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Full Length Research Paper

Response of pearl millet (*Pennistetum glaucum*) cultivars to post-flowering drought stress

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Two pearl millet (*Pennisetum glaucum* (L.) R. Br.) cultivars (Dadda and Shella) were evaluated at post-flowering stage for drought tolerance based on morpho-physiological criteria. The experiment was laid down in a randomized complete block design which comprised of a combination of two factors (cultivars and three water stress regimes; (well watered (36%), moderately stressed (21%) and severely stressed (9%)). Data on morpho-physiological variables such as total green leaf area, relative water content, potential quantum yield, root/ shoot ratio and yield per panicle revealed significant (P ≤ 0.05) differences between the cultivars at severely water stress treatment. Dadda showed the maximum relative water content (RWC) (45.70±1.13%) than Shella with the RWC of (32.00±1.06%) under SS. Under SS moisture level Dadda showed the maximum potential quantum yield of 0.77 μmolm^{-2}s^{-1} than Shella, 0.69 μmolm^{-2}s^{-1}. Dadda showed better drought stress tolerance than Shella in terns of relative water content, potential quantum yield, yield per panicle and root/ shoot ratio under sever water stress. The analysis of growth revealed the importance of total green leaf area, relative water content, potential quantum yield, yield per panicle and root/ shoot ratio as markers for drought tolerance during post-flowering stage in Pearl millet cultivars.

Key words: Cultivars, morpho-physiological criteria, pearl millet, post-flowering.

INTRODUCTION

Pearl millet (*Pennisetum glaucoma* (L.) R. Br.) is the sixth most important coarse-grain cereal grown in semi-arid tropical regions of Asia and Africa (Gari, 2002). In Ethiopia, the crop has been cultivated for its importance as food and fodder for animals and used in ritual activities. It has diverse adaptation mechanisms to grow and survive under relatively marginal environments. How-ever, under subsistence farming conditions in drought prone areas of the world, the crop has revealed low grain yield which was 500-700 Kgha^{-1} (Rai et al., 1999). Currently, drought is one of the most important limiting factors for crop production and becoming an increasingly severe problem in many regions of the world (Aslam et al., 2006). Moreover, the situation is currently exacer-bated due to the increasing population with high demand for food and the possible global climate change scenarios (Morison et al., 2008). This could lead to a decline in overall crop productions by affecting various aspects of plant growth (Rahman et al., 2004) and physiological pro-cesses (Hall et al., 1990). To advance crop productivity in such drought prone areas, it is necessary to understand the mechanism of plant responses with the ultimate goal of improving crop performance. Although the effects of water stress on growth and yield of maize (Aslam and Tahir, 2003; Rahman et al, 2004), wheat (Ahmad et al., 2003) and tropical legumes (Kumaga et al., 2003) have been studied during the last decade very little work has been done to study the effects of drought stress on cultivars of pearl millet in Ethiopia. On the other hand, many reports have indicated that, selection of drought stress tolerant plant species/cultivars would have econo-mic and efficient means of utilizing drought-prone areas (Turner, 1997). Elucidating variations and modifications in morpho-physiological traits under different drought stress

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Abbreviations: TGLA, Total green leaf area; **WW,** well watered; **MS,** moderately stressed; **SS,** severely stressed; **RWC,** relative water content; **PSII,** photosystem two; **RSR,** root /shootbiomass ratio; **FW,** fresh weight; **TW,** turgid weight; **DW,** dry weight.

Table 1. Physico-chemical properties of the soil used during the experiment.

Physico-chemical properties	Values
Bulk density	1.99 gm cm^{-3}
Soil moisture content	20%
Carbon content	6.3%,
pH	6.7

levels is crucial in improving yield under water limiting conditions. Moreover, it has been reported that identification at post-flowering drought tolerance under water deficit condition was crucial (Rauf and Sadaqat, 2008). Local cultivars are still the backbone of agricultural production in developing countries, which are adapted to various environments and preferred by farmers for many useful agronomic properties under stressful conditions (Brush, 1999), where the new developed cultivars are less reliable. Therefore, the present work was, therefore carried out to evaluate the post-flowering drought performance of Pearl millet's cultivars.

MATERIALS AND METHODS

Plant materials

Seeds of pearl millet cultivars were obtained from the collection made by College of Dry land Agriculture in collaboration with Biology Department of Mekelle University in the northern part of Ethiopia. The seeds were collected as the primary step for further crop improvement researches. Eight seeds of each Pearl millet [*Pennisetum glaucum* (L.) R. Br.] cultivars; Dadda and Shella were sown. Seedlings were completely emerged 7 days after sowing and seedlings with uniform vigor and height were retained while the remaining five were eliminated.

Growth conditions

A greenhouse experiment was carried out at College of Dry land Agriculture of Mekelle University, Northern Ethiopia (13°30'N and 39°29'E, 2,300 m above sea level) from January 2007 to May 2007. The assay was conducted under an average relative humidity of 43.23% and average minimum and maximum temperatures of 9.78 and 31.81°C, respectively.

Experimental design

The experiment was laid out in randomized complete block design with two factors (cultivars and water regimes) and three replications. The cultivars were randomly assigned within each block. Thirty-six plastic pots were used. Each pot was filled with 13 ± 0.05 Kg mixture of soil (Vertisol according to USAID classification), sand and manure in a proportion of 2:1:1, respectively. The physico-chemical nature of the mixed soil is summarized in Table 1. The field capacity of the mixed soil in the pot was determined following Abdallah El-Khoshiban (2010). This soil moisture level was maintained at the field capacity (referred as the control (WW)) till the plants reached a flowering stage. Following flowering stage plants were, watered to WW, moderately stressed (MS), 50% of WW and severely stressed (SS), 15% of WW. These moisture levels after measurements made using a soil moisture meter (Delta-T Devices Ltd., Cambridge, UK) were 36, 21 and 9% in the WW, MS and SS, respectively. The soil moisture levels were kept constant by restoring the amount of water lost within 48 h.

Measured parameters

Total green leaf area (TGLA)

TGLA was determined using a formula:

TGLA=Length x Width x 0.79

The correction factor 0.79 was obtained as the ratio of the average actual green leaf areas of 15 samples of each cultivar determined using leaf area meter (Area meter, AM 100 Analytical Development Company Ltd., UK) to the average green leaf areas of 15 samples of each cultivar taken by multiplying length and width.

Relative water content

Relative water content (RWC) was determined following the method used by Abdalla and El-khoshiban (2007). The second leaves from the tip were cut and placed in tagged polythene bags. Fresh weights (W_f) for 36 discs from the youngest fully expanded leaves were determined within an hour after excision. The discs were made turgid by soaking in distilled water for 24 h. the discs of leaves were kept in a chamber with light source of 6wm^{-2} to prevent the loss of weight by respiration (Turner, 1981). Subsequently, turgid weight (W_t) was determined after blotting with tissue paper. Dry weight (W_d) was obtained after drying the discs for 48 h at 70°C in an oven. Then RWC was computed using the following equation:

$$RWC = \frac{W_f - W_d \text{ X } 100}{W_t - W_d}$$

Chlorophyll fluorescence

Youngest and fully expanded leaves were selected to determine the chlorophyll fluorescence/the potential quantum yield using Plant Efficiency Analyzer (Hansatech Instruments Ltd., England). The leaves were covered with clips and kept in dark for 30 mn before measurements. The transients were induced by red light of 3000 µmol m^{-2} s^{-1}, which focused on the sample surface to give homogenous illumination over exposed area of sample surface and maximal quantum yield of photosystem two (PS II) was measured(Ashraf et al., 2007). Leaves of hundred and eight plants were considered during the measurement 70 days after germination or 12 days after flowering.

Root to shoot biomass ratio determination

Plants were harvested and separated into shoot and root parts. Subsequently the parts were dried at 80°C in oven until constant dry weight was obtained. Consequently the dry weights were determined using a triple balance. Root to shoot ratio was determined based on dry shoot and root biomass.

Yield/panicle

Dried panicles with mature seeds were obtained from each plant.

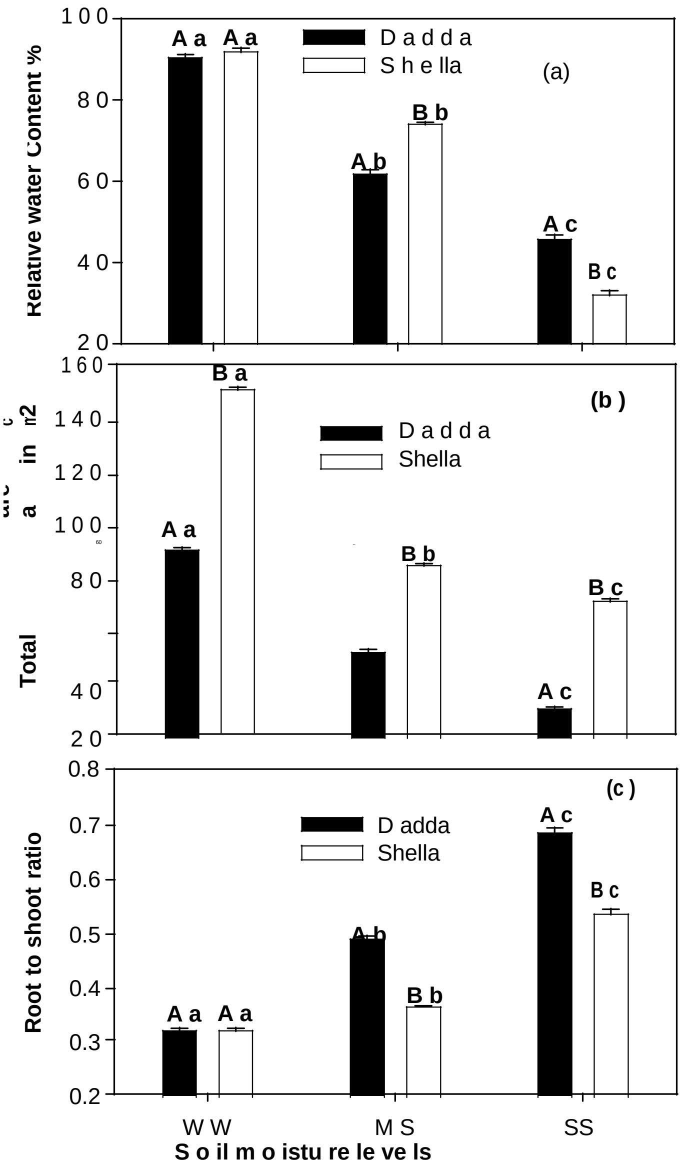

Figure 1. Relative water content (a), total green leaf area (b) and root to shoot ratio (c) of two cultivars of Pearl millet subjected to three water stress levels. Means followed by different lower case letters show significant variation among soil moisture treatment levels and different upper case letters show significant variation between cultivars within the same soil moisture level (P≤ 0.05). Bars represent the mean ±standard error.

Subsequently, the seeds were threshed mechanically (using hand). Grain yield/panicle was determined weighing the total seeds per panicle.

Statistical analysis

To determine the significance level of the treatments effect, data were subjected to analysis of variance using SPSS Version 17 (SPSS Inc). Treatment and varieties means were separated using the least significant differences test at 5% level of significance. Graphs were generated using Sigma Plot 8.0 (Systat Software, Inc.).

Table 2. Yield per panicle (g) of two farmer cultivars of Pearl millet under three soil moisture levels.

Varieties	Yield/panicle(g) Soil moisture levels		
	WW	MS	SS
Dadda	110.31Aa	86.01Bb	65.43Ac
Shella	160.65 Ba	96.67Bb	43.27Bc

*Means followed by different lower case letters show significant variation among water stress treatment levels and different upper case letters show significant variation between cultivars within the same water stress treatment level (P<0.05).

RESULTS

Relative water content and total green leaf area

RWC decreased significantly ($p \leq 0.05$) while the severity of stress level increased. There was no significant difference between the cultivars on WW; in contrast significant variation in RWC was noted in stressed water conditions (Figure 1a). Dadda showed the maximum RWC (45.70±1.13%) than Shella with the RWC of (32.00±1.06%) under SS.

Beside to this with an increasing in the intensity of water stress the tested cultivars showed a significant reduction in total green leaf area (TGLA) ($p \leq 0.05$) (Figure 1b). Also Shella showed the maximum TGLA across the three moisture levels. Under SS Shella and Dadda recorded the TGLA of (72.27±0.93) cm^2 and (31.50± 0.76) cm^2, respectively.

Root to shoot biomass ratio and yield per panicle

There was a significant difference in root to shoot ratio (RSR) of MS and SS plants at P<0.05 (Figure 1c). Under SS and MS Dadda exhibited maximum RSR than Shella. The tested cultivars showed a significant increase in RSR as water stress increased under the roots of plants; however, there was no significant variation in RSR ratio between the cultivars studied at the control treatment WW.

The performance of cultivars was variable according to the incidence of drought (Table 2). Both of the Dadda and Shella showed a great reduction in yield per panicle at the soil moisture levels studied. The highest (160.65 g) yield/panicle under the WW condition was obtained by the cultivar Shella, while the lowest (110.31 g) was produced by Dadda. However, under SS condition Dadda exhibited a highest yield/panicle of 65.43 g than Shella 43.27 g.

Potential quantum yield

The potential quantum yield was not significantly different between varieties in the WW and MS water supply

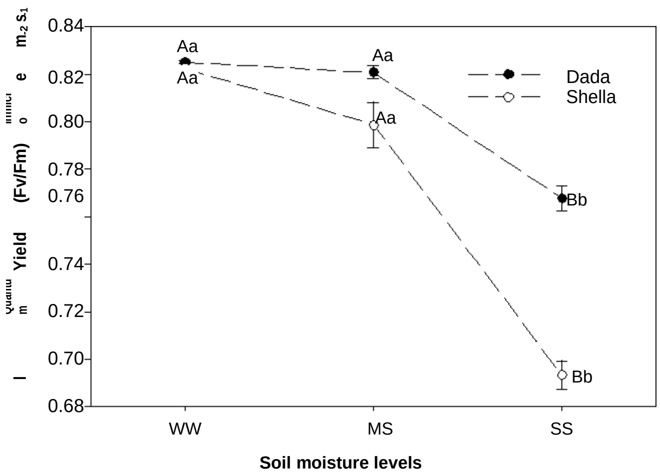

Figure 2. Potential quantum yield of two cultivars of Pearl millet subjected to three soil moisture levels. Means followed by different lower case letters show significant variation among soil moisture treatment levels and different upper case letters show significant variation between cultivars within the same soil moisture level (P≤ 0.05).Bars represent the mean ± standard error.

regimes (Figure 2). However, the varieties revealed significant reduction under severe water deficit condition. Under SS moisture level Dadda showed the maximum potential quantum yield of 0.77µmolm^{-2}s^{-1} than Shella which was 0.69 µmolm^{-2}s^{-1}.

DISCUSSION

The reduction in RWC observed is in agreement with reports made on wheat (Siddique et al., 2000) and rice varieties (Pirdashti et al., 2009).The decline might be triggered by water deficit in the soil as a consequence of water lost through the stomata. The genotypic variation in RWC under water stress level could be the difference in adaptation of the cultivars (Abdalla and El-khoshiban, 2007). Cultivars which maintain adequate leaf RWC can be in general considered as suitable for dry regions and are more tolerant to drought conditions and plasmolysis (Ahmadi and Siosemarideh, 2005). Detrimental effect of reduction of RWC on physiological and biochemical reactions and consequently on growth and productivity is well documented (Lawlor, 1995). Moreover, Khan et al. (2007) evaluated the physiological traits depending on the tolerance against drought stress in broad bean and reported that the RWC significantly was decreased during the stress and the tolerant varieties possessed the higher RWC than the non tolerant ones. In this regard Dadda found to be suitable; our results has also confirmed that measuring RWC is a potential tool for screening genotype under various degrees of water stress.

Similarly reduction in TGLA has been reported in soybean (Zhang et al., 2004), pearl millet (Winkel et al., 2001) and finger millet (Muhammad and Azam, 2007). How-ever, the response varies depending on genotypes and intensity and duration of water stress (Abdalla and El-Khoshiban, 2007). Likewise, Shella maintained its TGLA at SS than Dadda on the same water stress level. Such a reduction in TGLA might ascribe to water shortage which in turn led to a decrease in cell turgor and eventually cell growth. Crop cultivars with lower leaf area during post-flowering stage are considered as suitable for drought prone areas. Moreover, the difference in TGLA is probably a result of the greater evaporative demand differences in these cultivars.

Root to shoot ratio and Yield per panicle

The RSR raise under severe water deficit condition could be due to the shifts in photoassimilates which favor roots growth than the shoots (Bota et al., 2004). In addition, it might be due to water deficit which promote lesser bio-mass allocation to the shoot than to roots (Anwar et al., 2003; Salem, 2003). It is very possible that Dadda was more efficient than Shella in absorption and providing water to the top of the plant, a mechanism could lead to larger area of leaves (Spollen et al., 2000).

Similarly a study conducted on other pearl millet varieties also showed grains weight/panicle was reduced considerably by water stress (Mahalakshmi and Bidinger, 1986); nevertheless, the resistant cultivars showed superiority in grain yield over the susceptible ones (Khanna et al., 1994). Thus in this regard Dadda is high yielding and tolerant under severely water deficit con-dition. The decline in yield per panicle can be attributed to a decrease in source capacity which led to the reduction of seed weight, since water stress during seed develop-ment affect irreversibly the sink demand of panicles in pearl millet (Winkel et al., 1997). Moreover, the decrease in yield per panicle is associated with the effect of water stress during grain filling and loss of seed size (Seghatoleslami et al., 2008).Yadav et al. (1999) also demonstrated that water stress after pollination in pearl millet reduces seed yield through reduction of seed weight per panicle.

The present study support the suggestion that, water stress during post-flowering stage in crop plants had a direct effect on PSII photochemistry (Ashraf et al., 2007). Thus, water stress induced reduction in potential quantum yield is indicative of photoinhibition associated with an over-reduction of PSII (Piradshti et al., 2009; Praba et al., 2009). Under severely water stress regime Dadda showed the maximum potential quantum yield. The ability of this cultivar in maintaining high potential quantum yield under water deficit condition might indicate high efficiency of using radiation for photochemistry and carbon assimilation (Massaci and Jones, 1990). Moreover, measuring potential quantum yield has been considered as the most reliable screening tool for abiotic stresses like drought (Massaci and Jones, 1990). We found that Dadda showed better drought stress tolerance than shella. The results of the present study demonstrated the importance of total green leaf area, relative water content, potential quantum yield and root to shoot ratio as parameters for screening drought resistance during post-flowering stage. Further studies on other physiological and biochemical aspects of these cultivars under water stress conditions are still required.

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