

Growth and morphology of grain sorghum hybrids cultivated in semi-arid region under pre- and post-flowering drought stress¹

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ABSTRACT

Drought stress is a major factor limiting sorghum yield, especially in regions with irregular rainfall. This study investigated the morphophysiological and growth responses of grain sorghum hybrids to drought stress, comparing the effects at pre- and post-flowering stages. Twenty hybrids were evaluated under well-watered conditions and under drought imposed at the pre- and post-flowering stages, using a randomized complete block design, with three replications. The evaluated traits were plant height, leaf area index, shoot dry weight, and chlorophyll index. Growth parameters, including absolute growth rate, relative growth rate, leaf area ratio, leaf weight ratio, specific leaf area, and net assimilation rate, were also determined. The pre-flowering drought caused the greatest reductions in plant height, leaf area index, and dry weight accumulation, whereas the post-flowering drought had the greatest effect on leaf area ratio, chlorophyll index, and net assimilation rate. The grain yield under pre-flowering water stress was approximately 4,000 kg ha⁻¹, significantly higher than the 2,500 kg ha⁻¹ observed under post-flowering stress, suggesting a greater capacity for recovery and grain production when water availability is restored before the reproductive stage.

KEYWORDS: *Sorghum bicolor* L., abiotic stress, sorghum breeding.

RESUMO

Crescimento e morfologia de híbridos de sorgo granífero cultivado no semiárido sob estresse hídrico em pré- e pós-floração

O estresse hídrico é um dos principais fatores limitantes da produtividade do sorgo, especialmente em regiões com chuvas irregulares. Este estudo investigou as respostas morfofisiológicas e de crescimento de híbridos de sorgo granífero ao estresse hídrico, comparando os efeitos nos estádios de pré- e pós-florescimento. Vinte híbridos foram avaliados sob condições bem irrigadas e sob déficit hídrico imposto nos estádios de pré- e pós-florescimento, utilizando-se delineamento em blocos ao acaso, com três repetições. As características avaliadas foram altura de plantas, índice de área foliar, massa seca da parte aérea e índice de clorofila. Parâmetros de crescimento, incluindo taxa de crescimento absoluto e relativo, razão de área foliar, razão de massa foliar, área foliar específica e taxa de assimilação líquida, também foram determinados. O déficit hídrico no pré-florescimento causou as maiores reduções na altura de plantas, índice de área foliar e acúmulo de massa seca, enquanto o estresse no pós-florescimento teve maior efeito sobre a razão de área foliar, índice de clorofila e taxa de assimilação líquida. A produtividade de grãos sob estresse hídrico no pré-florescimento foi de aproximadamente 4.000 kg ha⁻¹, significativamente superior aos 2.500 kg ha⁻¹ observados sob estresse no pós-florescimento, sugerindo maior capacidade de recuperação e produção de grãos quando a disponibilidade hídrica é restabelecida antes da fase reprodutiva.

PALAVRAS-CHAVE: *Sorghum bicolor* L., estresse abiótico, melhoramento de sorgo.

INTRODUCTION

Drought stress is a major constraint to agriculture in many regions worldwide, affecting plant growth in almost all developmental stages

and significantly reducing crop yield (Virnodkar et al. 2020). In the context of ongoing climate change, identifying high-yielding and drought-adapted crops, such as sorghum, has become increasingly important.

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Sorghum [*Sorghum bicolor* (L.) Moench] is a key food and feed crop, and the fifth most produced cereal crop worldwide (Leon et al. 2024). Its ability to thrive in semiarid regions is associated with efficient physiological mechanisms that confer drought tolerance (Menezes et al. 2021, Campos et al. 2025).

Soil water availability at different growth stages directly influences sorghum yield by affecting several physiological processes (Ali et al. 2011). The magnitude of drought effects depends on their intensity, duration and particularly on the timing of stress relative to the crop's phenological cycle (Prasad et al. 2008, Campos et al. 2025).

In Brazil, sorghum is grown during the off-season as a second-season crop after soybean, a period often prone to both pre- and post-flowering drought stress (Menezes et al. 2015). In the Southeast and Midwest regions, post-flowering stress is more common; whereas, in the Northeast, pre- and post-flowering drought stress frequently occur due to irregular and poorly distributed rainfall (Guimarães & Landau 2015).

In grain sorghum, pre-flowering drought stress primarily affects panicle size, number of grains and overall yield; by contrast, post-flowering drought stress mainly affects grain size, leaf area and yield (Tolk et al. 2013, Borrell et al. 2014, Menezes et al. 2015, Souza et al. 2021).

Growth analysis is a technique used to identify morphophysiological changes in plants over time and assess photosynthetic efficiency through dry weight accumulation (Menezes et al. 2021). Despite the complexity of plant development,

growth analysis provides a robust framework for evaluating developmental patterns and quantifying the contribution of different physiological processes to plant performance through periodic biomass measurements (Benincasa 2003, Paul et al. 2017).

Evaluating growth in sorghum genotypes under drought conditions is therefore essential for identifying the main morphophysiological effects of drought stress throughout the crop cycle and supporting breeding programs aimed at selecting drought-tolerant genotypes (Prasad et al. 2021). Thus, this study aimed to evaluate the morphophysiological and growth responses of grain sorghum hybrids to drought stress at the pre- and post-flowering stages.

MATERIAL AND METHODS

The trials were conducted at an experimental site in Nova Porteirinha, Minas Gerais state, Brazil (15°48'10"S and 43°18'03"W), in 2017.

The soil at the site is classified as Red-Yellow Latosol (FAO 2022) with medium texture, and the regional climate as Aw, according to Köppen, characterized by dry winters and rainy summers. The region is semiarid, with irregular rainfall distribution, making it suitable for drought tolerance trials, since rainfall takes place between November and March, with a well-defined dry season from May to October. Therefore, water supply during the study was controlled exclusively through irrigation. Daily maximum and minimum temperatures and rainfall during the experiment are presented in Figure 1.

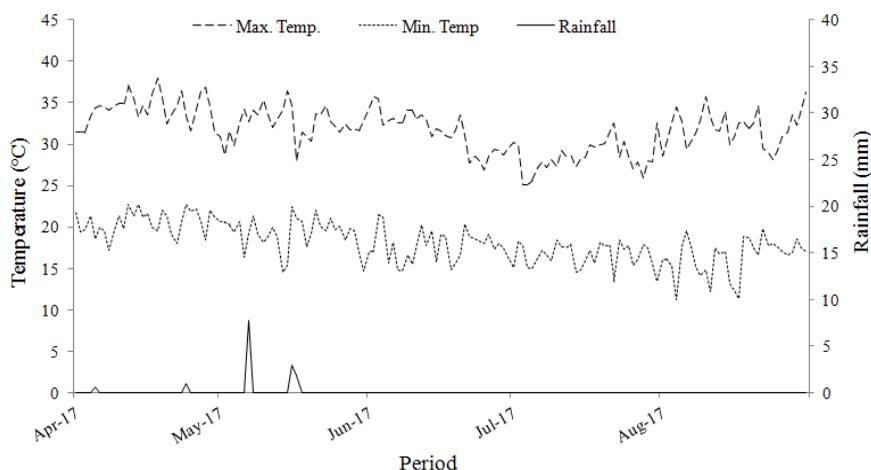


Figure 1. Daily rainfall and maximum and minimum temperatures recorded during the experiment (Nova Porteirinha, Minas Gerais state, Brazil), in 2017. Source: Brasil (2017).

Twenty grain-sorghum hybrids were evaluated (BRS 310, BRS 330, BRS 332, BRS 373, BRS3318, 1167048, 1105661, 1G 100, 1G220, 1G 282, 50A10, 50A70, BM 737, A9904, AG 1040, AG 1080, AG 1090, AS 4625, AS 4639, and DKB 590). These hybrids were grown under three water regimes: well-watered conditions, and pre- and post-flowering drought stress. In the pre-flowering drought stress treatment, irrigation was withheld from 30 (eight-leaf stage) to 60 days after sowing (DAS). In the post-flowering drought stress treatment, irrigation was discontinued at approximately 45 DAS, when most genotypes had reached the boot stage, thereby ensuring that drought stress occurred after flowering. In the well-watered treatment, irrigation was applied until plant maturity. The experiment was arranged in a randomized block design, in a 3 x 20 factorial scheme (water regimes × hybrids), with three replications. The plots comprised four 3-m-long rows, with the two central rows used as the working area.

Soil preparation involved conventional tillage, including one ploughing and two harrowings before planting. The area was then furrowed using a planter-fertilizer set at 0.5-m row spacing. Fertilization followed soil analysis recommendations and crop requirements, with a starter application of 250 kg ha⁻¹ of the 8-28-16 (NPK) formulation and side-dressing of 72 kg ha⁻¹ of N applied at 30 DAS, using urea as the nitrogen source. Sowing occurred in April 2017, with 20 seeds m⁻¹ sown manually. At 20 DAS, the plants were thinned to 10 plants m⁻¹, resulting in a final plant population of 200,000 plants ha⁻¹.

Irrigation was applied using conventional sprinklers spaced 12.0 x 12.0 m apart, operating at 250 kPa of pressure with 4.0 x 2.6 mm nozzles and a flow rate of 1.6 m³ h⁻¹. The total water depth, considering irrigation combined with rainfall, was 400.0 and 345.0 mm under pre- and post-flowering water stress conditions, respectively; whereas, under non-stress conditions, it was 600.0 mm.

The crop management consisted of two manual weedings and insecticide applications to control armyworm. Immediately after flowering, panicles in the working area were protected with polyethylene nets, which were stapled in place to prevent bird damage.

Five evaluations were conducted, starting at the eight-leaf stage (30 DAS) and continuing at 15-day intervals (45, 60, 75, and 90 DAS). Evaluated traits included plant height, leaf area index, shoot dry

weight, and chlorophyll index. The plant height was measured from the ground to the panicle apex, leaf area was estimated as leaf length x width x 0.75 (Petty et al. 2007), and leaf area index was calculated as the total leaf area divided by the ground area occupied by the plants. The shoot dry weight was determined by harvesting the canopy, separating leaves, oven-drying at 65 °C for 72 hours, and weighing the samples on an analytical scale. The chlorophyll index was measured using a SPAD-502 meter on the middle third of the flag leaf, avoiding edges, midribs and areas with chlorosis or necrosis. The grain yield was obtained by weighing the grain mass, correcting it to 13 % of moisture, and converting the results to kg ha⁻¹.

From the leaf area, leaf dry weight and total shoot dry weight, the following parameters were calculated: absolute growth rate - change in the dry weight over time; relative growth rate - change in the dry weight relative to the initial dry weight over time; leaf area ratio - leaf area per unit of dry weight; leaf weight ratio - leaf dry weight per unit of total dry weight; specific leaf area - leaf area per unit of leaf dry weight; and net assimilation rate - dry weight produced per unit of leaf area per unit of time. These growth parameters were estimated following Benincasa (2003).

Growth trait data were subjected to regression analysis as a function of DAS. Linear and polynomial models were tested, with the best-fitting model selected based on the significance of regression coefficients ($p \leq 0.05$), coefficient of determination (R^2), and adjusted R^2 . Model assumptions (normality and homoscedasticity of residuals) were verified through residual analysis. Grain yield data were subjected to analysis of variance (Anova), with water regime and hybrid as fixed effects and blocks as random effects. Significant effects were further analyzed using the Tukey test at 5 % of probability. Boxplots for grain yield were generated using the ggplot2 package in R (R Development Core Team 2016).

RESULTS AND DISCUSSION

No rain occurred during the stress period. Two light rain events were recorded before the pre-flowering stress period, but they did not affect the experiment (Figure 1). The temperature ranged between 25 and 35 °C, values considered optimal for grain sorghum growth (Prasad et al. 2021).

Drought stress significantly reduced plant height. Under well-watered conditions, plants were taller than those exposed to pre- and post-flowering drought stress, with the shortest plants under pre-flowering stress reaching approximately 95 cm at 90 DAS. Initial growth was slow, with plants reaching approximately 38 cm at 30 DAS, and plant height increased linearly between 30 and 75 DAS. After panicle emergence, plant height stabilized (Figure 2a). Height reduction under drought stress may result from decreased cell expansion and division (Ramazanzadeh & Asgharipour 2011). Similar reductions in sorghum height under drought conditions have been reported by Tardin et al. (2013) and Menezes et al. (2015). Since plant height is positively correlated with grain yield in sorghum (Leon et al. 2024), maintaining vegetative growth under drought stress is important for sustaining the yield potential.

The leaf area index increased until approximately 60 DAS, reaching maximum values

under all water regimes. The lowest leaf area index during this period was observed under pre-flowering water stress, with approximately $3.5 \text{ m}^2 \text{ m}^{-2}$. After 60 DAS, the leaf area index declined in all treatments due to leaf senescence, with the reduction being more pronounced under post-flowering drought stress. At 90 DAS, post-flowering-stressed plants exhibited a leaf area index of only $1 \text{ m}^2 \text{ m}^{-2}$, when compared with $3 \text{ m}^2 \text{ m}^{-2}$ under pre-flowering stress (Figure 2b).

Leaf area decreases under drought stress, but growth resumes once water becomes available again (as in the pre-flowering drought treatment) (Seleiman et al. 2021). This behavior confirms the sorghum's ability to enter a state of dormancy during stress and recover when water availability returns (Silva et al. 2023b). In the post-flowering stage, stress severity increases because water replenishment ceases and assimilates are redirected toward grain filling (Wang et al. 2023).

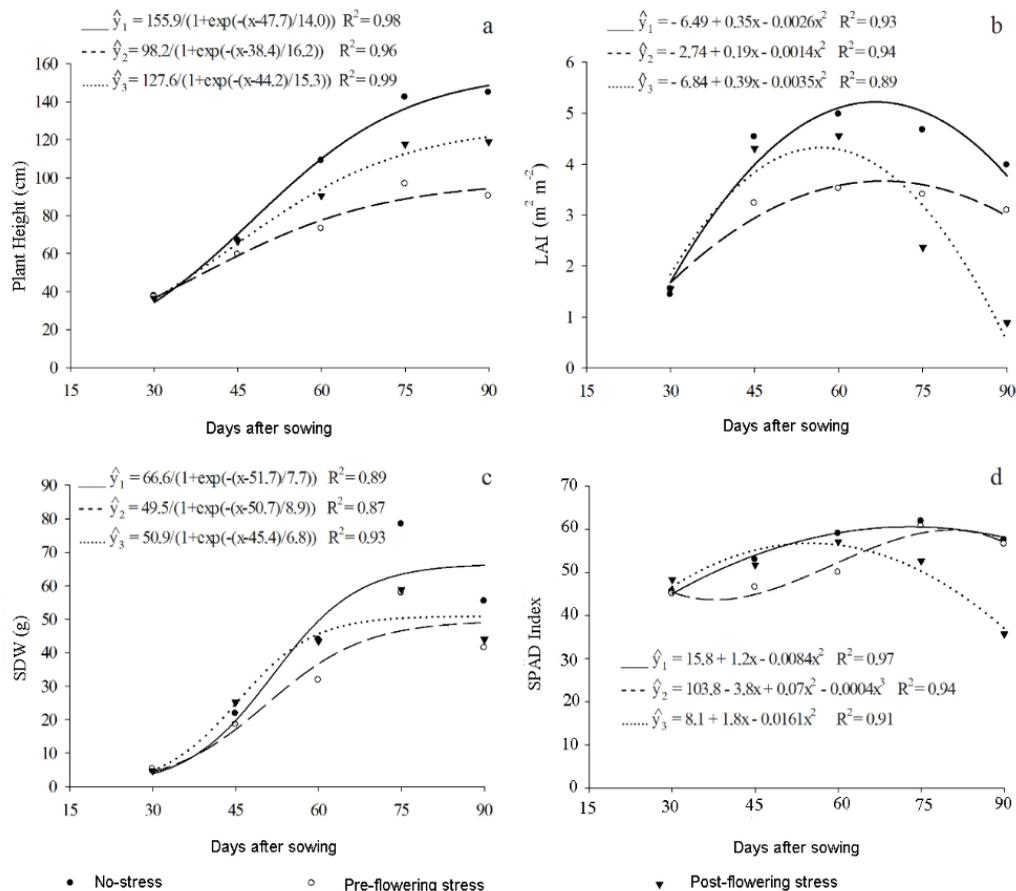


Figure 2. Plant height (a), leaf area index (LAI; b), shoot dry weight (SDW; c), and chlorophyll index (d) in grain sorghum hybrids grown under three water regimes: well-watered (—), pre-flowering drought stress (---), and post-flowering drought stress (····).

Pre-flowering stress reduced the plant growth and delayed new leaf emergence, leading to smaller leaf areas between 30 and 60 DAS. Under post-flowering drought stress, the leaf area reduction was primarily due to leaf senescence, particularly after 60 DAS.

Leaf expansion is driven by cell turgor and is highly sensitive to drought stress (Taiz et al. 2017). Drought-induced inhibition of cell expansion restricts leaf growth and represents one of the plant's primary physiological responses to drought (Taiz et al. 2017). According to Magalhães et al. (2015), leaf area is directly linked to final plant dry matter accumulation, because sorghum growth depends on photosynthetic activity and the total photosynthetically active leaf area.

The shoot dry weight increased slowly at the beginning of the cycle, followed by a linear increase between 45 and 60 DAS (Figure 2c), during which plants almost doubled their dry weight. After 60 DAS, the shoot dry weight gains slowed until 75 DAS, followed by a decline.

The lowest shoot dry weight throughout the cycle occurred under pre-flowering drought stress, although, by 90 DAS, the values under pre- and post-flowering drought stress were similar (Figure 2c). Reductions in dry matter production under drought depend on stress intensity, duration, plant development stage and capacity to recover once favorable conditions are restored (Xu et al. 2010). Reduced shoot growth, mainly due to decreased leaf area, may represent an adaptive response for plants under drought stress, given that it reduces the transpiration surface (Ashraf & Foolad 2007).

The reduction in plant dry weight under drought stress is associated with inhibited photosynthesis and reduced vegetative growth. According to Taiz et al. (2017), water deficiency induces cell turgor loss, triggering stomatal closure, reducing CO₂ assimilation, and inhibiting the photosynthetic process, thereby affecting electron transport, biochemical pathways, and enzyme activity.

Under well-watered conditions, the chlorophyll index increased until 75 DAS, reaching values close to 62. Subsequently, a slight reduction was observed, indicating that under optimal water availability grain sorghum hybrids maintain relatively high chlorophyll levels, even at later growth stages. Under post-flowering drought stress, the index increased only

until 60 DAS, followed by a marked reduction caused by drought stress (Figure 2d).

Under pre-flowering drought stress, the results differed from those observed in the other treatments. Between 30 and 60 DAS, while plants were subjected to water deficit, the chlorophyll index did not increase. Following rehydration at 60 DAS, a significant increase was observed until 75 DAS, followed by a decline.

Overall, these results show that drought stress at any developmental stage negatively affected the chlorophyll index, suggesting a reduced photosynthetic efficiency due to pigment degradation. However, high chlorophyll values are positively correlated with grain yield under drought stress, suggesting a delayed leaf senescence and greater carbon availability for grain filling, which may lead to improved grain size and overall yield (Kassahun et al. 2010, Borrell et al. 2014).

The absolute growth rate reflects the average rate of plant growth over a specific period (Silva et al. 2023a). A quadratic trend was observed for absolute growth rate across all three water regimes (Figure 3a). Between 30 and 60 DAS, the absolute growth rate was positive, coinciding with the period of maximum chlorophyll index values, whereas, in the well-watered treatment, it increased until 60 DAS, then declined and became negative at 90 DAS (approximately -1 g day⁻¹). Under pre-flowering drought stress, the absolute growth rate increased less markedly than in the other treatments between 30 and 60 DAS, whereas post-flowering stress caused a sharp decline after 60 DAS.

The absolute growth rate generally coincided with the onset of flowering, which occurred between 55 and 72 DAS. A higher absolute growth rate near the flowering period in sorghum was also reported by Ramazanadeh & Asgharipour (2011), in their evaluation of sorghum genotypes with different growth cycles.

The relative growth rate reflects the increase in dry mass per unit of initial dry weight over a given period (Santos et al. 2023). The maximum relative growth rate typically occurred early in the crop growth cycle (30 DAS) and declined as development progressed. This initial peak is associated with the predominance of young leaves exhibiting high photosynthetic capacity and rapid growth (Benincasa 2003, Aumonde et al. 2011). In the present study, the

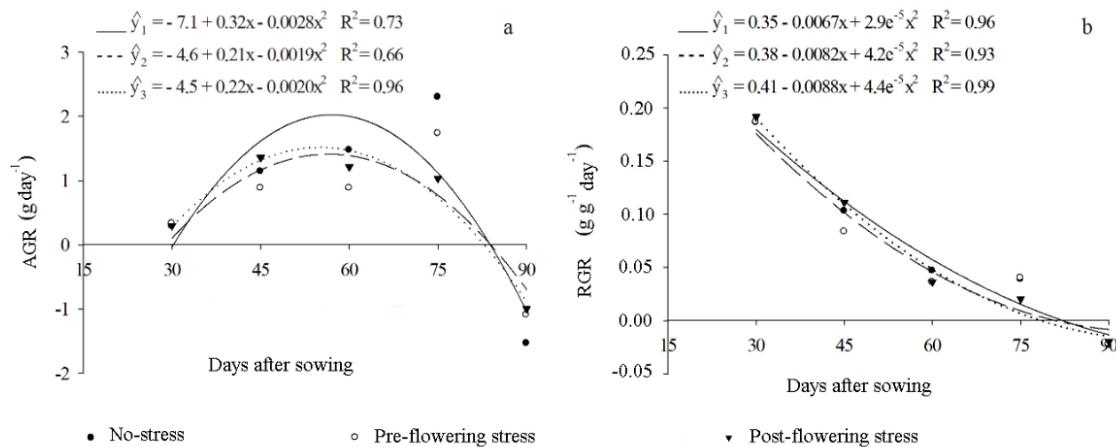


Figure 3. Absolute growth rate (AGR; a) and relative growth rate (RGR; b) in grain sorghum hybrids grown under three water regimes: well-watered (—), pre-flowering drought stress (---), and post-flowering drought stress (···).

relative growth rate was highest at 30 DAS across all water regimes and declined progressively, becoming negative at 90 DAS (Figure 3b), reflecting the natural decline in plant dry mass accumulation associated with reduced photosynthetically active leaf area (Santos et al. 2023).

Under pre-flowering drought stress, the relative growth rate was lower between 30 and 60 DAS when irrigation was withheld. By contrast, under post-flowering drought stress, it declined after 60 DAS (Figure 3b), demonstrating that the drought stress reduced the dry biomass accumulation. The results for relative growth rate and SPAD chlorophyll index in the sorghum hybrids under pre-flowering drought stress indicated the crop's capacity to resume growth once water availability was restored.

Under both well-watered and pre-flowering stress conditions, the leaf area ratio in sorghum decreased from about 160 cm² g⁻¹ at 30 DAS to approximately 40 cm² g⁻¹ at 75 DAS, followed by a slight increase until 90 DAS. By contrast, under post-flowering stress, it continued to decline throughout the evaluation period (Figure 4a).

Leaf area ratio represents the leaf area available for photosynthesis, reflecting the relationship between the leaf surface intercepting light and CO₂ and the total dry mass produced by the plant (Yin et al. 2019). This index is a morphophysiological trait resulting from the interaction between specific leaf area (morphological component) and leaf mass ratio (physiological component), and therefore varies according to plant structure and environmental

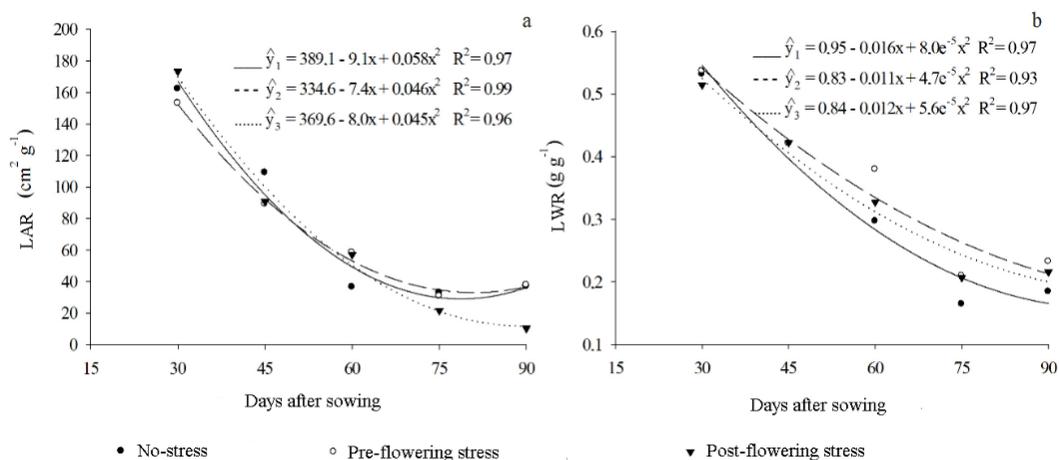


Figure 4. Leaf area ratio (LAR; a) and leaf weight ratio (LWR; b) in grain sorghum hybrids grown under three water regimes: well-watered (—), pre-flowering drought stress (---), and post-flowering drought stress (···).

conditions (Santos et al. 2023). The observed pattern in sorghum is expected, since upper leaves progressively shade lower leaves, thereby reducing the effective photosynthetic area (Benincasa 2003).

Leaf weight ratio expresses the fraction of biomass retained in leaves rather than allocated to other plant organs, considering that leaves are the primary sites of biomass production on which the growth of the remaining plant organs depends (Benincasa 2003). In this study, the leaf weight ratio decreased throughout the evaluation period across all water regimes. The smallest value was observed under well-watered conditions, whereas the highest one occurred under pre-flowering drought stress (Figure 4b). According to Vilela & Büll (1999), higher leaf weight ratio values in plants subjected to water deficit are related to reduced export of assimilates from leaves to other plant organs. These authors also reported higher leaf weight ratio values throughout the crop cycle in maize plants grown under water deficit.

Thus, the decreasing trend in leaf weight ratio indicates that leaves acted as the primary metabolic sink during the early stages of the crop cycle, due to the greater investment in leaf development to increase the effective photosynthetic area required for biomass production. After flowering, reproductive structures became the dominant metabolic sinks, exhibiting a high capacity for assimilate mobilization (Taiz et al. 2017).

The specific leaf area also declined throughout the evaluation period under well-watered and post-flowering drought stress conditions. Under well-

watered conditions, the reduction in specific leaf area among the hybrids was less pronounced. By contrast, under post-flowering drought stress the decline was more pronounced, especially after 60 DAS, when the hybrids were under water deficit. Conversely, under pre-flowering drought stress the most pronounced reduction in specific leaf area occurred during the stress period (30-60 DAS). Following rewatering, a slight increase was observed between 75 and 90 DAS (Figure 5a). A reduction under drought conditions was also reported by Liu & Stützel (2004), who observed that lower specific leaf area promotes the formation of thicker leaves, which aids in water conservation by reducing the surface-area-to-volume ratio. Since specific leaf area reflects the balance between biomass production and nutrient conservation efficiency and is strongly associated with carbon fixation capacity and turgor loss point, it is considered a key parameter in plant-water relations (Ramos Junior et al. 2013).

Net assimilation rate is a growth parameter that quantifies biomass accumulation per unit of leaf area over time (Benincasa 2003), reflecting the efficiency of the photosynthetic apparatus in converting assimilated carbon into plant biomass (Barbero et al. 2013). In this study, the net assimilation rate exhibited similar patterns across the three water regimes, with a slight reduction around 45 DAS, followed by an increase up to 75 DAS, and then a subsequent sharp decline, reaching negative values at 90 DAS.

Net assimilation rate is influenced by leaf area, canopy architecture, leaf orientation, and assimilate translocation and partitioning (Aumonde et al.

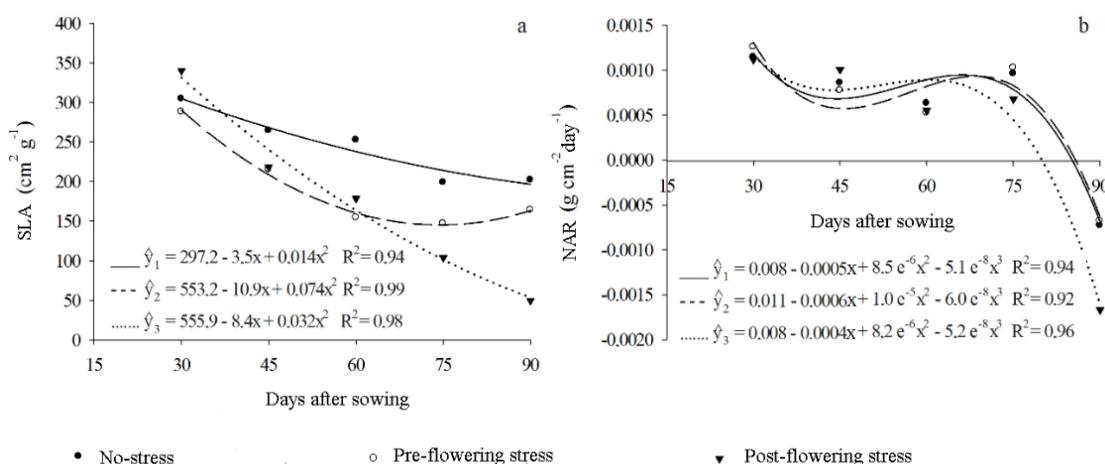


Figure 5. Specific leaf area (SLA; a) and net assimilation rate (NAR; b) in grain sorghum hybrids grown under three water regimes: well-watered (—), pre-flowering drought stress (---), and post-flowering drought stress (···).

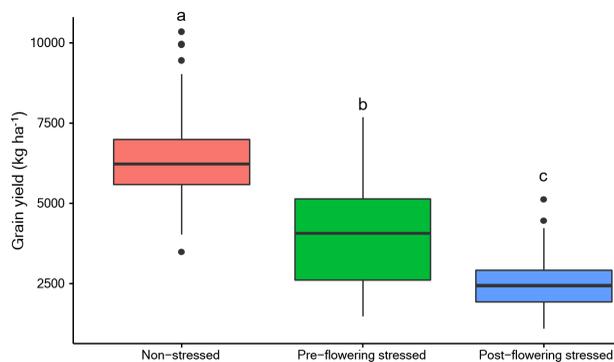


Figure 6. Grain yield of sorghum hybrids grown under three water regimes: well-watered (no-stress), pre- and post-flowering drought stress. Treatment means differed according to the Tukey test ($p < 0.05$).

2011). Typically, it decreases with plant age due to the progressive development of non-assimilating structures and shading of lower leaves within the canopy (Almeida et al. 2022). These results indicate that the post-flowering drought stress exerted a stronger negative effect on net assimilation rate than pre-flowering stress, primarily due to greater reductions in leaf area and chlorophyll content (chlorophyll index) toward the end of the crop cycle.

The grain yield was markedly reduced under drought stress (Figure 6). The lowest yield occurred under post-flowering drought stress, demonstrating that sorghum is more sensitive to drought during the reproductive stage than during vegetative growth.

Yield reduction under pre-flowering drought stress was associated with reductions in plant height, leaf area index, and shoot dry weight; whereas, under post-flowering drought stress, the yield was mainly influenced by reductions in leaf area index, SPAD chlorophyll index, and net assimilation rate. The decline in leaf area index under both stress conditions underscores its importance as a key trait for selecting drought-tolerant sorghum hybrids. A reduction in leaf area and photosynthetic activity ultimately lowers the net assimilation rate, leading to reduced assimilate availability for grain filling and, consequently, smaller grain size under post-flowering drought stress.

CONCLUSIONS

1. Drought stress significantly affected the morphophysiological traits and growth parameters of the evaluated grain sorghum hybrids;

2. The reduced photosynthetic efficiency, leaf area index, and net assimilation rate resulted in lower biomass accumulation and, consequently, smaller grains and reduced yield, particularly when water deficit occurred during the post-flowering stage;
3. Differences among hybrids and across developmental stages indicated that certain genotypes exhibit a greater tolerance to drought conditions.

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