



Effect of Supra-optimal Temperature on Seed Germination of Selected Sorghum Genotypes

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Abstract

Sorghum (*Sorghum bicolor*) is the most important traditional cereal crops of the hotter and drier regions. Its production is however affected by both abiotic and biotic factors. Among abiotic factors heat stress caused by supra-optimal temperatures affect seed germination, which ultimately leads to yield loss. The objectives of this study were therefore to i) evaluate the impact of supra-optimal temperature on seed germination and ii) identify a sorghum genotype with a highest resilience to germinate under heat-stressed condition. To achieve this objectives four popular sorghum genotypes Kuyuma, Sima, Vumba and ZSV 36R were tested and planted following a completely randomised design (CRD) replicated three times under two experimental temperature regime of 25° (Optimum) and 37°C (Supra-optimal). Data was collected on normal seedlings, abnormal seedlings, root length and shoot length. Results showed significance differences between the grand mean responses for all measured parameters across genotypes in the two temperatures regimes (Paired t-test, $P < 0.01$). Abnormal seedlings counted was the most affected parameter under a supra-optimal temperature exposure exhibiting a percentage increase of 183.2%, when compared to the optimum temperature environment. The genotypes ZSV 36R was identified as the most resilient performing genotype under heat stress.

Keywords: *Sorghum bicolor*, Temperature, Abnormal seedling, Impact, heat stress.

Introduction

Sorghum (*Sorghum bicolor*.) is an important traditional cereal crops in sub-Saharan Africa rich in carbohydrates, fiber, minerals and a gluten free crop (Christiansen, *et al.*, 2006). Globally it is the fifth most important cereal and second in sub-Saharan Africa after maize (Obalum, *et al.*, 2011). In Zambia sorghum is a staple food crop after maize, rice and sorghum varieties have been released by Zambia Agriculture Research Institute (ZARI) (Munkombwe *et al.*, 2020). Sorghum flour is used for making thick porridge, snacks, brewing of alcoholic, non-alcoholic beverages, ethanol fuel as well as livestock feed (Christiansen, *et al.*, 2006).

Sorghum yields among small scale farmers in sub-Saharan Africa are still low with an average of 0.85 t ha⁻¹ compared to potential yields of 3 to 5 tons per hectare (Muui *et al.*, 2013; ZAMSEED, 2020). The low productivity is attributed to both abiotic and biotic factors. Among the abiotic factors, exposure of sorghum crop to supra-optimal temperature is of great concern (Shah, *et al.*, 2017).

Temperature affects a variety of physiological processes in many plants, sorghum inclusive (Al-Shoaibi, 2020). It affects all phases of plant growth, from germination and early plant growth to canopy closure, photosynthesis and chlorophyll fluorescence. Optimum temperature range for germination in

sorghum ranges from 20 °C to 32 °C (Sari and Juniarti, 2023). Specifically, studies have shown that heat stress due to supra-optimal temperatures (above 32 °C) reduces fertility, grain quality, weighty, seed germination, ultimately leading to yield loss (Hill, *et al.*, 2022; Sari and Juniarti, 2023). In evaluating for germination, normal seedlings emerged, abnormal seedlings root length and shoot length were identified as key parameters to consider (Don, 2018). However, the effect of supra-optimal temperature on these parameters among sorghum genotypes is not clearly understood. The objectives of this study were therefore to i) evaluate the impact of supra-optimal temperature on seed germination and ii) identify a sorghum genotype with a highest resilience to germinate under heat-stressed condition.

Material and Methods

Location of study and germplasm used

The experiment was conducted at the Seed Control and Certification Institute (SCCI) (S 15° 34.0801 and E 28° 16.2785) in Chilanga, Zambia. The study was conducted in two independent chambers (sets), at different temperatures regimes, 25°C and 37°C (Optimum and supra-optimal temperature regimes respectively) for ten (10) days. Four popular genotypes of sorghum were used in the study: Kuyuma, Sima, Vumba and ZSV 36R.

Conduct of experiment

The experiment was laid out as a completely randomized design (CRD) in each germination chamber consisting of four genotypes (Kuyuma, Sima, Vumba and ZSV 36R) replicated three times, giving 12 experimental units per chamber. Thirty seeds for each genotype were plated per rep on sterilised moist sand, placed on 16 cm (diameter) x 8 cm (height) plastic pots

Data collection and analysis

Data was collected on all experimental units on: Normal seedlings (NS) counted, Abnormal seedlings (ABS) counted, shoot length and root length. Abnormal seedlings were identified as described by Don and Ducournau, (2018). Root and shoot length were measured using a metric centimetre (cm) ruler as done by Ndeke and Tembo (2019). The comparisons in the mean performance for each parameter in the two experimental set was analysed using a paired t- test. The impact of supra-optimal temperature for each measured parameter was assayed using the formula:

$I = ([a-b]/a) * 100$. Where

- I- Impact percentage in supra-optimal temperature
- a- Parameter mean performance in optimum temperature
- b- Parameter mean performance in supra-optimum temperature

Furthermore data collected within each experimental set was analysed using analysis of variance in GenStat statistical 18th edition software. Means was separated using fisher protected least significant difference (LSD) $\alpha = 0.05$

Results

Genotypic performance of measured parameters.

A comparison of grand mean performance for all parameter response between two temperatures regimes across genotypes revealed significant differences ($p < 0.05$) (Table 1) with regard to Normal seedlings counted, Abnormal seedlings counted, Root length and Shoot Length. Abnormal seedlings counted was the most affected parameter exhibiting a percentage increase of 183.2% under supra-optimal temperature when compared to the optimum temperature regime (Table 1). Observed representative appearance of seedlings across genotypes were observed and are as shown on plate 1.

Table 1: Comparison for mean performance of measured parameters across genotypes in optimum and supra-optimal temperature chambers at SCCI using paired t-test.

Parameters	Opt [*]	Supra ^y	Impact ^z	P- value
NS (count)	21.25	6.73	68.3	<0.001
AS (count)	3.33	9.43	183.2	<0.001
RL (cm)	17.03	10.1	40.7	0.008
SL (cm)	14.8	6.7	54.7	0.005

Foot Note- NS- Normal seedlings; AS- Abnormal seedlings; RL- Root length; SL- Shoot length, ^{*}- optimum temperature, 25 °C chamber, ^y- Supra-optimal temperature, 37°C, ^z- Impact in percentage calculated as comparison of performance in optimum, 25° C chamber and Supra-optimal temperature, 37° C

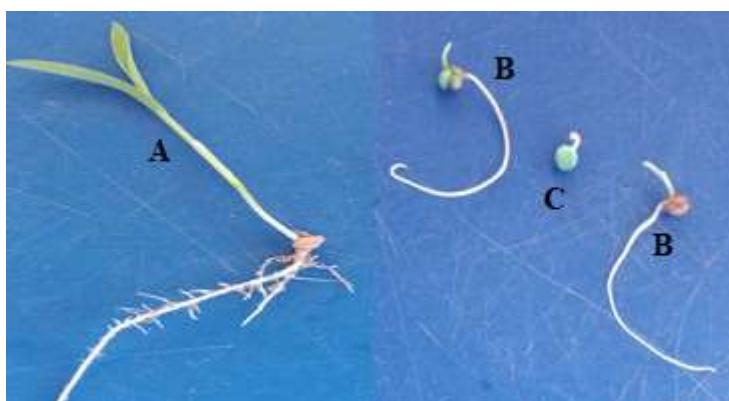


Plate 1: Observed representative normal and abnormal seedlings observed. A: Normal seedling: all the essential structures or features well developed, complete in proportion to each another and healthy. B: Abnormal seedling: Poorly developed coleoptile. C: Poorly developed coleoptile and no radicle

Analysis of variance for optimum experimental set 25°C showed that they were significant differences ($P < 0.05$) with respect to measured parameters among genotypes except for abnormal seedlings counted (Table 2). On the other hand, analysis of variance for a supra-optimal experimental set at 37°C, showed that they were no significant

difference with respect to measured parameters among genotypes except for 'normal seedlings' ($P < 0.05$) (Table 3). The genotypes ZSV 36R was identified as the best resilient genotype to germinate under heat stress. (Table 4 and 5). It was the best performer in both the optimum and supra-optimal set regimes.

Table 2: ANOVA of mean squares of measured parameter in sorghum evaluated at SCCI at, optimum temperature (25 °C)

Source of Variance	DF	NS (count)	ABS (count)	RL (cm)	SL(cm)
Genotype	3	10.53*	1.56 ^{ns}	5.29*	6.55**
Error	8	2.33	1.75	1.02	0.67
Total	11				

^{*}, ^{**} - data significantly different at $P = 0.05$, $P = 0.01$ respectively. Not significant ^{ns}. NS- Normal Seedlings, AS- Abnormal Seedlings, RL- Root Length, SL- Shoot Length

Table 3: ANOVA of mean squares of parameters in Sorghum evaluated at SCCI at supra-optimal temperature (37 °C)

Source of Variance	DF	NS (count)	ABS (count)	RL (cm)	SL (cm)
Genotype	3	30.08**	2.31 ^{ns}	2.60 ^{ns}	5.06 ^{ns}
Error	8	3.33	2.75	4.75	3.32
Total	11				

** - Significant at P = 0.01. Not Significant ^{ns}. NS- Normal Seedling, AS- Abnormal Seedlings, RL- Root Length, SL- Shoot Length. SOV- Source of variation.

Table 4: Genotypic mean performance on all measured parameters at optimum temperature, (25°C)

Sorghum Genotype	NS (count)	ABS (count)	RL (cm)	SL (cm)
Kuyuma	20.33	3.00	15.83	12.63
Sima	21.67	4.33	18.17	15.9
Vumba	19.33	3.33	3.33	15.3
ZSV 36 R	23.67	2.67	2.67	15.43
LSD ($\alpha = 0.05$)	2.88	-	1.9	1.54

LSD least significant difference test performed at P = 0.05, NS- Normal Seedling (NS), Abnormal Seedling (ABS), Root Length (RL) and Shoot Length (SL)

Table 5: Genotypic mean performance on all measured parameters at supra-optimal temperature (37°C)

Sorghum Genotype	NS (count)	ABS (count)	RL (cm)	SL (cm)
Kuyuma	6.33	9.00	10.47	7.33
Sima	5.00	11.67	8.93	6.2
Vumba	4.33	9.33	9.9	5.17
ZSV 36R	11.33	7.67	11.9	8.13
LSD ($\alpha = 0.05$)	3.65	-	-	-

LSD- least significant difference test performed at P = 0.05, NS- Normal Seedling, AS- Abnormal RL- Root Length, SL- Shoot Length

Discussion

Heat stress has an effect on crop productivity. Lately effect of climate change has led to increase in prevailing temperature leading to yield losses in crop productivity (Tandzi and Mutengwa, 2020). It has been established that temperature affects a variety of physiological processes including germination, photosynthesis and chlorophyll fluorescence in sorghum (AL-Shoaibi, 2020). Consequently, yield declines beyond the threshold of 32°C, and for every further increase of 1°C the yield drops by about 8.4% (Adhikari, *et al.*, (2015).

In this study, supra-optimal temperature was observed to reduce germination effect in sorghum seed. This was evident from the results obtained using paired t-test (Table 1) that showed better response for all measured parameters across genotypes in optimum temperature set (25°C) than in the

supra-optimal temperature set (37 °C) (P < 0.001). The greatest impact with supra-optimal temperature was observed on abnormal seedlings counted exhibiting 183.2 % increase when compared to the seedling response across genotypes under optimum temperature.

The poor performance of all genotypes in the supra-optimal environment set (37 °C) may be due to enhanced Abscisic Acid (ABA) production which leads to reduced production of gibberellic acid (GE) (Toh, *et al.*, 2008). It was suggested that GA exerts its influence in two ways, first by increasing the growth potential of embryo and by inducing hydrolytic enzymes, which in turn enhances germination effect (Gupta and Chakrabarty, 2013). In this study it can therefore be hypothesised that the effect on abnormal seedlings, shoots and roots in supra-optimal temperature environment was due to

inhibited expression of GA 20-oxidase genes which encodes for GA production (Wang, *et al.*, 2020, Yongli. 2020, Barnabás, *et al.*, 2007). Further analysis revealed that ZSV 36R was the most resilient genotype under the supra-optimal exposure. It also exhibited less abnormal seedlings compared to other genotypes (Table 5). Implying that the genotype can be the candidate of choice under heat-stressed conditions. Furthermore, ZSV 36R can be crossed with a least resilient genotype (Vumba) to heat stress, creating a mapping population essential in identifying quantitative trait loci linked to the effect of germination under heat stress (Tembo, *et al.*, 2014, Mbwando, *et al.*, 2016).

Evaluating for germination effect under field conditions is a challenge owing to variation and fluctuations in soil temperature and texture (Stewart, *et al.*, 2021). However it is imperative to investigate if normal seedling, abnormal seedling count, root length and shoot length utilised under laboratory conditions can be extended to be used as an indirect selection criteria for genotypic grain yield performance under field conditions in sorghum. Previous studies suggested that indirect selection can be employed for selection of traits (in this regard yield) that are difficult to select for (Tembo, *et al.*, 2016).

Conclusion

Exposing sorghum seed to supra-optimal temperature significantly affect seed germination. The largest effect on key parameters was with abnormal seedlings counted across genotypes which exhibited a percentage increase of 183.2% under supra-optimal temperature when compared to the optimum temperature regime. The genotypes ZSV 36R was identified as the best resilient performing genotype under heat stress.

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