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Morphophysiological indicators of drought tolerance in sorghum hybrids[1]

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ABSTRACT - The objective of this study was to assess the responses of sorghum hybrids to drought and select the most significant morphophysiological indicators to differentiate the hybrids grown under water deficit and well-watered conditions. Two field experiments were conducted simultaneously, one under well-watered and the other under water deficit conditions after the pre-flowering stage, evaluating four contrasting grain sorghum hybrids (DKB 540, BRS 310, BRS 332, and 50A10), in a randomized block experimental design with four replications. Means were subjected to individual and joint analysis of variance, and the effects of water conditions and sorghum hybrids were compared using the F-test ($p < 0.05$) and the Tukey's test ($p \leq 0.05$), respectively. Multivariate canonical variable analysis and Pearson correlation were also applied. Water deficit significantly reduced grain yield in 23.9%. The higher grain yields of the evaluated sorghum hybrids are associated with the higher relative chlorophyll content, photosynthetically active leaf area, panicle diameter and mass, grain mass per panicle, threshing index, and 100-grain mass, and the lower number of lodged plants. Grain mass per panicle, 100-grain mass, panicle mass, number of lodged plants, relative chlorophyll content, and leaf area are the most important indicators for explaining drought tolerance variations in grain sorghum hybrids grown under water deficit and well-watered conditions.

Key words: *Sorghum bicolor* (L.) Moench. Water deficit. Multivariate analysis.

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INTRODUCTION

Water scarcity is one of the main yield-limiting factors of cultivated plants worldwide. Variations in rainfall patterns and high air temperatures, mainly due to climate change, impose intense water stress on agricultural crops, leading to significant risks to global food and energy security (Fahad *et al*., 2017).

In addition to the unpredictable effects associated to droughts, the rapid global population growth requires the development of technologies to meet food demands, posing significant challenges for researchers to maximize crop yields under environments prone to water deficit (Molotoks; Smith; Dawson, 2021; Woldesemayat *et al*., 2018).

Of the cereals with the greatest potential for coexisting with drought, sorghum (*Sorghum bicolor* (L.) Moench) stands out, due to its adaptive capacity to stress conditions and potential for planting in the off-season, a growing season more prone to water restrictions. These characteristics have been attributed factors such as the evolution of the C4 photosynthetic pathway, the plant anatomical and morphophysiological structures, and physical and biochemical processes (Lopes *et al*., 2011; Magalhães *et al*., 2016; Pardo; Vanburen, 2021).

To cope with reduced water availability, plants display a complex series of water stress escape and tolerance mechanisms (Yadav; Sharma, 2016). These mechanisms interact and operate hierarchically, involving various plant structures, and affect crop yield under water stress conditions (Barnabás *et al*., 2008).

Information on sensitivity or tolerance responses to water stress, integrated with water-soil-plant-environment relations is essential for the development and enhancement of the scientific basis for adapting to and managing drought conditions. Additionally, it can aid in refining selection indices and developing technological solutions for sustainable agriculture to benefit society (Anjum *et al*., 2011; Pires *et al*., 2020). However, the effects of these parameters on different crop groups and drought conditions have not been fully elucidated (Zhang *et al*., 2018).

Research on this subject generates a large amount of information and require the use of tools for analyses to condense the results, eliminate variables with lower contributions, and select the most important indicators for explaining drought tolerance variations. In this context, Moreira *et al*. (2020) proposed a statistical approach using multivariate canonical variable analysis.

Therefore, considering the demand for technologies for adapting to drought conditions, the objective of this study was to assess the responses of sorghum hybrids to drought and select the most significant morphophysiological indicators to differentiate the hybrids grown under water deficit and well-watered conditions.

MATERIAL AND METHODS

Experimental area

The experiment was conducted at the Gorutuba Experimental Station of the Brazilian Agricultural Research Corporation (Embrapa Maize and Sorghum), in Nova Porteirinha, northern Minas Gerais (MG), Brazil. The experimental area is at the geographic coordinates: 15°45’25.5”S, 43°16’53.2”W, and an altitude of 524 m. The region’s climate is Aw, a tropical climate characterized by rainy summers and dry winters, according to the Köppen-Geiger classification (Alvares *et al*., 2013). This region is strategic for conducting water deficit studies due to its predictable drought occurrences throughout the year, as evidenced by historical data and confirmed during the experimental period (Figure 1A, B).

The soil of the experimental area was classified as *Latossolo Vermelho-Amarelo* (Typic Hapludox) (Santos *et al*., 2018) with medium texture. The main characteristics of the 0-20 cm soil layer were: pH in water = 5.8; organic matter = 1.6%; P = 18.1 mg dm^{-3}; K = 302.5 mg dm^{-3}; Ca = 2.9 $cmol_c$ dm^{-3}; Mg = 0.56 $cmol_c$ dm^{-3}; Al = 0.06 $cmol_c$ dm^{-3}; Cu = 0.77 mg dm^{-3}; Fe = 11.6 mg dm^{-3}; Zn = 4.1 mg dm^{-3}; and Mn = 16.0 mg dm^{-3}.

Experimental design and treatments

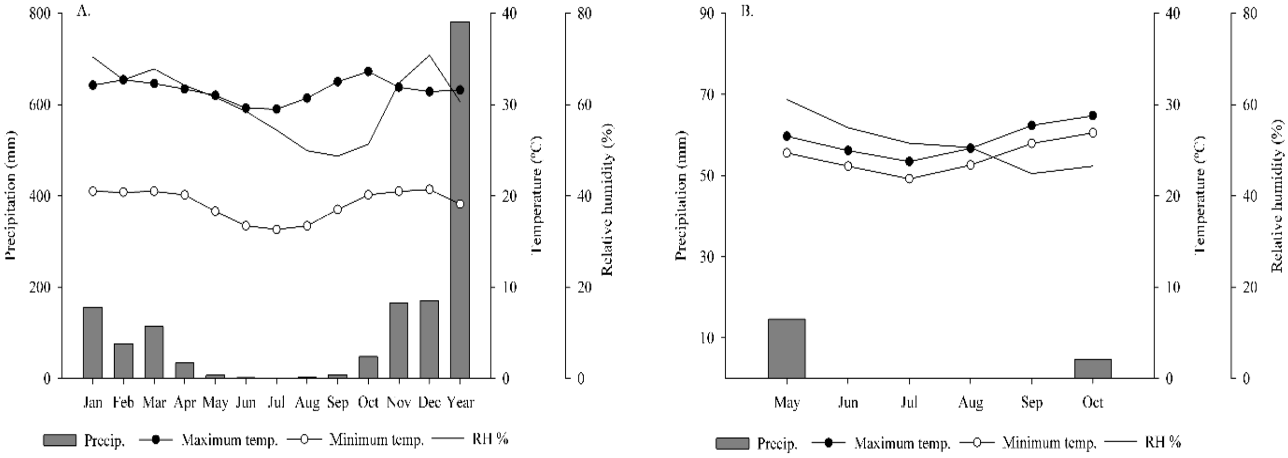
Two field experiments were conducted simultaneously, one under well-watered and the other under water deficit conditions following the pre-flowering growth stage. Four grain sorghum hybrids were evaluated (DKB 540, BRS 310, BRS 332, and 50A10) across both experiments.

A randomized block experimental design with four replications was used. The experimental plots consisted of four sorghum planting rows, each 4 m in length, spaced 0.5 m apart, covering a total area of 8 m^2. The evaluation area consisted of the two central rows of each plot, resulting in a total area of 4 m^2.

Implementation and conduction of experiments

The experiments were conducted from May 14 to October 08. The soil was prepared using a conventional system, with one plowing and two harrowing before planting, followed by furrowing and fertilizer application. Fertilizers were applied to the planting furrow based on soil analysis results and the crop demand, using 20 kg ha^{-1} of N,

Figure 1 - A. Climatological normal for Janaúba, MG, Brazil (1981-2010) with mean monthly and annual accumulated rainfall depths (mm), maximum and minimum temperatures (°C), and relative air humidity (%); B. observed climatological characteristics during the experimental period. (Source: Brazilian National Institute of Meteorology - INMET, 2024)



70 kg ha^{-1} of P_2O_5, and 40 kg ha^{-1} of K_2O. Topdressing was applied at the sorghum development stage 2 (visible fifth leaf ligule), applying 72 kg ha^{-1} of N, with urea as the source.

Sorghum seeds were manually planted in the furrow, with two seeds placed every 0.1 m. Thinning was carried out to avoid an excess of plants when they had three to four fully expanded leaves, leaving 10 plants m^{-1}, for a population of 200,000 plants ha^{-1}.

Cultural practices included manual weeding and control of pests and diseases through applications of chemical products approved for the crop, when necessary. Panicles of plants in the evaluation area were covered with polyethylene nets soon after flowering to prevent birds from feed on the grains.

The plants were irrigated using a conventional fixed sprinkler system, with sprinklers spaced 12 × 12 m apart, service pressure of 250 kPa, opening diameter of 4.0 × 2.6 mm, and flow of 1.6 m^3 h^{-1}. The experimental plots were irrigated to maintain soil moisture near field capacity until the water deficit treatment was applied.

Irrigation was suppressed in the experiment with water deficit starting from the beginning of the pre-flowering period (July 2), defined as phenological stage 5 or boot stage, according to Vanderlip and Reeves (1972), until the ending of crop cycle. A total water depth of 210.62 mm was used in this experiment.

In the experiment under well-watered conditions, plots were irrigated normally until the grain physiological maturation (R6), on August 27. A total water depth of 495.12 mm was used in this experiment.

Evaluated characteristics

The evaluated characteristics included relative chlorophyll content (RCC), chlorophyll fluorescence (FV/FM), number of days to flowering (NDF), leaf area (LA), plant height (PH), number of lodged plants (NLP), panicle length (PL), panicle diameter (PD), panicle mass (PM), grain mass per panicle (GMP), panicle index (PI), threshing index (TI), 100-grain mass (100G), grain yield (GY), and water use efficiency (WUE).

RCC was measured using a portable chlorophyll meter (SPAD 502, Minolta, Japan). Measurements were carried out on the middle third of the leaves, based on the mean of five readings per plant, in three plants per plot. FV/FM was measured using a fluorimeter (Plant Efficiency Analyser; Hansatech Instruments, King's Lynn, UK), with readings carried out between 9:00 and 10:00 on the middle third of previously dark-adapted leaves, in three plants per plot.

NDF was estimated by counting the number of days from sowing to the time when more than 50% of the plants in the plot reached anthesis. LA was estimated based on measurements of length (C) and width (L) of all leaves with photosynthetically active leaf area, in three randomly chosen plants per plot, according to the formula: LA (cm^2) = 0.75 × C × L (Francis; Rutger; Palmer, 1969).

PH (m) was determined by measuring the plants from the ground level to the panicle apex. Five plants in the evaluation area of each plot were evaluated during the grain physiological maturation stage. NLP was determined by counting the number of lodged plants with angles equal to or higher than 45° in each experimental plot.

PL (cm) was determined after harvest, by collecting ten randomly chosen panicles from each plot, and measuring them from the base to the apex using a ruler. PD (cm) was measured in the central part of panicles, using a caliper.

PM (g) was measured in ten randomly chosen panicles from each plot and separately weighed. GMP (g) was measured by weighing the grains of each panicle.

PI was determined by the ratio between the total number of panicles present in the useful area and the plant stand. TI was determined by the ratio between grain and panicle mass.

100 G (g) was determined by the mean mass of three subsamples of 100 grains randomly chosen from each plot, and converting the results to 13% moisture.

GY was determined by weighing the grain harvested in the evaluation area of each plot, correcting the values to 13% moisture, and converting them into kg ha^{-1}.

WUE (kg ha^{-1} mm^{-1}) was calculated by the ratio between the grain yield of each sorghum hybrid (kg ha^{-1}) and the irrigation water depth applied (mm).

Statistical analyses

The assumptions of normality and homoscedasticity of the data were confirmed. The means were subjected to individual analysis of variance and the homogeneity of residual variance was assessed. Thus, the data were subjected to joint analysis of variance, involving the two water availability conditions (water deficit and well-watered). When the effects of the water availability conditions were significant, the means were compared using the F-test at a 5% significance level ($p < 0.05$) and the means related to effects of the hybrids were compared using the Tukey's test at a 5% significance level ($p \leq 0.05$). NLP data were transformed using the ArcSen $\sqrt{x+1}$ method (arcsine square root transformation).

Additionally, multivariate analysis of variance (MANOVA) was applied to assess the grouping of the different morphophysiological responses of the plants. In this sense, the stats package of the MANOVA function was employed, using the Pillai test at a 5% significance level. After confirming the absence of multicollinearity, the data were subjected to canonical variable (CV) analysis, using the candisc package (Friendly; Fox, 2017). Pearson's correlation (r) was also calculated to evaluate the associations between the variables and grain yield. All statistical analyses were carried out using the software R (R Core Team, 2018).

RESULTS AND DISCUSSION

The joint analysis of variance of data from sorghum hybrids in environments under water deficit and well-watered conditions showed a significant effect of the hybrid-environment (HxE) interaction on leaf area (LA), number of lodged plants (NLP), and 100-grain mass (100 G). For the other variables, no significant interaction was observed (Table 1).

Isolated effects of the environment (E) and the hybrid (H) were observed simultaneously for panicle length (PL), panicle diameter (PD), panicle mass (PM), grain mass per panicle (GMP), threshing index (TI), and grain yield (GY). The environment factor affected relative chlorophyll content (RCC), chlorophyll fluorescence (FV/FM) and water use efficiency (WUE), while the hybrid factor affected NDF (Table 1).

The factors had no significant effect on plant height (PH) and panicle index (PI), which had mean values of 1.32 m and 0.98 panicles per plant, respectively (Table 1). Thus, the water deficit did not affect PH and PI, resulting in plants with adequate size and panicle index.

The results could illustrate the impacts of droughts at the pre-flowering stage on sorghum morphophysiological characteristics due to the absence of rainfall during the period in which water deficit was imposed (Figure 1B).

The effect of the environment within the hybrids and the effect of the hybrid within the environments indicated that the water deficit decreased the photosynthetically active leaf area (LA) of the hybrids 50A10, BRS 332, and DKB 540 by 39.8%, 39.7%, and 17.8%, respectively. However, the LA of the hybrid BRS 310 was similar in both environments. In the environment in which water restriction was administered, the hybrids DKB 540 and BRS 310 showed greater LA than the others. In the well-irrigated environment, no significant difference was found between the hybrids (Table 2).

The higher photosynthetically active leaf area observed in some hybrids in the water deficit environment can be attributed to drought-tolerance adaptations, a phenomenon termed stay-green. The persistence of plants with high LA in water deficit environments has advantages, including an extended duration of carbohydrate translocation, due to their ability to sustain photosynthesis (Thomas; Ougham, 2014). This leads to a grater production stability under conditions of low water availability and increase stem resistance to lodging.

According to Magalhães *et al*. (2014), photosynthetically active leaf area is strongly correlated to the final plant dry matter production, which is dependent on the ability of leaves to perform photosynthesis and their photosynthesis rate per unit of area. Thus, genotypes that maintain a higher LA present higher potential to tolerate drought periods.

The reduced photosynthetically active leaf area of

Table 1 - Analysis of variance of indicators of tolerance to drought in sorghum hybrids grown under water deficit and well-watered conditions

EC	SV	Block	Hybrid (H)	Environment (E)	H × E	Residue	Mean	CV (%)
	DF	6	3	1	3	18		
	Mean square							
RCC		9.2693^{ns}	12.6187^{ns}	1150.4022**	28.5325^{ns}	14.2834	47.89	7.89
FV/FM		0.0036^{ns}	0.0050^{ns}	0.0310**	0.0041^{ns}	0.0020	0.69	6.37
NDF		1.0625^{ns}	52.2083**	0.5000^{ns}	0.2500^{ns}	0.4792	60.81	1.14
LA		2269.882^{ns}	11268.268**	36634.569**	3960.944*	1214.230	235.60	14.79
PH		0.0084^{ns}	0.0030^{ns}	0.0014^{ns}	0.0085^{ns}	0.0036	1.32	4.54
NLP[1]		137.7544**	131.0557*	3316.5883**	128.8462*	30.8153	(10.28)	34.55
PL		2.3381*	12.4586**	17.1113**	0.6746^{ns}	0.8693	24.04	3.88
PD		0.7058**	0.4401*	5.4576**	0.0420^{ns}	0.1058	4.10	7.93
PM		20.5871^{ns}	367.9409**	408.8942**	33.0445^{ns}	25.3185	43.61	11.54
GMP		12.0738^{ns}	246.7179**	342.8271**	26.4608^{ns}	14.4421	31.12	12.21
TI		0.0008^{ns}	0.0037*	0.0076**	0.0003^{ns}	0.0009	0.71	4.14
PI		0.0005^{ns}	0.0013^{ns}	0.0001^{ns}	0.0001^{ns}	0.0007	0.98	2.67
100G		0.3886**	3.1373**	10.1813**	0.3665**	0.0325	2.59	6.97
GY		1541291.5^{ns}	3509709.1*	51174561.9**	1585663.5^{ns}	961147.3	9327.70	10.51
WUE		16.6163^{ns}	34.1081^{ns}	2281.9893**	15.6577^{ns}	11.3971	29.84	11.31

^{ns} = not significant ($p > 0.05$), ** = significant at 1% ($p < 0.01$), and * = significant at 5% ($p < 0.05$) by the F-test. [1] = observed mean value. EC = evaluated characteristic; SV = source of variation; DF = degrees of freedom; CV = coefficient of variation. RCC = relative chlorophyll content; FV/FM = chlorophyll fluorescence; NDF = number of days to flowering; LA = leaf area; PH = plant height; NLP = number of lodged plants; PL = panicle length; PD = panicle diameter; PM = panicle mass; GMP = grain mass per panicle; TI = threshing index; PI = panicle index; 100 G = 100-grain mass; GY = grain yield; WUE = water use efficiency

Table 2 - Leaf area (LA), 100-grain mass (100 G), and number of lodged plants (NLP) of sorghum hybrids grown under water deficit (WD) and well-watered (WW) environments

Hybrids	LA (cm^2)		100 G (g)		NLP[1] -	
	WD	WW	WD	WW	WD	WW
DKB 540	256.68 aB	312.33 aA	2.54 aB	3.87 aA	35.97(34.75) aA	5.74 (0.00) aB
BRS 310	239.47 aA	249.38 aA	1.59 bB	2.47 bA	16.42(7.25) bA	5.74 (0.00) aB
BRS 332	150.52 bB	249.56 aA	1.73 bB	2.41 bA	27.64(22.00) aA	6.34(0.25) aB
50A10	160.38 bB	266.48 aA	2.24 aB	3.87 aA	24.97(18.00) abA	5.74(0.00) aB
CV(%)	14.79		6.97		34.55	

Means followed by same lowercase letter in the columns (comparing hybrids), or uppercase letter in the rows (comparing environments), are not significantly different from each other by the Tukey's test ($p \leq 0.05$) and F-test ($p < 0.05$), respectively. [1]Means in parentheses are the original values without transformation

sorghum hybrids in the water deficit environment can be attributed to inhibition of cell division, elongation, and differentiation. This reduction inhibits leaf expansion and represents one of the initial responses to water deficiency in plants (Fahad *et al*., 2017; Taiz *et al*., 2017).

It was observed that, as a result of water restriction, the hybrids under study had their 100G reduced. Additionally, it was found that the hybrids BRS 310 and BRS 332 exhibit lower 100G than the others in both environments (Table 2). The reduction in grain mass under water deficit conditions can be attributed to the decrease in photosynthetic efficiency of plants.

According to Batista *et al*. (2019a), a reduced photosynthetic rate in sorghum plants result in lower translocation of carbohydrates to leaves and production of photoassimilates to other plant organs, mainly grains. This reduction explains the significant decreases in 100 G.

Barnabás *et al*. (2008) observed a reduction in grain mass in response to water or thermal stress. They attributed this reduction to compromised starch synthesis and a decrease in number of endosperm cells caused by the limited availability of assimilates to grains and the direct effects on grain development, affecting the crop yield. Therefore, genotypes that can sustain higher 100 G under water deficit conditions tend to show greater stability in grain yield under drought.

No lodging was found for the hybrids in the well-watered environment. The plants had a higher tendency for lodging in the water deficit environment, with the hybrid BRS 310 exhibiting a lower NLP compared to the hybrids DKB 540 and BRS 332. This is explained by the reduced photosynthetic rates and insufficient production of photoassimilates needed to maintain tissues (Silva *et al*., 2012). This weakens plant support structures, accelerates senescence and, consequently, increases susceptibility to lodging. Therefore, the asymmetric partition of photoassimilates from the stalk to the grains may have contributed to an increase in NLP values under water deficit conditions.

The sorghum plants produced longer panicles in the water deficit environment, with the hybrid BRS 332 exhibiting a lower mean value compared to the others. In contrast, pre-flowering water stress decreased PD (-18.2%), PM (-15.2%), GMP (-19%), and TI (-5.5%) of the hybrids, resulting in a GY loss of 23.9% (Table 3).

A decrease in grain productivity was expected under water deficit conditions. Since, in self-fertilizing species such as sorghum, the occurrence of water deficit prior to anthesis potentially reduces the number of grains, which is determined at the beginning of panicle differentiation, a period that is extremely sensitive to abiotic stresses (Dolferus; Ji; Richards, 2011).

Furthermore, water stress reduces the photosynthetic rate, accelerates plant senescence, and shortens the grain filling period. Thess effects are associated with the depletion of stem and sheath reserves and remobilization of assimilates from vegetative tissues to grains (Hussain *et al*., 2019). This leads to yield losses that are also evidenced by a decrease in starch accumulation, which generally composes a large part of the cereal dry weight (Barnabás *et al*., 2008).

These results indicate that, although sorghum is relatively adapted to drought-prone environments, it still requires a certain soil moisture to ensure the maintenance of grain yield under water deficit conditions. Moreover, it was observed that the losses and/or maintenance of productivity depend on the association of morphophysiological indicators and the extent to which these were influenced or not by water restriction. Therefore, the success of the crop's drought tolerance may be conditioned by the sorghum cultivars and their characteristics associated with maintaining productivity in water-restricted environments (Magalhães *et al*., 2016). This highlights the relevance of the information generated for establishing scientific foundations.

The hybrid BRS 310 exhibited a lower PD than BRS 332, and a lower PM and GMP than the others.

Table 3 - Panicle length (PL), diameter (PD), and mass (PM), grain mass per panicle (GMP), threshing index (TI), grain yield (GY), and number of days to flowering (NDF) in sorghum hybrids grown under water deficit (WD) and well-watered (WW) environments

Hybrids	PL	PD	PM	GMP	TI	GY	NDF
	(cm)		(g)		-	(kg ha^{-1})	(days)
DKB 540	24.39 a	4.02 ab	45.57 a	33.43 a	0.73 a	9533.40 ab	64.50 a
BRS 310	25.21 a	3.83 b	34.10 b	23.26 b	0.68 b	9142.98 ab	60.25 b
BRS 332	22.28 b	4.38 a	44.62 a	31.67 a	0.71 ab	8529.96 b	59.88 b
50A10	24.29 a	4.17 ab	50.15 a	36.14 a	0.72 ab	10104.44 a	58.63 c
WD	24.77 A	3.69 B	40.04 B	27.85 B	0.69 B	8063.10 B	-
WW	23.31 B	4.51 A	47.19 A	34.40 A	0.73 A	10592.29 A	-
CV(%)	3.88	7.93	11.54	12.21	4.14	10.51	1.14

Means followed by same lowercase letter in the columns (comparing hybrids), or uppercase letter in the rows (comparing environments), are not significantly different from each other by the Tukey's test ($p \leq 0.05$) and F-test ($p < 0.05$), respectively

This lower PM and GMP resulted in a TI of 68%, a value lower than that of the DKB 540 hybrid, whose TI indicated that 73% of the PM was allocated to grains (Table 3). It is understood that genotypes that allocate a higher proportion of mass to grains relative to panicle composition may contribute to higher yields under drought conditions.

Menezes *et al*. (2015) and Tardin *et al*. (2013) report that TI is a key trait linked to sorghum grain yield, as plants with high TI generally achieve higher yields. Thus, this characteristic can be used to assist in the selection of high-performance genotypes in both water deficit and well-watered environments.

The hybrid 50A10 was more productive than BRS 322, exhibiting higher grain yield combined with a shorter time for 50% of plants in the plot to reach anthesis (NDF) (Table 3). These different responses highlight the importance of adjusting flowering time as a crucial mechanism for adapting to environmental conditions, as it can help prevent abiotic stresses such as drought (Dolferus; Ji; Richards, 2011).

Sorghum crops are usually sown between summer crop seasons, at the end of the rainfall period. Thus, they are more prone to low water availability. In this context, early maturation in genotypes can be a favorable mechanism to escape drought effects, although late maturation hybrids usually result in higher yields (Menezes *et al*., 2015; Tardin *et al*., 2013). A strategy to minimize the risk of water stress in late maturation hybrids is to plant them at the beginning of the rainy season, while early maturation hybrids can be planted later in the season.

The effect of the environment factor (water deficit and well-watered) showed that plants subjected to water deficit conditions exhibited reduced RCC and FV/FM (Table 4). The observed reduction in photochemical efficiency may indicate decreased electron transport, increased heat dissipation, and consequently, greater photoinhibition.

According to Taiz *et al*. (2017), water deficiency causes loss of cell turgidity, pigment photooxidation, and chlorophyll degradation. This culminates in stomatal closure, reduced CO_2 assimilation and electron transport, inhibition of photosynthetic processes, and affects biochemical processes and enzymatic activity. Furthermore, lower photosynthetic efficiency can also reduce the transport of photoassimilates to the grains, since water, essential for this process, is limited, directly affecting production under water stress (Batista *et al*., 2017).

Plant mechanisms that include changes in chlorophyll synthesis, osmotic regulation, chloroplast functional and structural alterations, and photosynthetic rate directly or indirectly determine plant tolerance to water deficit (Anjum *et al*., 2011). Thus, a continuous study is important for improving this information and the progress of sorghum breeding programs focused on tolerance to drought.

Higher WUE was found in the water deficit environment (Table 4). This suggests that plants enhanced the conversion of water units into production units when subjected to water stress. This higher WUE can be attributed to the ability of plants to use the available water, regulating transpiration and asymmetrically allocating photoassimilates, which assist in ensuring grain production under unfavorable conditions (Lopes *et al*., 2011).

Barnabás *et al*. (2008) reported that increases in WUE under stress conditions can assist in the development of reproductive organs and, consequently, in the maintenance cereal crop production under drought conditions. This denotes the importance of scientifically exploring WUE to enhance understanding of crop responses to adverse conditions, benefiting agriculture.

A canonical variables (CV) analysis was performed to investigate influential traits and select indicators of drought tolerance. The vector matrix indicates the proportion of the total variance explained by each canonical variable and their respective correlations with the analyzed characteristics. The first two canonical variables (CV1, CV2) explained 80.58% of the total variation observed in the different environments (Figure 2).

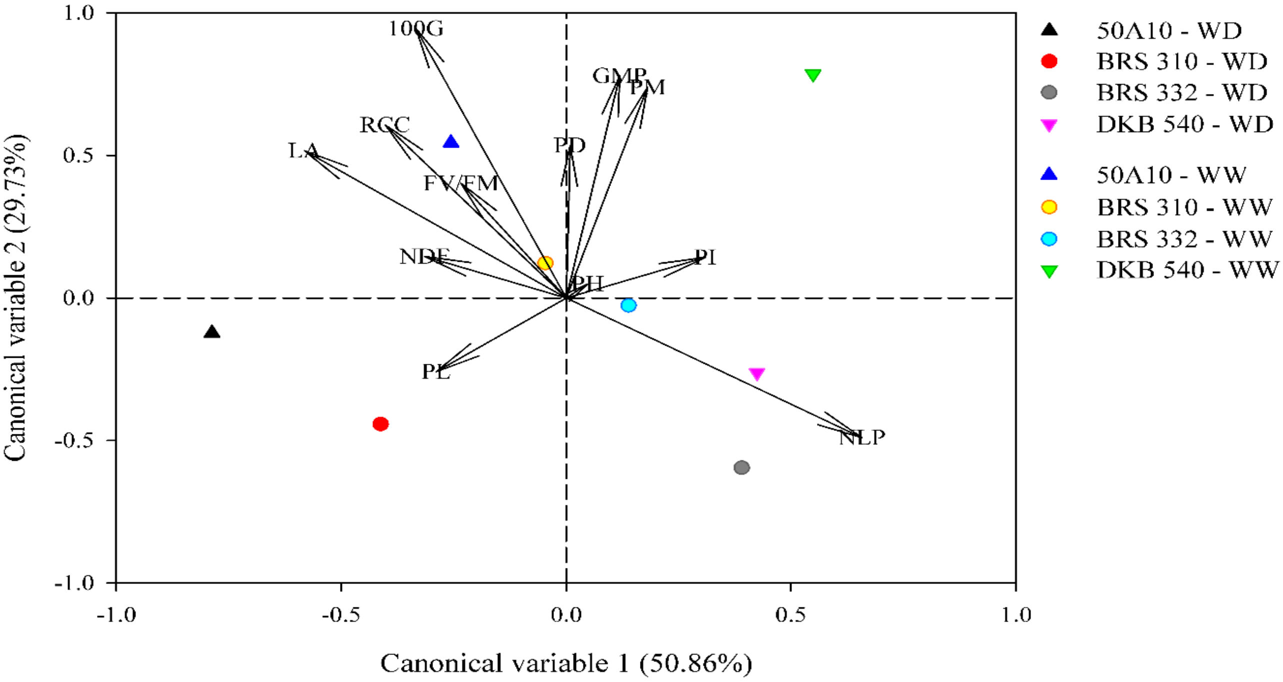
These results highlight the potential of canonical variables in identifying key traits associated with drought tolerance. The integration of this analysis into breeding programs can contribute to the development and selection of more resilient sorghum hybrids.

Table 4. Relative chlorophyll content (RCC), chlorophyll fluorescence (FV/FM), and water use efficiency (WUE) in sorghum hybrids grown under water deficit (WD) and well-watered (WW) environments

Environment	RCC	FV/FM	WUE (kg ha^{-1} mm^{-1})
WD	41.90 B	0.66 B	38.28 A
WW	53.89 A	0.72 A	21.39 B
CV(%)	7.89	6.37	11.31

Means followed by same letter in the columns are not significantly different from each other by the F-test ($p < 0.05$), respectively

Figure 2 - Graphical dispersion of canonical variables (CV1 and CV2) in sorghum hybrids grown under water deficit (WD) and well-watered (WW) environments



Canonical variable 1 (CV1) explained 50.86% of the total variation, with NLP and LA as the characteristics with the highest relative contribution. Canonical variable 2 (CV2) explained 29.73% of the total variation, with the greatest contribution from 100 G, followed by GMP, PM, and RCC (Figure 2).

Positive correlations are responsible for discriminating the treatments located to the right of VC1 and in the upper part of VC2. On the other hand, negative correlations are responsible for discriminating the treatments located to the left of VC1 and in the lower part of VC2 (Figure 2). The proximity of the hybrids to the respective correlation vectors of the variables suggests the characteristics in which they exhibit the greatest affinity, according to the environment (water deficit or well-watered) in which they are placed.

Characteristics with eigenvectors with the same sign have a direct effect, meaning that when the value of one increases, the value of the other also increases, and vice versa. Those with opposite signs exhibit an inverse relationship, meaning that when the value of one increases, the value of the other decreases. For example, as the RCC decreased, FV/FM, NDF, LA, 100 G decreased, while NLP increased (Table 5).

According to Sousa *et al*. (2018), the use of multivariate analyses is essential for the investigation and selection of indicators of tolerance to drought in hybrids. It allows researchers to determine characteristics responsible for tolerance to drought and exclude those

Table 5 - Canonical correlations (CV1 and CV2) referring to indicators of tolerance to drought in sorghum hybrids grown under water deficit and well-watered conditions

EC	CV1	CV2
RCC	-0.40	0.61
FV/FM	-0.23	0.40
NDF	-0.31	0.15
LA	-0.58	0.52
PH	0.05	0.05
NLP	0.65	-0.49
PL	-0.28	-0.26
PD	0.01	0.53
PM	0.18	0.74
GMP	0.12	0.78
PI	0.31	0.14
100 G	-0.33	0.95

EC = evaluated characteristic; RCC = relative chlorophyll content; FV/FM = chlorophyll fluorescence; NDF = number of days to flowering; LA = leaf area; PH = plant height; NLP = number of lodged plants; PL = panicle length; PD = panicle diameter; PM = panicle mass; GMP = grain mass per panicle; PI = panicle index; 100 G = 100-grain mass

Table 6 - Pearson's correlation coefficients (r) for morphophysiological characteristics of sorghum hybrids grown under water deficit and well-watered conditions

EC	GY	RCC	FV/FM	NDF	LA	PH	NLP	PL	PD	PM	GMP	TI	PI	100 G	WUE
GY	1	-	-	-	-	-	-	-	-	-	-	-	-	-	-
RCC	0.63**	1	-	-	-	-	-	-	-	-	-	-	-	-	-
FV/FM	0.33	0.64*	1	-	-	-	-	-	-	-	-	-	-	-	-
NDF	-0.02	0.09	0.36*	1	-	-	-	-	-	-	-	-	-	-	-
LA	0.57**	0.51**	0.58**	0.51**	1	-	-	-	-	-	-	-	-	-	-
PH	0.28	0.11	0.24	0.19	0.44*	1	-	-	-	-	-	-	-	-	-
NLP	-0.44*	-0.48**	-0.16	0.23	-0.18	0.09	1	-	-	-	-	-	-	-	-
PL	-0.27	-0.38*	-0.32	0.07	0.11	-0.14	0.22	1	-	-	-	-	-	-	-
PD	0.51**	0.57**	0.36*	-0.03	0.22	0.11	-0.42*	-0.69**	1	-	-	-	-	-	-
PM	0.51**	0.28	0.19	-0.01	0.21	0.2	-0.27	-0.38*	0.59**	1	-	-	-	-	-
GMP	0.55**	0.33	0.26	0.05	0.24	0.2	-0.29	-0.42*	0.60**	0.98**	1	-	-	-	-
TI	0.46**	0.37*	0.43*	0.28	0.2	0.1	-0.2	-0.45*	0.38*	0.54**	0.68**	1	-	-	-
PI	-0.02	-0.06	-0.08	0.02	-0.18	-0.06	0.17	-0.2	0.09	0.22	0.27	0.39*	1	-	-
100 G	0.72**	0.51*	0.42*	0.31	0.62**	0.21	-0.21	-0.11	0.32	0.64**	0.68**	0.55**	0.07	1	-
WUE	-0.43*	-0.80**	-0.54**	-0.04	-0.38*	0.09	0.63**	0.47**	-0.61**	-0.32	-0.36*	-0.34	0.06	-0.46**	1

** = significant at 1% ($p < 0.01$), and * = significant at 5% ($p < 0.05$) by the F-test. EC = evaluated characteristic; GY = grain yield; RCC = relative chlorophyll content; FV/FM = chlorophyll fluorescence; NDF = number of days to flowering; LA = leaf area; PH = plant height; NLP = number of lodged plants; PL = panicle length; PD = panicle diameter; PM = panicle mass; GMP = grain mass per panicle; TI = threshing index; PI = panicle index; 100 G = 100-grain mass; WUE = water use efficiency

of little contribution to distinguishing hybrids under water deficit (Wattoo *et al*., 2018). Therefore, optimizing resources and the efficiency of breeding programs with an emphasis on selection for drought tolerance.

In order to determine the associations between morphophysiological traits of drought tolerance and grain yield, a Pearson's correlation analysis (r) was conducted. GY showed significant positive correlations with RCC, LA, PD, PM, GMP, TI, and 100G. However, significant negative correlations were observed with NLP and WUE (Table 6). Characteristics with significant positive correlations with grain yield contributed directly to the performance of the hybrid under stress conditions and should be considered as indicators for selection (Sousa *et al*., 2018).

These findings complement the information generated by the canonical variable analysis, further highlighting the significant correlation of RCC with FV/FM, LA, and NLP, and other characteristics related to grain yield (Table 6).

According to Batista *et al*. (2019b), the coefficient of correlation determines the linear association level between two characteristics and, combined with other multivariate analyses, assists in the understand of plants dynamics and their different mechanisms of tolerance under water deficit condition. Thus, it enables the determination of useful indicators for distinguishing and selecting genotypes tolerant to drought.

Therefore, the results of the present work demonstrated that maintaining high RCC, LA, PD, PM, GMP, TI, and 100 G and a low NLP under water deficit conditions is essential for sustaining grain yield and tolerance to drought. It is worth noting that these indicators are easy to measure and can significantly assist sorghum breeding programs in creating and manipulating scientific bases that enable the development of technological solutions for coping with drought. As a result, ensuring the sustainability of productive activities and providing food security for society.

CONCLUSIONS

1. The occurrence of water deficit from the pre-flowering stage onwards reduces the grain yield of sorghum by 23.9%;
2. Higher grain yield of the sorghum hybrids is associated with higher relative chlorophyll content (RCC), leaf area (LA), panicle diameter, panicle mass (PM), grain mass per panicle (GMP), threshing index, 100-grain mass (100 G), as well as a lower number of lodged plants (NLP);
3. GMP, 100 G, PM, NLP, RCC, and LA are the most important indicators for explaining drought tolerance variations in grain sorghum hybrids grown under water deficit and well-watered conditions.

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REFERENCES

ALVARES, C. A. *et al*. Köppen's climate classification map for Brazil. **Meteorologische zeitschrift**, v. 22, n. 6, p. 711-728, 2013.

ANJUM, S. A. *et al*. Morphological, physiological and biochemical responses of plants to drought stress. **African Journal of Agricultural Research**, v. 6, n. 9, p. 2026-2032, 2011.

BARNABÁS, B.; JÄGER, K.; FEHÉR, A. The effect of drought and heat stress on reproductive processes in cereals. **Plant, Cell and Environment**, v. 31, n. 1, p. 11-38, 2008.

BATISTA, P. S. C. *et al*. Índices de seleção para identificar genótipos de sorgo granífero tolerantes ao estresse hídrico. **Revista Brasileira de Milho e Sorgo**, v. 18, n. 3, p. 379-395, 2019b.

BATISTA, P. S. C. *et al*. Performance of grain sorghum hybrids under drought stress using GGE biplot analyses. **Genetics and Molecular Research**, v. 16, n. 3, p. 1-12, 2017.

BATISTA, P. S. C. *et al*. Selection of sorghum for drought tolerance in a semiarid environment. **Genetics and Molecular Research**, v. 18, n. 1, p. 1-11, 2019a.

DOLFERUS, R.; JI, X.; RICHARDS, R. A. Abiotic stress and control of grain number in cereals. **Plant Science**, v. 181, n. 4, p. 331-341, 2011.

FAHAD, S. *et al*. Crop production under drought and heat stress: plant responses and management options. **Frontiers in Plant Science**, v. 8, p. 1-16, 2017.

FRANCIS, C. A.; RUTGER, J. N.; PALMER, A. F. E. A rapid method for plant leaf area estimation in maize (*Zea mays* L.). **Crop Science**, v. 9, n. 5, p. 537-539, 1969.

FRIENDLY, M.; FOX, J. **Candisc**: visualizing generalized canonical discriminant and canonical correlation analysis. Version R package version 0.6-5. 2017.

HUSSAIN, S. *et al*. Drought stress in plants: an overview on implications, tolerance mechanisms and agronomic mitigation strategies. **Plant Science Today**, v. 6, n. 4, p. 389-402, 2019.

INMET - Instituto Nacional de Meteorologia. **Normais Climatológicas do Brasil** – (1981- 2010). Disponível em:< https://portal.inmet.gov.br/>. Acesso em: 01 out. 2024.

LOPES, M. S. *et al*. Enhancing drought tolerance in C4 crops. **Journal of Experimental Botany**, v. 62, n. 9, p. 3135-3153, 2011.

MAGALHÃES, P. C. *et al*. Exigências edafoclimáticas e fisiologia da produção. *In*: BORÉM, A.; PIMENTEL, L. D.; PARRELLA, R. A. C. (ed.). **Sorgo**: do plantio à colheita. 1. ed. Viçosa, MG: UFV, 2014. cap. 3, p. 58-88.

MAGALHÃES, P. C. *et al*. Phenotypic plasticity of root system and shoots of *Sorghum bicolor* under different soil water levels during pre-flowering stage. **Australian Journal of Crop Science**, v. 10, n. 1, p. 81-87, 2016.

MENEZES, C. B. *et al*. Evaluation of grain yield in sorghum hybrids under water stress. **Genetics and Molecular Research**, v. 14, n. 4, p. 12675-12683, 2015.

MOLOTOKS, A.; SMITH, P.; DAWSON, T. P. Impacts of land use, population, and climate change on global food security. **Food and Energy Security**, v. 10, n. 1, p. 1-20, 2021.

MOREIRA, S. A. F. *et al*. Maize hybrids contrasting for drought tolerance differ during the vegetative stage. **Semina: Ciências Agrárias**, v. 41, n. 4, p. 1093-1106, 2020.

PARDO, J.; VANBUREN, R. Evolutionary innovations driving abiotic stress tolerance in C4 grasses and cereals. **The Plant Cell**, v. 33, n. 11, p. 3391-3401, 2021.

PIRES, M. V. *et al*. Yield-related phenotypic traits of drought resistant maize genotypes. **Environmental and Experimental Botany**, v. 171, p. 1-10, 2020.

R CORE TEAM. **R**: a language and environment for statistical computing. Vienna, Austria: R Foundation for Statistical Computing, 2018.

SANTOS, H. G. *et al*. **Brazilian Soil Classification System**. 5 th ed. Brasília, DF: Embrapa, 2018. 356 p.

SILVA, M. R. *et al*. Desempenho agronômico de genótipos de milho sob condições de restrição hídrica. **Revista de Ciências Agrárias**, v. 35, n. 1, p. 202-212, 2012.

SOUSA, R. S. *et al*. Identification of drought-tolerant corn genotypes by multivariate analysis. **Pesquisa Agropecuária Tropical**, v. 48, n. 3, p. 204-211, 2018.

TAIZ, L. *et al*. **Fisiologia vegetal**. 6. ed. Porto Alegre: Artmed, 2017. 888 p.

TARDIN, F. D. *et al*. Avaliação agronômica de híbridos de sorgo granífero cultivados sob irrigação e estresse hídrico. **Revista Brasileira de Milho e Sorgo**, v. 12, n. 2, p. 102-117, 2013.

THOMAS, H.; OUGHAM, H. The stay-green trait. **Journal of Experimental Botany**, v. 65, n. 14, p. 3889-3900, 2014.

VANDERLIP, R. L.; REEVES, H. E. Growth stages of sorghum [*Sorghum bicolor*, (L.) Moench.]. **Agronomy Journal**, v. 64, n. 1, p. 13-16, 1972.

WATTOO, F. M. *et al*. Identification of drought tolerant maize genotypes and seedling based morpho-physiological selection indices for crop improvement. **Sains Malaysiana**, v. 47, n. 2, p. 295-302, 2018.

WOLDESEMAYAT, A. A. *et al*. Cross-species multiple environmental stress responses: an integrated approach to identify candidate genes for multiple stress tolerance in sorghum (*Sorghum bicolor* (L.) Moench) and related model species. **PloS One**, v. 13, n. 5, p. 1-30, 2018.