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Effects of Water Stress on Chlorophyll Content and Yield Performance of Selected Cowpea (Vigna Unguiculata) Genotypes Under Controlled Conditions

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Abstract
Cowpea (*Vigna unguiculata* (L.) Walp) is a drought-tolerant legume vital for food security in Zambia, but its production is constrained by water stress. This study evaluated the physiological and agronomic responses of three cowpea genotypes (Msandile, Bubebe, and Namuseba) to different water regimes. A completely randomized design with three replications was used. Plants were subjected to three weekly water levels: 1000 mL (control, simulating Agro-Ecological Zone III), 800 mL (moderate stress, AEZ II), and 600 mL (severe stress, AEZ I). Chlorophyll content (SPAD), pod number, grain weight, and biomass were measured. Water stress significantly ($p < 0.05$) reduced all measured parameters. Chlorophyll content declined with increasing stress; Msandile and Namuseba showed higher values under control conditions (56.9 and 54.43, respectively) but Msandile experienced the sharpest decline under severe stress. Pod number and grain weight followed similar trends, with Msandile showing the highest yield under control (41.67 pods, 0.27 g/plant) but poor performance under stress. Namuseba demonstrated the most stable yield performance across stress levels. A strong positive correlation was found between chlorophyll content and pod count ($r = 0.82$), pod weight ($r = 0.76$), and biomass ($r = 0.70$). The genotypes exhibited differential tolerance to water stress. Namuseba is recommended for drought-prone environments due to its stability, while Msandile is suitable for high-potential areas. Chlorophyll content is a reliable physiological marker for screening drought tolerance in cowpea.

Keywords: Cowpea, Drought, Genotypic Variation SPAD, Yield Components, Zambia

1. Introduction
Cowpea (*Vigna unguiculata* (L.) Walp) is a crucial legume for food security and soil fertility in sub-Saharan Africa, particularly in semi-arid regions [1]. Its drought tolerance makes it a strategic crop for climate-resilient agriculture. However, water stress remains a primary abiotic constraint, leading to significant reductions in photosynthetic efficiency and yield [2, 3]. Chlorophyll content, a key indicator of photosynthetic capacity, is highly sensitive to water deficit and serves as a reliable proxy for plant health and stress tolerance [4, 5].
In Zambia, cowpea production is predominantly rain-fed, making it vulnerable to increasing rainfall variability. While genotypic variation for drought tolerance exists, the physiological responses of locally adapted varieties like Msandile, Bubebe, and Namuseba are not well-documented. Understanding how these genotypes maintain chlorophyll content and yield under stress is critical for varietal selection and breeding [6, 7].
This study aimed to:
1. Determine the effect of water stress on chlorophyll content
2. Assess its impact on yield parameters (pod count, grain weight, and biomass)
3. Analyze the relationship between chlorophyll content and yield performance in three selected cowpea genotypes.

2. Materials and Methods
2.1 Experimental Site and Design
The study was conducted in a greenhouse at Mount Makulu Research Station, Zambia (15.547609°S, 28.245138°E). A Completely Randomized Design (CRD) with a 3 × 3 factorial arrangement (3 genotypes × 3 water regimes) and three replications was used, totaling 27 experimental units.

2.2 Plant Materials and Treatments
The cowpea genotypes used were **Msandile**, **Bubebe**, and **Namuseba**. The water regimes applied weekly were:
1000 mL (Control, no stress)
800 mL (Moderate stress)
600 mL (Severe stress)
Water stress treatments commenced two weeks after germination. Seeds were sown in pots containing 10 kg of sandy loam soil. Standard agronomic practices were followed without fertilizer or pesticide application.

2.3 Data Collection
Chlorophyll Content
Measured at flowering using a SPAD-502 Plus meter on the uppermost fully expanded leaves.

Pod Count
Total number of pods per plant at maturity.

Grain Weight
Weight of grains per plant after harvesting and shelling.

Biomass
Total above-ground plant dry weight at maturity.

2.4 Statistical Analysis
Data were analyzed using Analysis of Variance (ANOVA) in SPSS v26. Treatment means were separated using Fisher's LSD test at $p < 0.05$. Pearson's correlation analysis was performed to assess relationships between chlorophyll content and yield parameters.

3. Results

Table 1: Mean values of measured parameters for three cowpea genotypes under different water regimes

Genotype	Water (mL/wk)	Chlorophyll (SPAD)	Pod Count	Grain Weight (g)	Biomass (g)
Bubebe	1000	43.93	39.00	0.13	0.93
	800	47.33	36.67	0.11	0.66
	600	39.33	31.67	0.09	0.53
Msandile	1000	56.90	41.67	0.27	0.77
	800	46.33	34.00	0.13	0.58
	600	32.67	20.33	0.09	0.42
Namuseba	1000	54.43	36.67	0.13	0.88
	800	43.67	35.33	0.12	0.71
	600	38.33	33.33	0.10	0.59

3.1 Chlorophyll Content
Water stress significantly ($p = 0.00076$) reduced chlorophyll content across all genotypes.
(Table 1). Under control conditions, Msandile (56.9) and Namuseba (54.43) had the highest SPAD values. Under severe stress (600 mL), all genotypes showed a decline, with Msandile experiencing the most substantial reduction (32.67). Bubebe showed relative stability under stress (Fig. 1A).

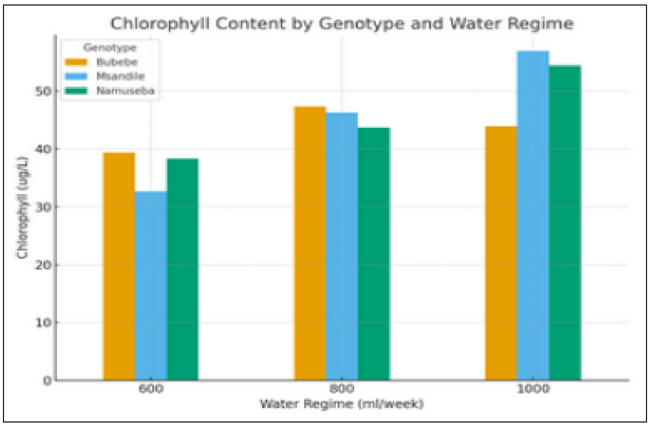


Fig 1A

3.2 Yield Parameters
Water stress significantly reduced pod number ($p = 0.00011$) and grain weight ($p = 0.003118$). Msandile produced the highest pod count (41.67) and grain weight (0.27 g) under control but showed a drastic drop under severe stress (20.33 pods, 0.09 g). Namuseba maintained more stable pod numbers and grain weight across stress levels (Fig. 1B, 1C). Biomass was also significantly reduced by stress ($p = 0.000000000253$), with Namuseba consistently producing the highest biomass under stress conditions.

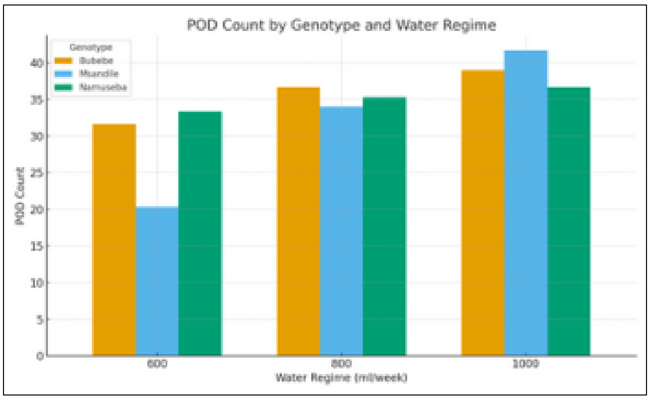


Fig 1B

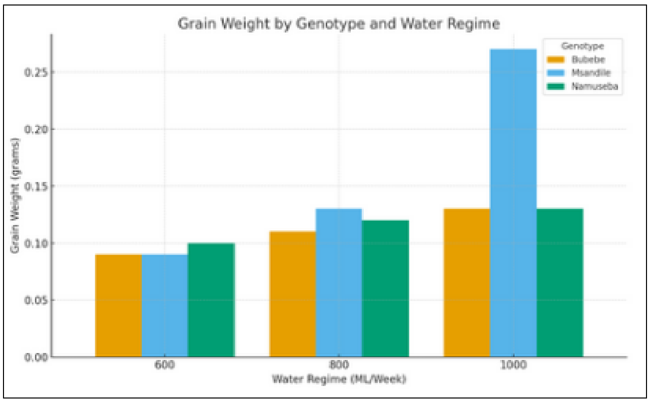
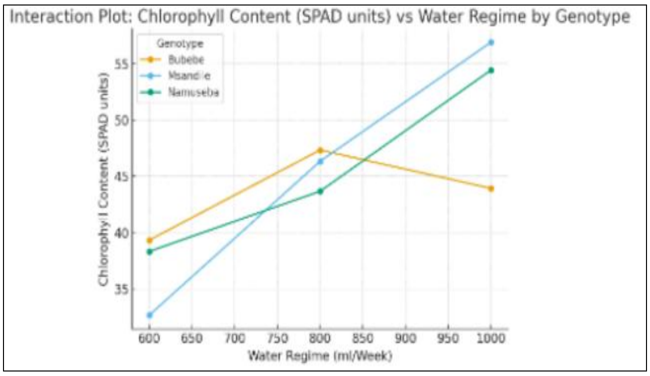


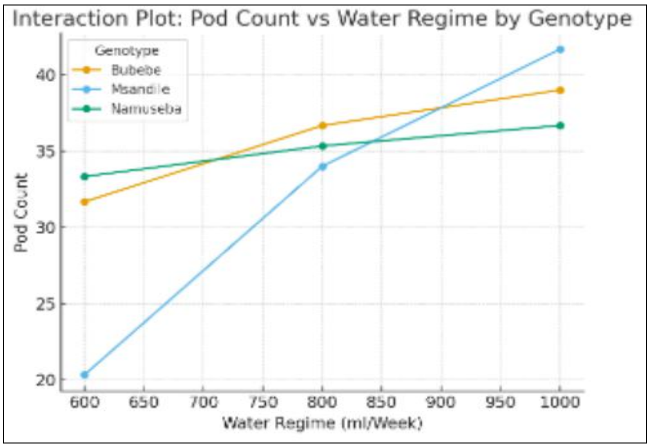
Fig 1C

3.3 Genotypic Differences and Correlation

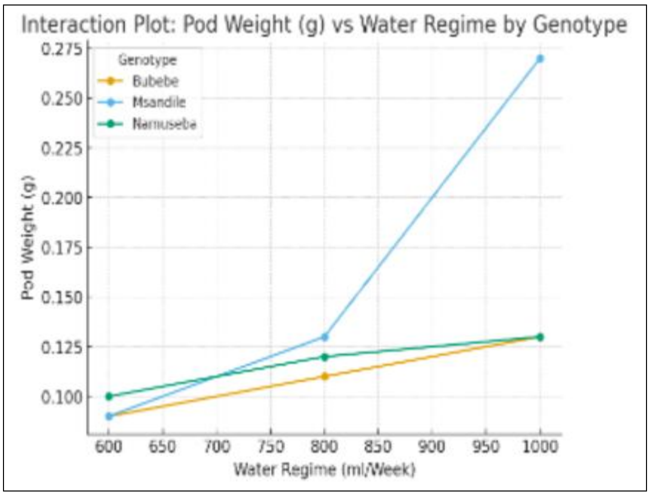
Interaction plots showed largely parallel responses, indicating that the main effect of water stress was consistent across genotypes, with no significant genotype-by-water interaction for most traits. However, Namuseba consistently performed better under stress.



Interaction plot Fig. 3A



Correlation analysis revealed strong positive relationships between chlorophyll content and pod count ($r = 0.82$, $p < 0.01$), grain weight ($r = 0.76$, $p < 0.01$), and biomass ($r = 0.70$, $p < 0.01$).



Interaction plot Fig

4. Discussion

The decline in chlorophyll content under water stress is consistent with previous studies [4, 5], attributed to oxidative damage and impaired chloroplast function. The high initial chlorophyll in Msandile and Namuseba suggests greater photosynthetic potential, but Msandile's sharp decline indicates sensitivity. Bubebe's relative stability may be due to better antioxidant protection [8]. In the present study, Msandile and Namuseba exhibited higher chlorophyll levels under control conditions, reflecting greater photosynthetic potential. However, Msandile showed a sharper decline under severe stress compared to Bubebe, which remained relatively stable chlorophyll content. This pattern supports the view that genotypic variability in chlorophyll retention is a key physiological marker of drought tolerance (Nkebiwe *et al*., 2022; Mwale *et al*., 2023) [5, 7].

Yield components (pod count and grain weight) followed a similar declining trend with increasing water stress. Msandile, which produced the highest pod count (41.67) and grain weight (0.27 g) under control conditions, showed drastic yield reductions under stress, especially at 600 ml/week. This indicates that Msandile is a high-potential but stress-sensitive variety, suitable for environments with reliable rainfall or irrigation.

Namuseba, on the other hand, showed more stable yield performance across stress levels, maintaining relatively higher pod counts (33.33 at 600 ml/week) and grain weight compared to Msandile. This suggests that Namuseba possesses stress-adaptive traits, possibly through early maturity or efficient assimilate partitioning. This finding aligns with Mofokeng *et al.* (2021), who observed that certain cowpea varieties exhibit yield stability under drought due to drought escape mechanisms.

Bubebe consistently produced lower yields across treatments but showed moderate stability, implying that it may not be the most productive under favourable conditions but could perform relatively reliably in marginal environments. Msandile could serve as a parental line for improving yield potential, but it requires introgression of drought-tolerance traits to withstand stress. Namuseba shows promise as a drought-tolerant variety due to its ability to sustain pod production under water stress, making it suitable for drought-prone areas. Bubebe, while less productive, may harbour physiological traits (e.g., chlorophyll stability) that could be useful in breeding programs targeting stress resilience. This aligns with Kamara *et al.* (2021) [6] and Singh *et al.* (2022) [9], who emphasized the need to combine high-yielding and stress-tolerant genotypes in breeding pipelines to produce resilient cowpea varieties.

The positive correlation observed between chlorophyll content and yield parameters reinforces the role of chlorophyll as a physiological predictor of productivity under stress. Similar findings were reported by Nkebiwe *et al.* (2022) [5], who found significant correlations between SPAD values and grain yield in cowpea under drought.

The severe yield reduction in Msandile under stress highlights its sensitivity despite high yield potential. Namuseba's stable performance suggests the presence of drought-adaptive traits, such as efficient assimilate partitioning or osmotic adjustment [6, 9]. The strong correlation between chlorophyll content and yield parameters confirms SPAD readings as a valuable, non-destructive tool for early screening of drought-tolerant genotypes [5, 7].

5. Conclusion and Recommendations

Water stress significantly impairs chlorophyll content and yield in cowpea, but genotypic differences offer opportunities for mitigation. **Namuseba** is recommended for drought-prone areas due to its stability. **Msandile** is suitable for high-potential regions with reliable moisture. **Bubebe** may contribute stress-resilience traits for breeding.

Breeding programs should integrate physiological traits like chlorophyll content for efficient selection. Future research should validate these findings in multi-location field trials and investigate the molecular basis of the observed tolerance in Namuseba.

6. References

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