Phenotypic plasticity of root system and shoots of *Sorghum bicolor* under different soil water levels during pre-flowering stage

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Abstract

In order to understand possible relationships among the features of plant organs located above and below soil, their correlation and how it might affect strategies for drought adaptation, two sorghum lines contrasting in drought tolerance (BR007-sensitive and 99100-tolerant) were cultivated under different soil water levels during pre-flowering stage. Half of the plants of each genotype remained under daily irrigation to maintain the soil moisture close to FC, and the other half was subjected to WD. After 10 days in these conditions, physiological and anatomical characteristics of the lines were evaluated and then water supply was restored and maintained at optimum levels by the end of the cycle. At physiological maturity, the root morphology and agronomic parameters associated to productivity were analyzed. Regardless the variation in the soil moisture, plants of the line 99100 presented values significantly superior for HI, RDB and LA compared to plants of the BR007 line, whereas the *A* values were lower. Only line 99100 plants increased VFRL compared to its counterparts under FC. The lack of change in root morphology in line BR007 plants was accompanied by increase in CAT activity and lower DBV in leaves. The increase in CAT activity was not sufficient to reduce the pool of H_2O_2 in leave cells of BR007 under WD, leading to a decrease in F_vF_m. Thus, the H_2O_2 accumulated in the leaf was deviated to the lignin biosynthesis, corroborating the lower DVB in leaves of BR007 line under WD, compared to line 99100 under WD. In addition, BR007 plants showed smaller DBC under WD, which simultaneously led to an increase in NBC. Such structural and functional adjustment at the leaf level would compensate the absence of changes in the root architecture in response to the stress generated by the WD.

Keywords: Antioxidant enzymes; chlorophyll-*a* fluorescence; gas exchange; micromorphometry; WinRhizo.
Abbreviations: *A*_Photosynthesis rate; ABE_Abaxial Epidermis thickness; ADE_Adaxial Epidermis thickness; APX_Ascorbate Peroxidase; BSD_Abaxial Stomatal Density; BSI_Abaxial Stomatal Index; CAB_Thickness of Abaxial Cuticle; CAD_Thickness of Adaxial Cuticle; CAT_Catalase activity; C_i_Internal CO_2 concentration; DBC_Polar Diameter of Bulliform Cells; DSD_Adaxial Stomatal Density; DSI_Adaxial Stomatal Index; DVB_Distance between Vascular Bundles; *E*_Transpiration rate; FC_Field Capacity; FLO_Phloem area; FRL_Fine Roots Length; FRSA_Fine Roots Superficial Area; FRV_Fine Roots Volume; F_v/F_m_Ratio between variable and maximum fluorescence of the photosystem II; g_s_Stomatal conductance; HI_Harvest Index; LA_Leaf Area; MXD_Metaxylem Diameter; NBC_Number of Bulliform Cells; PDB_Panicle Dry Biomass; POD_Guaiacol peroxidase; RDM_Root Dry Biomass; TDB_Total Dry Biomass; TL_Thickness of Lamina; TORL_Total Roots Length; TORSA_Total Roots Superficial Area; TORV_Total Roots Volume; TRL_Thick Roots Length; TRSA_Thick Roots Superficial Area; TRV_Thick Roots Volume; VFRL_Very Fine Roots Length; VFRSA_Very Fine Roots Superficial Area; VFRV_Very Fine Roots Volume; WD_Water Deficit.

Introduction

Originally from the tropical Africa, sorghum [*Sorghum bicolor* (Moench) L.] is the cereal most adapted to drought, and is a source of food for over 500 million people in 98 countries (Pennizi, 2009). In Brazil, this cereal is traditionally used as raw material for animal feed and for ethanol production from the early 80`s (Dutra et al., 2013). The sorghum genome (~720 bp) was completely sequenced and available in public databases (Pennizi, 2009). Thereafter, the complete description of each transcription unit has become a major challenge not only to understand the biology of this cereal, but also to know as it tolerates drought (Pennizi, 2009). Nowadays, it is known that sorghum has structural and morphophysiological evolutionary features that increase the efficiency of water use, and also extra-copies of several genes associated to adaptation to drought (Paterson et al., 2009).

Currently, two main strategies for adapting to drought have been proposed, each one with its specificities, tolerance and avoidance (Lopes et al., 2011; Fang and Xiong, 2015). Drought tolerance is characterized by continuous maintenance of the physiological processes under low water availability. Drought tolerant species can reduce xylem cavitation (Cochard et al., 2008; Souza et al., 2013) and the rate of oxidation of macromolecules by the reduction/elimination of reactive oxygen species (Mittler, 2002), without changing the variables of leaf gas exchange (Horta et al., 2014). It may also accumulate solutes to maintain cell turgidity in relatively low water potentials

(Mutava et al., 2011; Horta et al., 2014). Drought avoidance, in turn, can be achieved by maximizing the access to water (Uga et al., 2013) and minimizing the loss to the atmosphere (Singh and Reddy, 2011). Features associated with this second strategy include increase in root system biomass, high specific root length, small leaf area and efficient stomatal control of transpiration. The choice for one or another adaptive strategy; however, depends on the ability of the plant to adjust its phenotype properly and with the right speed.

Phenotypic plasticity, defined as the ability of a genotype to produce different phenotypes under different environmental conditions (Valladares et al., 2000), has rarely been studied connected to strategies of drought adaption in sorghum. The relationship between the allocation of carbon and reduced water utilization has been studied mainly in organs located above ground, with particular emphasis to the leaves (Liu et al., 2010; Dugas et al., 2011; Mutava et al., 2011). According to the functional balance theory (Brouwer, 1962), plants increase the biomass allocation to the shoots if the carbon gain is affected by limiting resources above the ground, such as light and CO_2. Similarly, the plants increase the biomass allocation to the roots when subjected to low levels of resources below ground, i.e. water and nutrients (Poorter and Nagel, 2000). Despite this flexibility of biomass allocation to capture and store, organs constitute a key point in the coordination of morphological, anatomic and physiological changes in the leaf in environments with low water availability in the soil (Davies and Zhang, 1991). So, little is known about this relationship (Fort et al., 2012).

Adaptation to drought requires coordination between the organs of plant located above and under the soil (Hajek et al., 2013). Under drought condition, the adaptive strategies of the organs located under the soil overpass the above-soil organs (Fort et al., 2012), and variations occur in the different characteristics of the same species (Liu et al., 2010). The present study addressed features of phenotypic plasticity in the root system and shoots of two sorghum lines cultivated under different soil water levels during pre-flowering stage. The main objective of this study was to identify the possible dependence relationship between the features of plant organs located above and below soil, how strong is the relationship, and how it would affect the adaptive strategies in this species.

Results

Drought tolerance in sorghum 99100 line is related to increase in Very Fine Roots Length (VFRL)

Despite the variation in the soil water levels, plants of grain sorghum line 99100 showed higher RDM compared to the BR007 (Table 1). The TORL (Fig.1) of the plants of 99100 line exposed to WD was increased compared to values obtained for plants of the same line when the soil moisture was maintained near FC. On the other hand, the roots of plants BR007 showed no significant differences for these variables under WD, compared to the control at FC (Fig. 1). Since in plants of line 99100 under WD the TORL was greater and the RDM was not changed compared to the control plants, this increase was associated with the occurrence of fine roots, corroborated by the morphometric image analysis of these roots, previously scanned in scanner table using the WinRhizo program.

The total LA was also higher in 99100 line than in the BR007, and both lines showed significantly lower LA values under WD (Table 1). Furthermore, an increase in TDB value was observed for plants under WD in both lines compared to the control. However, this increase was much more pronounced in the 99100 line than in BR007 (Table 1). The plants of line 99100 also presented 45.8% higher PDB compared to those observed in BR007 plants (Table 1) dissociated of the reduction in biomass of this reproductive structure in both lines under WD. Consequently, the HI of the line 99100 was 54.6% higher (on average) compared to the line BR007. Under WD, regardless of lineage, the HI was reduced 63.4% on average compared to the values observed in the respective controls (Table 1).

The lack of change in root morphology in plants of line BR007 under WD was compensated by structural and functional adjustment at the leaf level

The A, g_s and E were significantly higher in BR007 plants compared to the values observed in leaves of plants of the line 99100, while the C_i did not change (Table 2). A positive correlation was found between A and E (r² = 0.96), affecting water use efficiency of these lineages at different levels of water. Under WD conditions both lines presented values of A, g_s and E similar to the respective controls; however, C_i was tended to decrease (Table 2).

The F_v/F_m in control plants of both lines and in 99100 plants exposed to WD was 0.77, and only in leaves of plants of the BR007 the F_v/F_m ratio was decreased (Table 2). Enzyme assays identified significant differences in the CAT enzyme activity only in leaves of the BR007 line, depending on the level of soil water, which was greater under WD (Table 2). This increase was not associated to significant changes in the activities of APX and POD (Table 2).

Regarding the structural characteristics, leaves from plants of line 99100 presented thicker CAB and ABE, higher DSD and DSI, fewer NBC and DVB when compared to BR007 line (Table 3). In WD conditions, an increase of 35.6% in the NCB was observed in the 99100 line compared to control (Table 3). However, the MXD and FLO presented similar values in control plants of both lines and in the plants of line 99100 exposed to WD, and only in the leaves of plants BR007 under WD, which showed small size in such area (Table 3). In addition, only the BR007 plants under WD adjusted their leaf structure via reduction of the DVB, and in the NBC and DBC, compared to their respective controls (Table 3).

Discussion

Only few studies have focused on the characteristics of roots associated with leaves traits and productivity, in plants grown under drought (Uga et al., 2013; Horta et al., 2014). Uga et al. (2013) reported that with altering the structure such as change in the root angle and growth, plants may exploit more water from the deeper layers of the soil (using drought avoidance strategy), allowing significant improvement in A and grain yield. In the present study, a root elongation was observed mainly in the fine roots of plants in line 99100. It is important to notice that only plants with the conservative characteristics from the arid environments present enhancement in the efficiency of capture of water by producing very fine roots (Liu et al., 2010). It is suggested; therefore, that this line presents a strategy belowground not only of drought avoidance, but also conservative of the water absorbed from the soil. Apparently, this is the greatest divergence between the lines.

Stomatal closure and decrease in A are initial symptoms of dehydration of leaves, and is frequently observed in C4 plants (Maroco et al., 1997; Massad et al., 2007), such as sorghum

Table 1. Production parameters in two sorghum lines under different levels of water in the soil (field capacity – FC, and water deficit – WD) (n=6)

Parameter	BR007		99100	
	FC	WD	FC	WD
RDM	7.100 aB	6.360 aB	10.21 aA	9.410 aA
PDB	5.140 aB	4.460 bB	7.870 aA	7.800 aA
TDB	24.53 aA	29.98 aB	22.60 bA	37.88 aA
HI	0.210 aB	0.149 bB	0.348 aA	0.206 bA
LA	0.100 aB	0.044 bB	0.191 aA	0.130 bA

Abbreviations/unit: Root dry biomass (RDM, g); panicle dry biomass (PDB, g); total dry biomass (TDB, g), harvest index (HI, g. g^{-1}) and leaf area (LA, m²). Means followed by the same letter are not statistically different from each other. Uppercase letters denote comparisons between genotypes within the same soil water level, and lowercase letters denote comparisons between the soil water levels within the same genotype. Means were compared by Scott-Knott test at 5% probability.
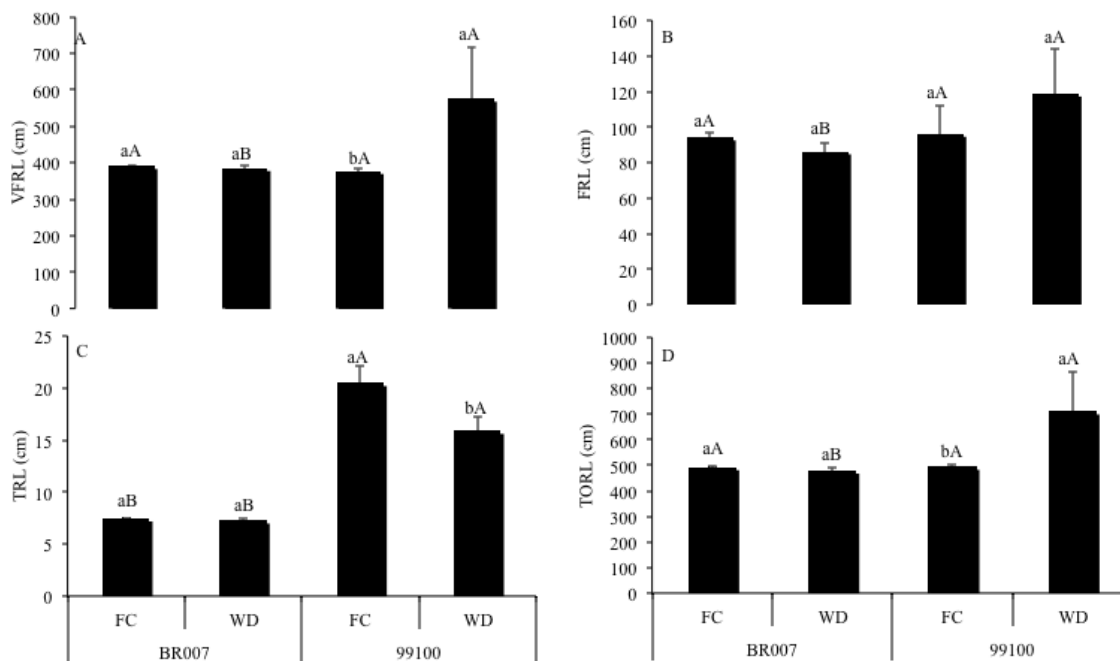
Fig 1. Morphometric characterization of root length (A-D) in two sorghum lines under different levels of water in the soil (field capacity – FC, and water deficit – WD) (n=3). Abbreviations/unit: Length of very fine roots (VFRL), fine (FRL), thick (TRL) and total (TORL). Uppercase letters denote comparisons between genotypes within the same soil water level, and lowercase letters denote comparisons between the soil water levels within the same genotype. Means were compared by Scott-Knott test at 5% probability.

(Xie and Su, 2012; Zegada-Lizarazu and Monti, 2013), that associated with decreases in C_i liable to cause an increase in the PR. In this study, a reduction in LA was observed in plants of both lines grown under WD, dissociated from changes in A, g_s and C_i. Unlike expected, the A was greatly increased, especially in leaves from line BR007. According to the theory proposed by Davies and Zhang (1991), signals originated from the root system could affect g_s, and the ability to move water efficiently to long distances is crucial to maintain cell turgor and gas exchange under WD (Ocheltree et al., 2014). In addition, it is possible that, in the present study, part of the glucose originated in photosynthesis (and also in the starch breakdown) has been directed toward the biosynthesis of sucrose in leaves, increasing cell osmolarity. Zegada-Lizarazu and Monti (2013) observed that the accumulation of compatible solutes is very important in mechanisms of protection to the stress generated by the WD in mature sorghum plants, since the increase in the concentration of sugars would prevent photo-oxidative destruction of reaction center of PSII in the leaves. Nevertheless, a decrease in the ratio F_v/F_m and an increased

activity of CAT were observed in plants of BR007 line under WD. Under WD, the PR can be considered as an adaptive mechanism against the production of toxic levels of reactive oxygen by the consumption of photo-assimilates during photosynthesis, favoring the availability of CO_2 to the chloroplast, and also maintaining a high consumption of electrons and ATP for carbon fixation (Zhang and Kirkham, 1996). In addition, PR increases glycolate oxidase activity, which in turn, stimulates the H_2O_2 production. It is possible that the increase in CAT activity was not sufficient to reduce the pool of H_2O_2 in leaf cells of BR007 line under WD. Another possibility is the deviation of the H_2O_2 accumulated in the leaf to the lignin biosynthesis; thus, corroborating the lower DVB in leaves of this line under WD, compared to the other treatments. The increased lignification of leaf cells prevents excessive water loss and xylem cavitation (Cochard et al., 2008; Tyree and Sperry, 1988). In addition, plants of BR007 showed smaller DBC under WD, which could cause an increase in leaf rolling. However, loss of leaf turgor occurred simultaneously to an increase in the density of bulliform cells. In grasses, the bulliform cells are specifically

Table 2. Parameters of gas exchange and chlorophyll-a fluorescence obtained *in situ* and enzymes of the antioxidant system in two sorghum lines under different levels of water in the soil (field capacity – FC, and water deficit – WD) (n=6).

Parameter	BR007		99100	
	FC	WD	FC	WD
A	31.05 aA	33.28 aA	22.03 aB	23.57 aB
g_s	0.295 aA	0.265 aA	0.216 aB	0.210 aB
E	5.710 aA	5.710 aA	4.850 aB	4.850 aB
C_i	142.2 aA	106.4 aA	181.8 aA	132.0 aA
F_vF_m	0.760 aA	0.720 bB	0.770 aA	0.770 aA
CAT	0.039 bA	0.076 aA	0.047 aA	0.069 aA
APX	1.256 aA	1.256 aA	0.699 aA	0.699 aA
POD	0.092 aA	0.092 aA	0.077 aA	0.077 aA

Abbreviations/unit: Photosynthesis rate (A, μmol CO_2 $m^{-2}s^{-1}$), stomatal conductance (g_s, mol H_2O $m^{-2}s^{-1}$); transpiration rate (E, mol H_2O $m^{-2}s^{-1}$); internal CO_2 concentration (C_i, μmol CO_2 $m^{-2}s^{-1}$); ratio between variable and maximum fluorescence of the photosystem II 2 (F_v/F_m); catalase activity (CAT, μmol H_2O_2.min^{-1} mg $protein^{-1}$); ascorbate peroxidase (APX, μmol de ascorbato min^{-1} mg $proteína^{-1}$); guaiacol peroxidase (POD, μmol guaiacol min^{-1} mg $protein^{-1}$). Uppercase letters denote comparisons between genotypes within the same soil water level, and lowercase letters denote comparisons between the soil water levels within the same genotype. Means were compared by Scott-Knott test at 5% probability.
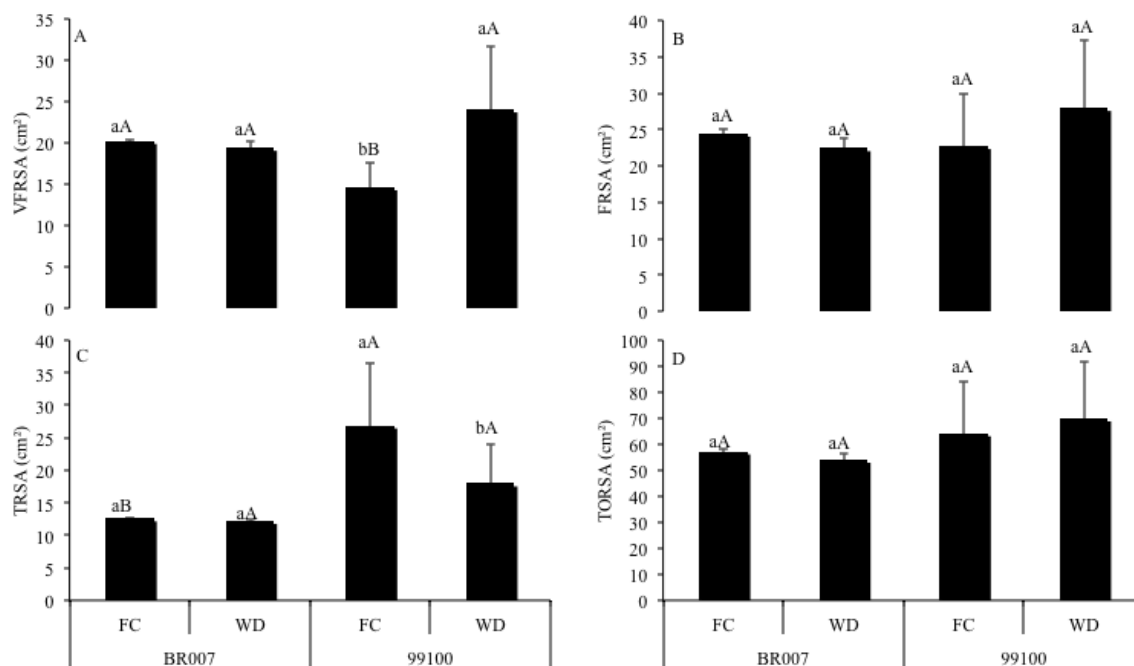
Fig 2. Morphometric characterization of surface area (A-D) of roots in two sorghum lines under different levels of water in the soil (field capacity - FC, and water deficit - WD) (n=3). Abbreviations/unit: Superficial area of very fine roots (VFRSA), fine (FRSA), thick (TRSA) and total (TORSA). Uppercase letters denote comparisons between genotypes within the same soil water level, and lowercase letters denote comparisons between the soil water levels within the same genotype. Means were compared by Scott-Knott test at 5% probability.

located in the adaxial side of the leaves, and when plants are exposed to WD. These cells eliminate water inducing an adaxial leaf curl (Li et al., 2010). Such events would equalize the absence of changes in the root architecture in response to the stress generated by the WD.

In turn, the expansion of the root system associated with enhance in the VFRL of plants of 99100 line under WD increased the capture of water by the plant, eliminating the need for osmotic adjustment in tissues of this organ. This fact allows *per se* the reduction of oxidative stress and the accumulation of large amounts of soluble carbohydrates in leaf cells for the purpose of osmotic adjustment.

Materials and Methods

Plant material and cultivation conditions

The experiment was carried out during the months of November and February 2010, under greenhouse conditions

at the National Maize and Sorghum Research Center (EMBRAPA), Sete Lagoas, Minas Gerais, Brazil (altitude 732 m, latitude South 19°28', longitude West 44°15'). The average maximum and minimum temperature recorded during the growth period were 29°C and 23°C, respectively. The relative humidity ranged from 54% to 79%. Plant material consisted of two sorghum lines (*Sorghum bicolor* L. Moench) contrasting to drought tolerance, BR007 and 99100. The line 99100 is more tolerant to WD than BR007.

Plants were grown in plastic pots containing 20 kg of typical dystrophic Red Latosol soil previously adjusted with limestone and fertilized with NPK + Zinc (8-28-16). The water content in the soil was monitored daily between 9:00 a.m. and 3:00 p.m., with a moisture sensor (GB Reader N1535)(Measurement Engineering, Australia) installed at the center of each pot with the aid of a screw auger at a depth of 20 cm. These sensors detect the water content in the soil based on electrical resistance and are coupled to digital meters. Water replacement by irrigation was based on the

Table 3. Leaf anatomic characterization in two sorghum lines under different levels of water in the soil (field capacity - FC, and water deficit - WD) (n=6).

| | BR007 | | 99100 | |
	FC	WD	FC	WD
ADE	56.70 aA	56.70 aA	61.46 aA	61.46 aA
ABE	49.21 aB	49.21 aB	57.03 aA	57.03 aA
CAD	10.68 aB	10.68 aB	14.05 aA	14.05 aA
CAB	9.420 aB	9.420 aB	14.20 aA	14.20 aA
TL	96.97 aA	88.48 bA	103.1 aA	65.12 bB
NBC	26.00 bA	29.00 bA	23.00 bB	35.00 aA
DBC	78.87 aA	68.50 aA	58.78 bA	57.94 aA
DVB	169.9 aA	150.3 aB	94.98 bA	86.00 bA
MXD	87.12 aA	72.73 bB	74.61 aA	78.48 aA
FLO	7689 aA	5914 bB	7696 aA	7635 Aa
DSI	8.560 aB	8.560 aB	11.98 aA	11.98 aA
BSI	15.94 aA	15.94 aA	14.38 aA	14.38 aA
DSD	50.00 aB	50.00 aB	72.00 aA	72.00 aA
BSD	79.00 Aa	79.00 aA	83.00 aA	83.00 aA

Abbreviations: Thickness of adaxial epidermis (ADE, µm), thickness of abaxial epidermis (ABE, µm), thickness of adaxial cuticle (CAD, µm); thickness of abaxial cuticle (CAB, µm); thickness of lamina (TL, µm); number of bulliform cells (NBC, nº); polar diameter of bulliform cells (DBC, µm); distance between vascular bundles (DVB, µm); metaxylem diameter (MXD, µm); phloem area (FLO, µm²); adaxial stomatal index (DSI, [(stomata number. mm^{-2})/epidemic cells number]*100); abaxial stomatal index (BSI, [(stomata number. mm^{-2})/epidemic cells number]*100); adaxial stomatal density (DSD, stomata number.mm^{-2}) and abaxial stomatal density (BSD, stomata number.mm^{-2}). Uppercase letters denote comparisons between genotypes within the same soil water level, and lowercase letters denote comparisons between the soil water levels within the same genotype. Means were compared by Scott-Knott test at 5% probability.
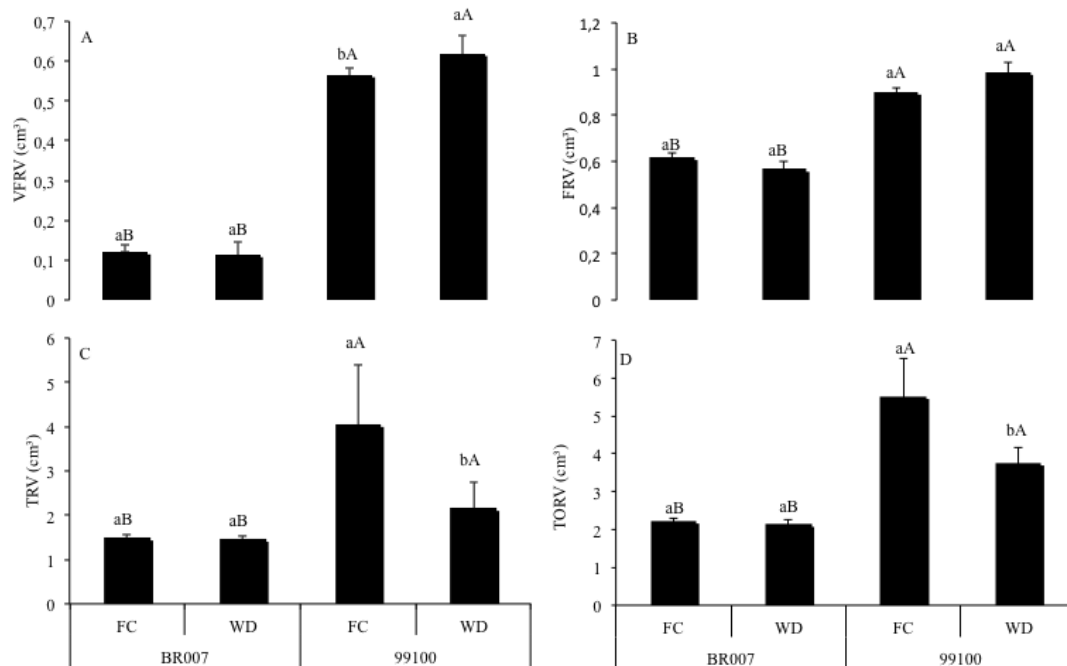
Fig 3. Morphometric characterization of volume (A-D) of roots in two sorghum lines under different levels of water in the soil (field capacity - FC, and water deficit - WD) (n=3). Abbreviations/unit: Volume of very fine roots (VFRV), fine (FRV), thick (TRV) and total (TORV). Uppercase letters denote comparisons between genotypes within the same soil water level, and lowercase letters denote comparisons between the soil water levels within the same genotype. Means were compared by Scott-Knott test at 5% probability.

data obtained with the sensor and water was added to reach FC during the period preceding the imposition of the treatments. The water replacement calculations were performed with a spreadsheet and based on a soil water retention curve. In parallel, all necessary cultural and phytosanitary treatments were performed.

At the pre-flowering stage, half of each initial treatment was subjected to WD. The other part remained under daily irrigation in order to maintain the soil moisture close to FC, i.e., soil water tension at –18 kPa (Souza et al., 2013; Souza et al., 2014). The WD was reached by applying 50% of the total available water until soil water tension becomes at least –100 kPa (Souza et al., 2013; Souza et al., 2014). After 10

days under these conditions, physiological and anatomical characteristics of the lines were evaluated and water supply was restored and maintained at optimum levels by the end of the cycle. At physiological maturity, the root morphology and agronomic parameters associated to productivity were analyzed.

Physiological evaluation

Gas exchange an chlorophyll-a fluorescence

The *A* and *E* were measured using an infrared gas analyzer (IRGA–Infrared Gas Analyzer), model LI 6400 (LI-COR,

Lincoln, NE, USA). The C_i and g_s were calculated by the equipment from the values of A and E (Faquhar et al., 1980). Measurements were made between 09:00 and 10:00 am under artificial photosynthetic active radiation of 1500 μmol photons m^{-2} s^{-1} at leaf level, with 21% O_2 and 400 μmol CO_2 mol^{-1} air. The F_v/F_m was evaluated by the saturating pulse method, using a fluorimeter with modulated pulse amplitude (Plant Efficiency Analyser, Hansatech Instruments King's Lynn, UK) at the same time of gas exchange evaluations.

Enzymatic assays

The activity of the enzymes of the antioxidant system from homogenized plant material was determined: CAT (EC 1.11.1.6), APX (EC 1.11.1.11) and POD (EC 1.11.1.7) in a medium containing potassium phosphate buffer 0.1M (pH 6.8), 0.1 mM EDTA, 1 mM DTT, 1 mM PMSF and 1% PVPP (w/v). Additional details are described in Pereira et al. (2010). The activity of these enzymes was expressed as units (U) per amount of protein. Each U corresponds to the oxidation of 1 μmol H_2O_2 min^{-1} to CAT, 1 μmol ascorbate min^{-1} for the APX and 1μmol guaiacol min^{-1} for POD.

Anatomical evaluation

The anatomical characteristics of leaves were measured as described by Souza et al. (2010). Images were obtained with an Olympus BX-60 photomicroscope and micromorpho-metric analysis was performed using the UTHSCSA ImageTool (University of Texas, San Antonio, USA).

Agronomic parameters and root morphology evaluation

Plants were partitioned into stem, leaves, roots and reproductive parts and dried in an oven with forced air circulation at 70°C for 72 h. The grain dry weight and total dry biomass was used to estimate HI (grain dry weight/total dry biomass). The LA was measured with a LICOR area meter (Model LI-3100, Li-Cor, Lincoln, NE, USA).

   The computerized system WinRhizo Pro 2007a (Regent Instruments, Sainte-Foy, QC, Canada) was used for evaluations of the root system. For this purpose, roots were scanned using a professional scanner (Epson, EXPRESSION 10000 XL, Epson America, Inc., Long Beach, CA, USA), equipped with an additional light source. The definition of 400 dpi was used to capture images. The root morphology were measured according to Magalhães et al. (2011).

Statistical analysis

Treatments were evaluated in a 2×2 factorial design, with two sorghum lines (BR007-sensitive and 99100-tolerant) and two levels of soil moisture (control and water deficit), totaling four treatments. The experimental unit was the pot containing two plants, with six replications per treatment. Results were subjected to analysis of variance followed by Scott-Knott test at 5% probability.

Conclusion

The results showed that the sorghum lines BR007 and 99100 present similar tolerances to dehydration of leaf tissues, besides the different strategies of adaptation to drought. Plants of 99100 present characteristics of drought avoidance in the roots, in addition to drought tolerance. Such divergence demanded differential plasticity in the leaf structure and functionality of leaves in plants of BR007 exposed to WD, which was not necessary in leaves of the tolerant line 99100, in most cases.

References

Brouwer R (1962) Distribution of dry matter in the plant. Neth J Agr Sci. 10: 399-408.

Cochard H, Barigah ST, Kleinhentz M, Eshel A (2008) Is xylem cavitation resistance a relevant criterion for screening drought resistance among *Prunus* species? Plant Physiol. 165: 976-82.

Davies WJ, Zhang J (1991) Root signals and the regulation of growth and development of plants in drying soil. Annu Rev Plant Phys. 42: 55-76.

Dugas DV, Monaco MK, Olsen A, Klein RR, Kumari S, Ware D, Klein PE (2011) Functional annotation of the transcriptome of *Sorghum bicolor* in response to osmotic stress and abscisic acid. BMC Genomic. 12: 514.

Dutra ED, Barbosa-Neto AG, Souza RB, Morais-Jr MA, Tabosa JN, Menezes RS (2013) Ethanol production from the stem juice of different sweet sorghum cultivars in the state of Pernambuco, northeast of Brazil. Sugar Tech. 15: 316-321.

Fang Y, Xion L (2015) General mechanisms of drought response and their application in drought resistance improvement in plants. Cell Mol Life Sci. 72: 673-689.

Farquhar GD, von Caemmerer S, Berry JA (1980) A biochemical model of photosynthetic CO_2 assimilation in leaves of C3 species. Planta. 149: 78-90.

Fort F, Jouany C, Cruz P (2012) Root and leaf functional trait relations in Poaceae species: implications of differing resource-acquisition strategies. Plant Ecol. 2: 1-9.

Hajek P, Hertel D, Leuschner C (2013) Intraspecific variation in root and leaf traits and leaf-root traits linkages in eight aspen demes (*Populus tremula* and *P. tremuloides*). Plant Sci. 4: 1-11.

Horta LP, Braga MR, Lemos-Filho JP, Modolo LV (2014) Organ-coordinated response of early-post-germination mahogany seedlings to drought. Tree Physiol. 0: 1-12.

Li L, Shi ZY, Li L, Shen GZ, Wang XQ, An LS, Zhang JL (2010) Overexpression of *ACL1* (abaxially curled leaf 1) increased bulliform cells and induced abaxial curling of leaf blades in rice. Mol Physiol. 3: 807-817.

Liu G, Frescher GT, Pan X, Cornelissen JHC, Li Y, Dong M (2010) Coordinated variation in leaf and root traits across multiple spatial scales in Chinese semi-arid and arid ecosystems. New Phytol. 188: 543-553.

Lopes MS, Araus JL, van Heerden PDR, Foyer CH (2011) Enhancing drought tolerance in C4 crops. J Exp Bot. 0: 1-19.

Magalhães PC, Souza TC, Cantão FRO (2011) Early evaluation of root morphology of maize genotypes under phosphorus deficiency. Plant Soil Environ. 57: 135-138.

Maroco JP, Ku MSB, Edwards GE (1997) Oxygen sensitivity of C4 photosynthesis: evidence from gas exchange and chlorophyll fluorescence analyses with different C4 subtypes. Plant Cell Environ. 20: 1525-1533.

Massad RS, Tuzet A, Bethenod O (2007) The effect of temperature on C4-type leaf photosynthesis parameters. Plant Cell Environ. 30: 1191-1204.

Mittler R (2002) Oxidative stress, antioxidants and stress tolerance. Trends Plant Sci. 7: 405-410.

Mukberjee SP, Choudhuri MA (1983) Implications of water stress-induced changes in the levels of endogenous ascorbic acid and hydrogen peroxide in *Vigna* seedlings. Physiol Plantarum. 58: 166-170.

Mutava RN, Prasad PVV, Tuinstra MR, Kofoid KD, Yu J (2011) Characterization of sorghum genotypes for traits related to drought tolerance. Field Crop Res. 123: 10-18.

Ocheltree TW, Nippert JB, Kirkham MB, Vara P, Prasad V (2014) Partitioning hydraulic resistance in *Sorghum bicolor* leaves reveals unique correlations with stomatal conductance during drought. Funct Plant Biol. 41: 25-36.

Paterson AH, Bowers JE, Bruggmann R, Dubchak I, Grimwood J, Gundlach H, Haberer G, Hellsten U, Mitros T, Poliakov A, Schmutz J, Spannagl M, Tang H, Wang X, Wicker T, Bharti AK, Chapman J, Feltus FA, Gowik U, Grigoriev IV, Lyons E, Maher CA, Martis M, Narechania A, Otillar RP, Penning BW, Salamov AA, Wang Y, Zhang L, Carpita NC, Freeling M, Gingle AR, Hash T, Keller B, Klein P, Kresovich S, McCann MC, Ming R, Peterson DG, Rahman M, Ware D, Westhoff P, Mayer KFX, Messing J, Rokhsar DS (2009) The Sorghum bicolor genome and the diversification of grasses. Nature. 457: 551-556.

Pennizi E (2009) How sorghum withstands heat and drought. Science. 323: 573.

Pereira FJ, Magalhães PC, Castro EM, Souza TC, Alves JD (2010) Antioxidant system activity and aerenchyma formation in 'Saracura' maize roots. Pesqui Agropecu Bras. 45: 450-456.

Poorter H, Nagel O (2000) The role of biomass allocation in the growth responses of plants to control levels of light, CO_2 and water: a quantitative review. Aust J Plant Physiol. 27: 595-607.

Singh SK, Reddy KR (2011) Regulation of photosynthesis, fluorescence, stomatal conductance and water-use efficiency of cowpea *(Vigna unguiculata* (L.) Walp.) under drought. J Photoch Photobio B. 105: 40-50.

Souza TC, Magalhães PC, Pereira FJ, Castro EM, Silva-Jr JM, Parentoni SN (2010) Leaf plasticity in successive selection cycles of 'Saracura' maize in response to periodic soil flooding. Pesqui Agropecu Bras. 45: 16-24.

Souza TC, Magalhães PC, Castro EM, Albuquerque PEP, Marabesi MA (2013) The influence of ABA on water relation, photosynthesis parameters, and chlorophyll fluorescence under drought conditions in two maize hybrids with contrasting drought resistance. Acta Physiol Plant. 35: 515-527.

Souza TC, Magalhães PC, Castro EM, Portilho NP, Padilha FA, Gomes CJC (2014) ABA application to maize hybrids contrasting for drought tolerance: changes in water parameters and in antioxidant enzyme activity. Acta Physiol Plant. 73: 205-217.

Souza TC, Castro EM, Magalhães PC, Lino LO, Alves EA, Albuquerque PEP (2013) Morphophysiology, morphoanatomy, and grain yield under field conditions for two maize hybrids with contrasting response to drought stress. Acta Physiol Plant. 35: 3201-3211.

Tyree MT, Sperry JS (1988) Do woody plants operate near the point of catastrophic xylem dysfunction caused by dynamic water stress? Plant Physiol. 88: 574-580.

Uga Y, Sugimoto K, Satoshi OS, Rane J, Ishitani M, Hara N, Kitomi Y, Inukai Y, Ono K, Kanno N, Inoue H, Takehisa H, Motoyama R, Nagamura Y, Wu J, Matsumoto T, Takai T, Okuno K, Yano M (2013) Control of root system architecture by *DEEPER ROOTING 1* increases rice yield under drought conditions. Nature Gen. 45: 1097-1105.

Valladares F, Wright SJ, Lasso E, Kitajima K, Pearcy RW (2000) Plastic phenotypic response to light of 16 congeneric shrubs from a Panamanian rainforest. Ecol. 81: 1925-1936.

Xie T, Su P (2012) Canopy and leaf photosynthetic characteristics and water use efficiency of sweet sorghum under drought stress. Russ J Plant Physiol. 59: 224-234.

Zegada-Lizarazu W, Monti A (2013) Photosynthetic response of sweet sorghum to drought and re-watering at different growth stages. Physiol Plantarum 159: 56-66.

Zhang J, Kirkham B (1996) Antioxidant responses to drought in sunflower and sorghum seedlings. New Phytol. 132: 361-373.