassam (1979), as the ratio between relative yield decrease (1 - Ya/Ym) and relative evapo-transpiration deficit (1 - ETa/ETm); where Ya/Ym is actual/maximum yield; ETa/ETm is actual/maximum evapo-transpiration. SDW and water supply in the 70% FC treatment were assumed as Ym and ETm, respectively; SDW and water supply in the combined 30% and 50% FC treatments were assumed as Ya and ETa, respectively. Ky was therefore only assessed for the variety factor.

Statistical analysis

Normal distribution and equal variance of data were controlled through the Kolmogorov-Smirnov and Bartlett tests, respectively. Data were then submitted to analysis of variance (ANOVA) through the CoStat 6.3 software (CoHort Software, Monterey, CA, USA): in each trait the significance of the investigated sources (cultivars, moisture levels and their interaction) was determined. The Scott - Newman-Keuls test ($P \leq 0.05$) was adopted to separate means of statistically significant sources. Pearson's correlation (r) was assessed among functional, leaf water and biomass traits.

Conclusion

In forage sorghum, the choice of a suitable variety remains a fundamental strategy to cope with drought. In severe soil pot conditions, the three tested cultivars showed different abilities to withstand the effects of water shortage. The two recent varieties (JS-2002 and Chakwal Sorghum) outperformed the old one (JS-263) under all viewpoints. In fact, the same ranking was consistently repeated in plant morphology, functional growth, leaf water status, biomass yield and water effects. This means that in terms of general behaviour a top performer at high moisture like JS-2002 has a competitive edge over JS-263 that is reduced, not reversed at low moisture. This finding is seldom echoed in the literature, contrasting the belief that in many cases old varieties and landraces are better suited to face drought than modern varieties at high potential under favourable circumstances. In this light, our results support the drive to replace old genotypes, although specific field tests are needed to validate pot trial results and assess actual yield gains in varying crop conditions. Our work thus promotes further research at laboratory, greenhouse and field plot level, in the quest for higher and consistent sorghum performances.

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