



# Drought tolerance assessment of Bambara groundnut (*Vigna subterranea* L. Verdc) accessions based on morphological and physiological traits

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## ABSTRACT

Understanding the adaptive responses of Bambara groundnut (*Vigna subterranea* L. Verdc) under water-limited conditions is vital for improving resilience of this crop in semi-arid agriculture. This study evaluated the growth, yield, physiological, and photochemical responses of 24 Bambara groundnut accessions under drought stress and non-stress conditions. Significant variation ( $p < 0.05$ ) was observed among accessions for key morphological traits such as leaf length, plant height, and petiole number. Drought stress markedly reduced biomass and seed yield, with Acc 97 and Acc 100 demonstrating superior yield under stress. Physiological responses showed significant declines in photosynthetic rate, stomatal conductance, and chlorophyll fluorescence parameters under drought. However, accessions such as Acc 25, Acc 61, and Acc 87 maintained relatively higher water-use efficiency and photochemical stability, indicating better drought adaptation. Drought indices highlighted Acc 200, Acc 190, and Acc 175 as promising genotypes, combining high yield potential and stress tolerance. Principal component analysis revealed that photosynthetic efficiency and biomass traits predominantly contributed to phenotypic variation under stress. Overall, this study identified genetically diverse and drought-resilient Bambara groundnut accessions with potential for targeted breeding programs aimed at enhancing drought tolerance and securing food production in marginal environments.

## 1. Introduction

Bambara groundnut (*Vigna subterranea* (L.) Verdc.), a leguminous crop native to sub-Saharan Africa, is known for its exceptional drought tolerance and adaptability to marginal environments (Chai et al., 2016). The crop produces stable yields under limited water conditions, thus playing an essential role in the legume food systems of arid and semi-arid regions. (Chibarabada et al., 2014). Its drought tolerance and nutritional value position Bambara groundnut as a strategic crop for improving food security in areas most vulnerable to climate variability (Mayes et al., 2019).

Despite its potential, Bambara groundnut remains underutilized and under-researched. Smallholder farmers cultivate unimproved landraces, valued for their tolerance but lacking the genetic refinement seen in commercial crops (Khan, Rafii, Ramlee, Jusoh, & Al-Mamun, 2021). In countries like South Africa, where multiple landraces are mostly

planted, variability in traits such as drought tolerance highlights the urgent need for evaluation to guide breeding programs (Unigwe et al., 2016). Unfortunately, the crop remains one of the most neglected species in agricultural research, limiting progress in its improvement (Muhammad et al., 2020). Bambara groundnut is becoming increasingly important due to its potential to address climate change challenges (Abu & Buah, 2011; Chibarabada et al., 2014). Its high water-use efficiency and adaptability make it a model crop for resource-poor farmers in marginal areas. However, water limitation still significantly affects its vegetative growth and yield, with varying responses among accessions (Muhammad et al., 2016). Understanding this variation is essential for identifying drought tolerant traits and developing strategies to improve crop performance under water-limited conditions.

Existing studies have demonstrated the significance of evaluating Bambara groundnut under drought conditions. Research has focused on physiological traits such as stomatal conductance, photosynthetic rate,

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and morphological traits like leaf length and biomass to understand drought tolerance mechanisms. For instance, Mwale et al. (2007) examined Bambara groundnut's water-use efficiency and physiological responses under varying drought conditions, while Mabhaudhi et al. (2013) highlighted its photosynthetic adaptations to water stress. Additionally, studies by Massawe et al. (2005) investigated morphological traits contributing to drought tolerance, such as root architecture and biomass. Recent investigations including drought indices have further emphasized the importance of traits like stress tolerance index (STI), mean productivity (MP), geometric mean productivity (GMP), as well as gas exchange and chlorophyll fluorescence parameters in assessing drought tolerance among accessions (Kunene et al., 2022; Mandizvo et al., 2022; Rahmah et al., 2020). These indices provide a quantitative framework for evaluating performance under stress and non-stress conditions, offering insights into the variability among accessions (Kunene et al., 2022). While the crop's drought tolerance is well-investigated, systematic evaluations of these indices across diverse accessions remain limited, leaving a knowledge gap essential for breeding programs targeting enhanced drought tolerance.

In this study, we assessed the impact of drought stress on physiological and morphological traits, as well as drought indices, in 24 Bambara groundnut accessions. By characterizing these traits and indices' responses under water stress, we aimed to identify key traits and accessions for drought tolerance.

## 2. Materials and methods

### 2.1. Plant material

Twenty-four Bambara groundnut accessions were obtained from the Agricultural Research Council (ARC) genebank in South Africa. These accessions represent a range of phenotypic diversity, including variation in seed coat colour, which has been suggested to influence growth and performance (Kunene et al., 2022; Mandizvo & Odindo, 2019). All accessions were landraces maintained at the ARC genebank. They are core accessions that are adapted to South African agro-ecologies based on preliminary evaluations (Kunene et al., 2022). The 24 accessions comprised 16 seed coat colours: ochre brown, graphite black, red-brown, sepia brown, brown beige, mahogany brown, golden yellow, red-brown, clay brown, jet black, fawn brown, signal brown, ochre brown, steel blue, terra brown, and brown olive.

### 2.2. Tunnel environment

The experiment was conducted from October 2020 to February 2021 at the Controlled Environment Research Unit (CERU) located on the Pietermaritzburg campus of the University of KwaZulu-Natal, South Africa. The tunnel environment was regulated to maintain a day/night temperature of 27/15°C, a relative humidity of 65 %, and exposure to natural day length. The controlled environment allowed for the precise imposition of drought stress at critical growth stages, thereby enhancing the reliability of accession's comparisons.

### 2.3. Experimental design and trial management

The experiment was set up in a randomized complete block design with a 2 × 24 factorial treatment structure and three replications, resulting in 144 experimental units. Treatments included two irrigation levels (well-watered and water-stressed) and 24 accessions. The well-watered treatment involved regular irrigation to maintain field capacity throughout all growth stages. Under drought-stress treatment, irrigation was withheld entirely for 14 days at three distinct growth stages: vegetative, flowering, and pod-filling. Stress was applied sequentially, meaning that each growth stage experienced a separate 14-day drought period before irrigation was resumed. These specific growth stages were selected based on their known sensitivity to water availability and their

critical influence on plant development and yield formation. Drought stress during the vegetative stage can impair biomass accumulation and canopy development, while stress at the flowering stage is widely recognized to reduce reproductive success and pod formation. Stress at the pod-filling stage can lead to poor seed development and reduced final yield. This approach was guided by prior studies demonstrating the importance of evaluating genotype responses to water stress at different developmental phases. It allowed for the assessment of both early and late-stage drought tolerance mechanisms among accessions (Beebe et al., 2013; Liu et al., 2004; Prasad et al., 2008; Wei et al., 2018).

Each 10-liter pot was spaced 0.4 m apart, with rows separated by 1 m. Three seeds were hand-sown per pot and thinned to one seedling after emergence. Large seeds were selected to ensure uniform germination and vigorous seedling growth (Finch-Savage & Bassel, 2016; Kunene et al., 2022). The experiment was conducted using a clay loam soil, which was well-drained and suitable for legume growth. Prior to planting, the soil was analysed and found to be moderately fertile, with a pH of 6.5.

### 2.4. Data collection

Data collection commenced two weeks after planting and included morphological and physiological traits. Morphological traits included leaf length (LL), leaf width (LW), petiole length (PL), plant height (PH), number of petioles per plant (NPP), total biomass (TB), and seed yield (SY) (Table 2). Physiological measurements included gas exchange and chlorophyll fluorescence parameters, such as stomatal conductance (gs), transpiration rate (T), net CO<sub>2</sub> assimilation rate (A), intercellular CO<sub>2</sub> concentration (Ci), the ratio of assimilation to intercellular CO<sub>2</sub> concentration (A/Ci), the ratio of intercellular to atmospheric CO<sub>2</sub> (Ci/Ca), intrinsic water-use efficiency (A/g<sub>s</sub>), and instantaneous water-use efficiency (F<sub>o</sub>'), maximum fluorescence (F<sub>m</sub>'), maximum quantum efficiency of photosystem II (F<sub>v</sub>/F<sub>m</sub>), effective quantum efficiency of PSII (ΦPSII), photochemical quenching (qP), non-photochemical quenching (qN), electron transport rate (ETR), electron transport relative to assimilation (ETR/A), and alternative electron sink activity (AES). In addition, drought tolerance was evaluated using six indices: stress susceptibility index (SSI), tolerance (TOL), mean productivity index (MPI), stress tolerance index (STI), geometric mean productivity (GMP), and yield stability index (YSI).

#### 2.4.1. Gas exchange and chlorophyll fluorescence parameters

Gas exchange and chlorophyll fluorescence measurements were conducted using the LI-6400XT Portable Photosynthesis System (LI-COR Biosciences, Lincoln, NE, USA), equipped with an integrated infrared gas analyzer (IRGA) and a leaf chamber fluorometer (LCF, Model 6400-40B; 2 cm<sup>2</sup> leaf area). During measurements, the reference CO<sub>2</sub> concentration (Ca) was set to 400 μmol mol<sup>-1</sup>, while photosynthetically active radiation (PAR) was maintained at 1000 μmol m<sup>-2</sup> s<sup>-1</sup> using an artificial light source. The leaf temperature was regulated at 25°C. Additionally, the flow rate was kept constant at 500 μmol s<sup>-1</sup>, and relative humidity within the chamber was adjusted to 43 %, resulting in a vapor pressure deficit (VPD) of approximately 1.7 kPa to prevent stomatal closure due to low humidity.

Measurements were taken between 08:30 and 11:30 a.m. on the third half-expanded leaf from the apex, under both non-stress and drought-stress conditions. For each accession, readings were obtained from three biological replicates. The parameters recorded included stomatal conductance (gs), net CO<sub>2</sub> assimilation rate (A), transpiration rate (T), intercellular CO<sub>2</sub> concentration (Ci), and the ratio of Ci to ambient CO<sub>2</sub> concentration (Ci/Ca). From these, intrinsic water-use efficiency (WUE<sub>i</sub> = A/g<sub>s</sub>) (Mandizvo et al., 2022) and instantaneous water-use efficiency (WUE<sub>ins</sub> = A/T) (Medrano et al., 2015) were calculated. The photosynthetic efficiency (A/Ci) was determined following the approach described by Kitao et al. (2003).

Chlorophyll fluorescence parameters were assessed under steady-state photosynthesis using a saturation pulse intensity of 1300  $\mu\text{mol m}^{-2} \text{s}^{-1}$ . Minimum ( $F_o'$ ) and maximum ( $F_m'$ ) fluorescence yields, as well as steady-state fluorescence ( $F_s$ ), were recorded from light-adapted leaves under ambient glasshouse conditions. Variable fluorescence ( $F_v'$ ) and fluorescence change ( $\Delta F$ ) were derived using the equations:

$$F_v' = F_m' - F_o' \quad (1)$$

$$\Delta F = F_m' - F_s \quad (2)$$

Additional parameters were estimated in accordance with Evans (2009), including the maximum quantum yield of PSII ( $F_v'/F_m'$ ), effective quantum yield of PSII ( $\Phi\text{PSII}$ ), photochemical quenching ( $qP$ ), non-photochemical quenching ( $qN$ ), and electron transport rate (ETR). The ratio of ETR to net assimilation ( $A$ ) provided a relative measure of electron transport to oxygen evolution, while the alternative electron sink (AES) was calculated as the ratio between  $\Phi\text{PSII}$  and the quantum yield of carbon assimilation (Ort & Baker, 2002). All measurements were conducted on fully expanded leaves to ensure consistency across accessions.

#### 2.4.2. Drought indices

To evaluate the drought tolerance of each accession, six drought selection indices were calculated using yield data under non-stress ( $Y_p$ ) and drought-stress ( $Y_s$ ) conditions. The indices (Table 1) included mean productivity (MP), stress tolerance index (STI), geometric mean productivity (GMP), tolerance index (TOL), stress susceptibility index (SSI), and yield stability index (YSI).

#### 2.5. Data analysis

All data were analyzed using GenStat® version 18 (VSN International, UK), with Duncan's multiple range test applied at a 5 % significance level to compare treatment means. Advanced analyses, including principal component analysis (PCA), hierarchical clustering, and correlations analysis, were conducted using R version 4.4.2 (R Foundation for Statistical Computing, Vienna, Austria). Hierarchical clustering was performed using Euclidean distance with the complete linkage method to group accessions into three clusters. The resulting clusters were visualized in a heatmap. The following R packages were used: factoextra, ggplot2, Hmisc, pathview, and ggraph.

**Table 1**

Drought tolerance indices used to evaluate Bambara groundnut accessions for drought tolerance.

| Drought tolerance indices         | Equation   | Reference                                     |
|-----------------------------------|--|---|
| Stress Susceptibility Index (SSI) | $\text{SSI} = \frac{1 - \frac{Y_s}{Y_p}}{1 - \frac{\bar{Y}_s}{\bar{Y}_p}}$ | (Ayed et al., 2021) [Equation 1]              |
| Tolerance (TOL)                   | $\text{TOL} = Y_p - Y_s$   | (Belay et al., 2021) [Equation 2]             |
| Mean Productivity Index (MPI)     | $\text{MPI} = \frac{Y_p + Y_s}{2}$   | (Pour-Aboughadareh et al., 2019) [Equation 3] |
| Stress Tolerance Index (STI)      | $\text{STI} = \frac{Y_p \times Y_s}{(\bar{Y}_p)^2}$                        | (Ekbic et al., 2017) [Equation 4]             |
| Geometric Mean Productivity (GMP) | $\text{GMP} = \sqrt{(\bar{Y}_p)(\bar{Y}_s)}$                               | (Grzesiak et al., 2018) [Equation 5]          |
| Yield Stability Index (YSI)       | $\text{YSI} = \frac{Y_s}{Y_p}$   | (Sánchez-Virosta et al., 2021) [Equation 6]   |

$Y_p$ : yield of each accession under non-stress,  $Y_s$ : yield of each accession under drought stress,  $MP$ : mean productivity,  $STI$ : stress tolerance index,  $GMP$ : geometric mean productivity,  $TOL$ : tolerance index,  $SSI$ : stress susceptibility index, and  $YSI$ : yield stability index.

**Table 2**

List of quantitative agronomic traits recorded from 24 Bambara groundnut accessions.

| Traits                      | Code | Description   | Measurement type |
|-----------------------------|------|---|------------------|
| Leaf length                 | LL   | Length of the leaf from the base to the tip                         | Measuring tape   |
| Leaf width                  | LW   | Width of the leaf from the widest part of the leaf                  | Measuring tape   |
| Petiole length              | PL   | Length of panicle from its base to the tip                          | Measuring tape   |
| Plant height                | PH   | Height of main stalk from the ground to the tip of the main panicle | Measuring tape   |
| Total biomass               | TB   | Weight of the whole plant including roots                           | Weighing balance |
| Number of petiole per plant | NPP  | Number of petioles per plant  | Counting         |
| Seed yield                  | SY   | Total quantity of seeds produced per plant                          | Counting         |

### 3. Results

#### 3.1. Assessment of growth and yield in response to drought stress

Drought stress had a significant impact on the growth and yield performance of Bambara groundnut accessions. Table 3 shows analyses of variance with mean squares and significant tests for growth and yield traits in Bambara groundnut accessions grown under drought stress and non-stressed conditions. A significant variation ( $p < 0.05$ ) was observed between drought-stress and non-stress conditions in terms of leaf length (Fig. 1a). Among the accessions, Acc 199, Acc 151, and Acc 175 exhibited the longest leaf lengths under drought stress, measuring 8.73, 7.70, and 8.70 cm, respectively (Fig. 1d–e). In terms of total biomass and seed yield (Fig. 1b–c), the variation between the two conditions was highly significant ( $p < 0.001$ ). Under drought stress, Acc 190 had the highest biomass (21.33 g), followed by Acc 100 (15.00 g) and Acc 25 (14.67 g). In contrast, under non-stressed conditions, Acc 190 had the highest biomass (46.67 g), followed by Acc 200 (36.67 g). Under drought stress, Acc 97 had the highest yield (9.00 g), followed by Acc 100 (7.33 g).

Drought stress had a significant impact on growth and yield-related traits in Bambara groundnut accessions, as shown by the violin plots (Fig. 2). Under drought-stress conditions, both leaf length (Fig. 2a) and leaf width (Fig. 2b) exhibited reduced median values and broader distributions, indicating greater variability among accessions. In contrast, under non-stress they showed narrower, more centralized distributions, reflecting more uniform trait expression. A similar trend was observed for petiole length (Fig. 2c) and plant height (Fig. 2d), where drought stress resulted in visibly compressed distributions and overall reductions in trait magnitude. The number of petioles per plant (Fig. 2e) also decreased under drought stress, showing a wider and flatter distribution, suggesting inconsistent responses among accessions. Conversely, non-stress conditions resulted in greater uniformity, as shown by the higher density of values near the median. Among all traits, total biomass (Fig. 2f) and seed yield (Fig. 2g) were strongly affected by drought. There was substantial leftward shift in distribution and decreased density, proving the inhibitory effect of water limitation on biomass accumulation.

#### 3.2. Performance of Bambara groundnut accessions under drought stress: insights from drought indices

Table 4 shows variability among accessions with respect to drought tolerance indices. Acc 200, Acc 190, Acc 175, and Acc 87 showed highest tolerance (TOL), indicating superior performance under both drought and well-watered conditions. Notably, Acc 200 recorded the

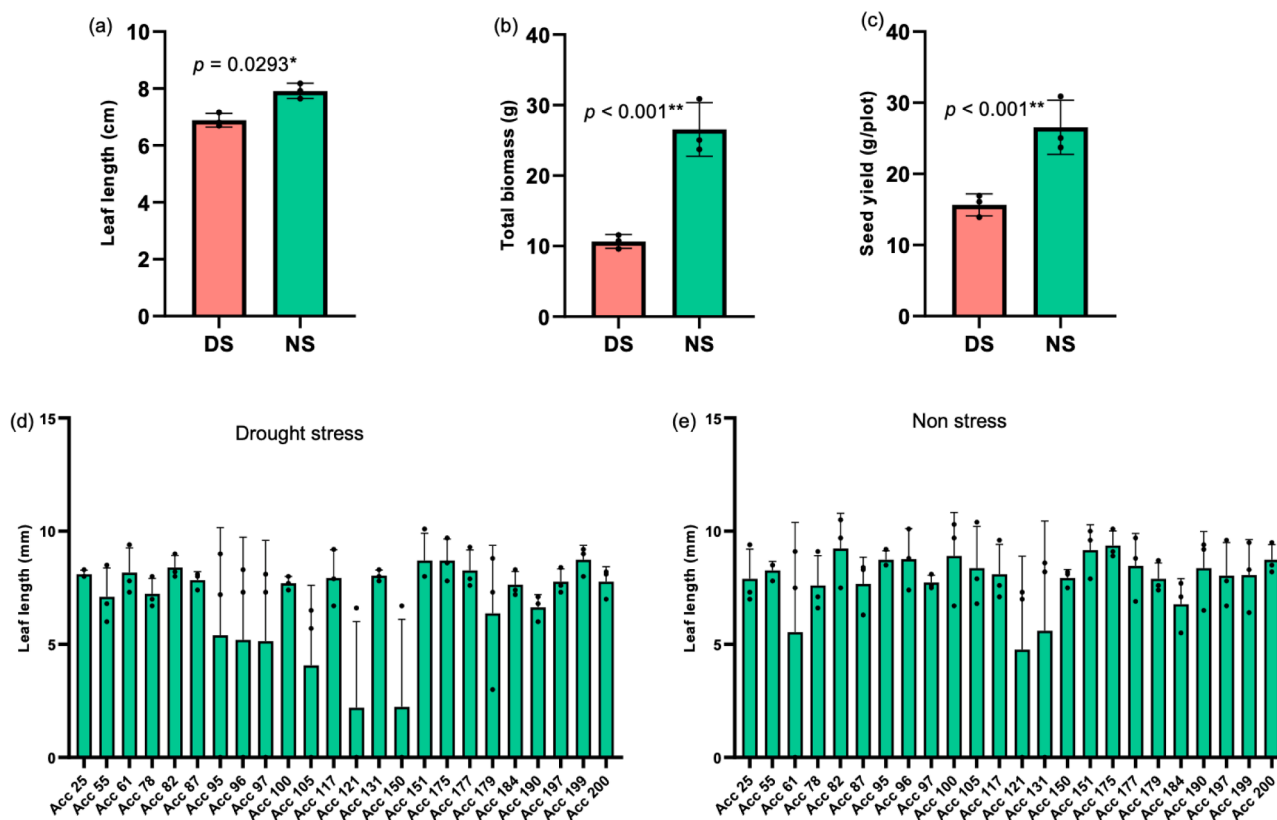


Fig. 1. Effects of drought stress (DS) and non-stress (NS) conditions on morphological traits of Bambara groundnut accessions. (a) leaf length (LL), (b) total biomass (TB), and (c) seed yield (SY), (d) and (e) highlight the effects of drought and non-stress conditions on leaf length of 24 accessions. Asterisks indicate statistical significance based on ANOVA followed by Tukey's test: \* $p < 0.05$ , \*\* $p < 0.01$ , \*\*\* $p < 0.001$ . Error bars represent the standard error of the mean (SE).

Table 3

Combined analyses of variance with mean squares and significant tests for agronomic traits in Bambara groundnut accessions grown under drought stress and non-stressed conditions.

| Source of variation | df | LL                  | LW                  | PH                    | PL                   | SY                 | TB                  | NPP                   |
|---------------------|----|---------------------|---------------------|-----------------------|----------------------|--------------------|---------------------|-----------------------|
| Accessions          | 23 | 9.333**             | 1.601 <sup>ns</sup> | 100.528 <sup>ns</sup> | 39.332 <sup>ns</sup> | 51.4 <sup>ns</sup> | 108.9 <sup>ns</sup> | 100.839 <sup>ns</sup> |
| Stress              | 1  | 38.028**            | 0.681 <sup>ns</sup> | 141.016 <sup>ns</sup> | 105.233*             | 4876.7**           | 9091.6**            | 1.361 <sup>ns</sup>   |
| Accessions x stress | 23 | 5.716 <sup>ns</sup> | 1.169 <sup>ns</sup> | 93.898 <sup>ns</sup>  | 46.751*              | 42.7 <sup>ns</sup> | 149.2 <sup>ns</sup> | 93.941 <sup>ns</sup>  |
| Residual            | 96 | 4.598               | 1.037               | 62.69                 | 26.294               | 42.3               | 100.2               | 82.75                 |

d.f; degrees of freedom, LL; leaf length, LW; leaf width, PH; plant height, PL; petiole length, SY; seed yield, TB; total biomass, NPP; number of petioles per plant. \* and \*\* denote significant at 5 and 1 % probability levels, respectively. ns, non-significant.

highest tolerance index (TOL = 20.00) and a high yield stability index (YSI = 0.47), highlighting its high productivity under optimal conditions. Conversely, accessions such as Acc 177, Acc 95, Acc 97, and Acc 95 demonstrated relatively higher yields under stress (Ys), high STI values, and stable YSI, suggesting better drought resilience. High stress susceptibility index (SSI) in Acc 105 (0.93), Acc 97 (0.75), and Acc 95 (0.58) indicate increased vulnerability to water deficit. In contrast, Acc 197, Acc 190, Acc 175, Acc 131, and Acc 82 showed lower SSI values, reflecting better tolerance under stress. Furthermore, high mean productivity (MP) and geometric mean productivity (GMP) values in Acc 55, Acc 78, Acc 82, Acc 87, Acc 97, Acc 100, Acc 177, Acc 197, Acc 84, and Acc 200 show their consistent yield performance across both environments.

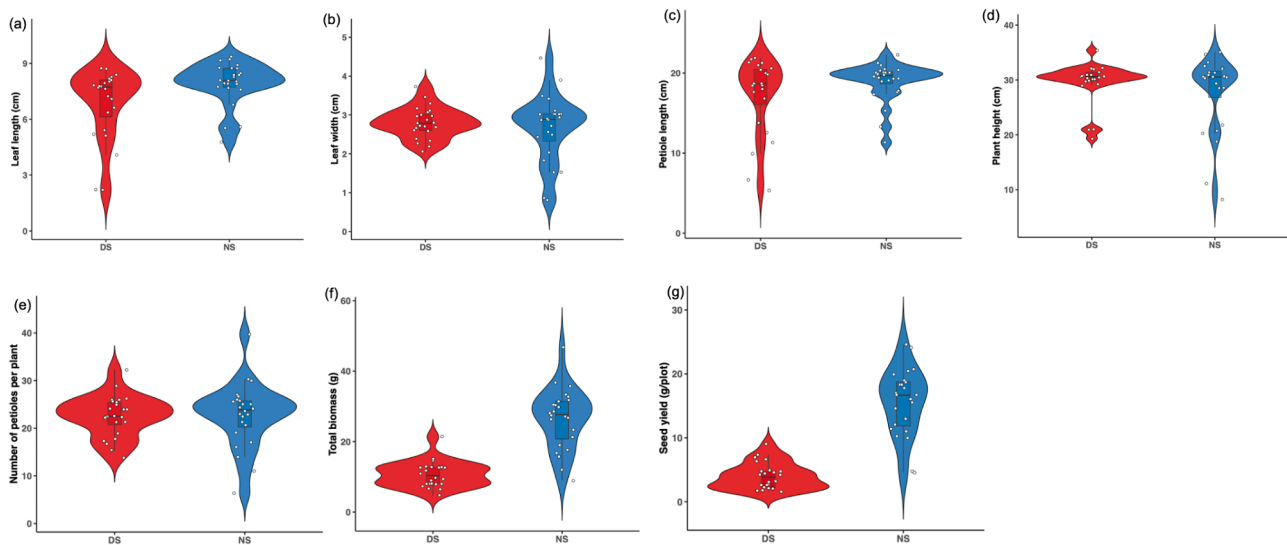
### 3.3. Physiological and photochemical responses of Bambara groundnut accessions to drought stress

Table 5 shows that accessions and drought treatment significantly affected key gas exchange traits, including stomatal conductance ( $g_s$ ), photosynthetic rate (A), transpiration (T), intercellular  $CO_2$

concentration ( $C_i$ ), and water use efficiency (WUEi and WUEinst) ( $p < 0.01$ ). Significant effects were also observed for all chlorophyll fluorescence traits, while variation among accessions was showed in  $F_m'$ , ETR, ETR/A, and AES. Notably, the accession  $\times$  stress interaction was significant for  $g_s$ , A, WUEi, WUEinst, ETR, and AES.

Under non-stress conditions,  $g_s$  ranged from 0.09 to 0.34  $mol\ m^{-2}\ s^{-1}$ , and A from 3.24 to 23.50  $\mu mol\ m^{-2}\ s^{-1}$ , with Acc 87 showing the highest  $g_s$  and Acc 200 the highest A. Drought stress led to a general decline in  $g_s$ , A, and A/ $C_i$ , indicating both stomatal and non-stomatal limitations (Table 6). Acc 25 and Acc 61 maintained relatively higher A and WUEi values under drought stress, suggesting better carbon assimilation and water use efficiency (Table 7).

Chlorophyll fluorescence responses also declined under drought, particularly  $F_v/F_m$  and  $\Phi PSII$ , reflecting reduced PSII efficiency (Table 7). However, Acc 25, Acc 61, and Acc 87 exhibited moderate reductions, indicating better photoprotection. ETR and ETR/A values declined under drought in most accessions but remained comparatively higher in Acc 25 and Acc 61, suggesting more efficient electron transport relative to carbon fixation. AES followed similar patterns, with drought-sensitive accessions showing the lowest values.



**Fig. 2.** Violin plots illustrating the effect of drought stress (DS) and non-stress (NS) conditions on key morphological and yield-related traits in Bambara groundnut accessions. (a) Leaf length, (b) leaf width, (c) petiole length, (d) plant height, (e) number of petioles per plant, and (f) total biomass (g). Each plot displays the full distribution of trait values across accessions, with the width of the violin representing the kernel density of the data. Box plots within each violin indicate the median and interquartile range.

**Table 4**

Drought indices of 24 Bambara groundnut accessions evaluated under non-stress and drought stress conditions.

| Accession | Yp(g/plot) | Ys(g/plot) | STI   | SSI   | TOL   | MP    | GMP   | YSI   |
|-----------|------------|------------|-------|-------|-------|-------|-------|-------|
| Acc 25    | 10.33      | 4.33       | 0.18  | 0.42  | 6.00  | 7.33  | 6.69  | 0.18  |
| Acc 55    | 20.33      | 4.67       | 0.39  | 0.23  | 15.66 | 12.50 | 9.74  | 0.39  |
| Acc 61    | 13.00      | 2.00       | 0.11  | 0.15  | 11.00 | 7.50  | 5.10  | 0.11  |
| Acc 78    | 16.00      | 4.67       | 0.31  | 0.29  | 11.33 | 10.33 | 8.64  | 0.31  |
| Acc 82    | 17.67      | 2.33       | 0.17  | 0.13  | 15.34 | 10.00 | 6.42  | 0.17  |
| Acc 87    | 24.00      | 5.00       | 0.49  | 0.21  | 19.00 | 14.50 | 10.95 | 0.49  |
| Acc 95    | 11.00      | 6.33       | 0.28  | 0.58  | 4.67  | 8.67  | 8.35  | 0.28  |
| Acc 96    | 11.33      | 1.67       | 0.08  | 0.15  | 9.66  | 6.50  | 4.35  | 0.08  |
| Acc 97    | 12.00      | 9.00       | 0.44  | 0.75  | 3.00  | 10.50 | 10.39 | 0.44  |
| Acc 100   | 18.33      | 7.33       | 0.55  | 0.40  | 11.00 | 12.83 | 11.59 | 0.55  |
| Acc 105   | 4.67       | 4.33       | 0.08  | 0.93  | 0.34  | 4.50  | 4.50  | 0.08  |
| Acc 117   | 18.67      | 6.67       | 0.51  | 0.36  | 12.00 | 12.67 | 11.16 | 0.51  |
| Acc 121   | 4.67       | 2.00       | 0.04  | 0.43  | 2.67  | 3.34  | 3.06  | 0.04  |
| Acc 131   | 15.67      | 1.67       | 0.11  | 0.11  | 14.00 | 8.67  | 5.11  | 0.11  |
| Acc 150   | 10.00      | 2.67       | 0.11  | 0.27  | 7.33  | 6.33  | 5.16  | 0.11  |
| Acc 151   | 14.67      | 2.00       | 0.12  | 0.14  | 12.67 | 8.34  | 5.42  | 0.12  |
| Acc 175   | 20.00      | 2.67       | 0.22  | 0.13  | 17.33 | 11.33 | 7.30  | 0.22  |
| Acc 177   | 18.33      | 7.00       | 0.52  | 0.38  | 11.33 | 12.67 | 11.33 | 0.52  |
| Acc 179   | 17.00      | 3.00       | 0.21  | 0.18  | 14.00 | 10.00 | 7.14  | 0.21  |
| Acc 184   | 16.67      | 4.33       | 0.30  | 0.26  | 12.34 | 10.50 | 8.50  | 0.30  |
| Acc 190   | 20.67      | 2.67       | 0.23  | 0.13  | 18.00 | 11.67 | 7.42  | 0.23  |
| Acc 197   | 16.67      | 1.67       | 0.11  | 0.10  | 15.00 | 9.17  | 5.27  | 0.11  |
| Acc 199   | 19.00      | 3.33       | 0.26  | 0.18  | 15.67 | 11.17 | 7.96  | 0.26  |
| Acc 200   | 24.67      | 4.67       | 0.47  | 0.19  | 20.00 | 14.67 | 10.73 | 0.47  |
| LSD       | 6.27       | 57.05      | 6.97  | 13.95 | 58.2  | 28.28 | 12.42 | 13.92 |
| CV %      | 33.27      | 51.66      | 29.81 | 45.81 | 33.68 | 62.35 | 29.81 | 71.30 |
| p-value   | 0.55       | 0.73       | 0.83  | 0.13  | 0.57  | 0.83  | 0.91  | 0.13  |

**Yp**; yield under non-stress conditions, **Ys**; yield under drought stress conditions, **STI**; stress tolerance index, **SSI**; stress susceptibility index, **TOL**; tolerance, **MP**; mean productivity, **GMP**; geometric mean productivity, **YSI**; yield stability index, **LSD**; least significant difference, **CV %**; coefficient of variation, **p-value**; probability value.

Under drought stress (Fig. 3), Bambara groundnut accessions showed notable variation in gas exchange and chlorophyll fluorescence responses compared to the non-stress condition. Most accessions showed a significant decrease in stomatal conductance ( $g_s$ ) and net  $CO_2$  assimilation rate ( $A$ ) under drought. Interestingly, transpiration rate ( $T$ ) remained relatively unchanged (Fig. 3b). In some accessions, intercellular  $CO_2$  concentration ( $C_i$ ) increased slightly under drought stress (Fig. 3d). Both the  $A/C_i$  ratio and the  $C_i/C_a$  ratio declined under stress conditions (Fig. 3e–f). Additionally, intrinsic water-use efficiency ( $A/g_s$ ) and instantaneous water-use efficiency ( $A/T$ ) decreased in many accessions (Fig. 3e, h). Chlorophyll fluorescence parameters were also

adversely affected. Minimum fluorescence ( $F_o'$ ) and maximum fluorescence ( $F_m'$ ) decreased under drought stress (Fig. 3i–j), along with a reduction in the maximum quantum efficiency of PSII photochemistry ( $F_v/F_m$ ) (Fig. 3j). A significant decline was also observed in the effective quantum efficiency of PSII ( $\Phi_{PSII}$ ) (Fig. 3i). Both photochemical quenching ( $q_P$ ) and non-photochemical quenching ( $q_N$ ) decreased under drought conditions compared to the non-stressed condition (Fig. 3m–n). Furthermore, the electron transport rate ( $ETR$ ), the  $ETR/A$  ratio, and alternative electron sink ( $AES$ ) values all declined under drought stress (Fig. 3o–q).

**Table 5**

Analysis of variance showing mean squares and significant tests for leaf gas exchange and chlorophyll fluorescence parameters of 24 Bambara groundnut accessions evaluated under drought-stress and non-stress conditions.

| Gas exchange parameters             |    |                     |                    |                      |                      |                      |                      |          |                     |          |
|-------------------------------------|----|---------------------|--------------------|----------------------|----------------------|----------------------|----------------------|----------|---------------------|----------|
| Source of variation                 | df | gs                  | T                  | A                    | Ci                   | A/Ci                 | Ci/Ca                | WUEi     | WUEinst             |          |
| Accessions                          | 23 | 0.01526**           | 753.32**           | 108.46**             | 4086**               | 0.0654 <sup>ns</sup> | 0.086 <sup>ns</sup>  | 2.1339** | 8247.9**            |          |
| Stress                              | 1  | 0.49294**           | 6.72**             | 824.67**             | 73748**              | 7.1515**             | 7.9532**             | 16.233** | 291.9**             |          |
| Accessions x stress                 | 23 | 0.00361**           | 0.06 <sup>ns</sup> | 14.91**              | 94 <sup>ns</sup>     | 0.0628 <sup>ns</sup> | 0.0622 <sup>ns</sup> | 0.704**  | 89.4**              |          |
| Residual                            | 96 | 0.00053             | 0.14               | 0.14                 | 438                  | 0.1412               | 0.1412               | 0.1412   | 10.8                |          |
| Chlorophyll fluorescence parameters |    |                     |                    |                      |                      |                      |                      |          |                     |          |
| Source of variation                 | df | Fo'                 | Fm'                | Fv/Fm                | ΦPSII                | qP                   | qN                   | ETR      | ETR/A               | AES      |
| Accessions                          | 23 | 562.9 <sup>ns</sup> | 3371**             | 0.0627 <sup>ns</sup> | 0.0771 <sup>ns</sup> | 0.0731 <sup>ns</sup> | 0.0829 <sup>ns</sup> | 3442**   | 931.66**            | 2.1339** |
| Stress                              | 1  | 21628.6**           | 104211**           | 7.2335**             | 11.2025**            | 10.9364**            | 3.5232**             | 422262** | 1625.57**           | 16.233** |
| Accessions x stress                 | 23 | 120.6 <sup>ns</sup> | 125 <sup>ns</sup>  | 0.0618 <sup>ns</sup> | 0.0624 <sup>ns</sup> | 0.0621 <sup>ns</sup> | 0.0653 <sup>ns</sup> | 738*     | 66.58 <sup>ns</sup> | 0.704**  |
| Residual                            | 96 | 438.1               | 438                | 0.1412               | 0.1412               | 0.1412               | 0.1412               | 438      | 46.18               | 0.1412   |

d.f; degrees of freedom, gs; stomatal conductance, T; transpiration rate, A; net CO<sub>2</sub> assimilation rate, A/Ci; CO<sub>2</sub> assimilation rate/intercellular CO<sub>2</sub> concentration, Ci; intercellular CO<sub>2</sub> concentration, Ci/Ca, ratio of intercellular and atmospheric CO<sub>2</sub>, WUEi; intrinsic water use efficiency, WUEinst; instantaneous water-use efficiency, Fv/Fm; maximum quantum efficiency of photosystem II photochemistry, ΦPSII; the effective quantum efficiency of PSII photochemistry, qP; photochemical quenching, qN; non-photochemical quenching, ETR; electron transport rate, ETR/A; relative measure of electron transport to oxygen molecules, AES; alternative electron sinks. \*and \*\* denote significant at 5 and 1 % probability levels, respectively. ns, non-significant.

### 3.4. Phenotypic diversity and trait contributions under drought and non-stress conditions

Table 8 shows the principal component analysis (PCA) results, including factor loadings, eigenvalues, and percentage variance for the evaluated traits under drought stress and non-stress conditions. Under drought stress, the first five principal components (PCs) accounted for 87.43 % of the total variance, with PC1 contributing 34.36 %, followed by PC2 (22.62 %), PC3 (17.44 %), PC4 (7.78 %), and PC5 (5.23 %). In non-stress conditions, the first five PCs explained 88.22 % of the total variance, with PC1 accounting for 29.97 %, followed by PC2 (21.10 %), PC3 (18.19 %), PC4 (12.04 %), and PC5 (6.91 %). Under drought stress, PC1 was predominantly influenced by high positive loadings from qP (0.303), Fm' (0.302), Fv/Fm (0.286), Ci (0.284), gs (0.284), and ETR (0.272), indicating the key role of photosynthetic performance and gas exchange in drought adaptation. PC2 reflected variation in water-use efficiency and fluorescence traits, with moderate negative loadings from T (−0.285), A (−0.300), and MP (−0.325). PC3 highlighted yield and biomass attributes under drought, with high loadings from TB (0.381) and SY (0.400). PC4 was defined by morphological traits, especially PL (0.292) and LL (0.283), while PC5 showed moderate loadings from PH (−0.451) and NPP (−0.511), reflecting the impact of drought on plant height and reproductive development. Under non-stress conditions, PC1 showed strong negative contributions from PH (−0.245), TB (−0.251), and SY (−0.303), suggesting that biomass and yield performance were central axes of variation. PC2 was mainly influenced by negative loadings from qP (−0.340), ΦPSII (−0.342), Fm' (−0.276), and Ci/Ca (−0.334), reflecting differences in photosynthetic efficiency. PC3 was characterized by moderate positive contributions from ETR (0.258), Fm' (0.241), and Ci (0.248), associated with electron transport and fluorescence under optimal conditions. PC4 emphasized morphological plasticity, with PL (0.262) and LL (0.215) contributing positively. Finally, PC5 captured variation in plant height and yield traits, with PH (0.310) and NPP (0.264) showing high positive loadings.

The principal component (PC) biplots revealed the phenotypic relationships among Bambara groundnut accessions under both conditions. Traits positioned closely together or aligned in the same direction indicated strong positive correlations, while vectors pointing in opposite directions reflected negative correlations. Under drought stress, the PCA of agronomic traits and drought indices (Fig. 4a–b) showed that PC1 and PC2 accounted for 43.57 % and 29.19 % of the total variation, respectively. Under non-stress conditions, PC1 and PC2 accounted 54.10 % and 22.93 % of the total variation, respectively. Accessions such as Acc 200, Acc 199, Acc 184, and Acc 177 were distinctly separated along PC1 under drought stress, reflecting their unique drought response profiles

and high performance in drought tolerance indices such as STI, GMP, and MP. Interestingly, accessions were more dispersed under non-stress conditions, indicating underlying phenotypic variability even in optimal environments. Although traits such as TOL, STI, MP, and GMP continued to exert strong influence, the directional grouping was less defined compared to drought stress conditions. Specifically, STI, MP, GMP, and SY contributed positively and strongly to PC1, emphasizing their close association with drought tolerance. Under non-stress conditions, MP, GMP, and STI were most closely associated with Acc 177, Acc 100, Acc 200, and Acc 87, while TB and TOL were more strongly linked to Acc 82, Acc 190, and Acc 179. For leaf gas exchange and chlorophyll fluorescence traits under drought stress (Fig. 4c), PC1 and PC2 explained 59.79 % and 26.86 % of the total variation, respectively. Traits such as intrinsic water-use efficiency (WUEinst), non-photochemical quenching (qN), minimum fluorescence (Fo'), electron transport rate per assimilation (ETR/A), and alternative electron sink (AES) were closely aligned and contributed positively to PC1, highlighting their importance in physiological adaptation to water deficit. Accessions such as Acc 61, Acc 150, and Acc 175 clustered around these vectors, indicating superior physiological performance under drought. Under non-stress conditions (Fig. 4d), PC1 and PC2 accounted for 37.98 % and 34.16 % of the variation, respectively. Accessions like Acc 25 and Acc 97 were widely separated, indicating diversity in photosynthetic efficiency and photo-protective traits. The clustering pattern was less compact, reflecting a broader range of physiological responses in the absence of drought.

### 3.5. Impact of drought stress on phenotypic stability and stress tolerance indices

Hierarchical clustering analysis revealed three distinct groups of Bambara groundnut accessions under both conditions, based on agronomic traits, drought tolerance indices, and physiological parameters (Fig. 5a–d). Under drought stress (Fig. 5a), accessions in cluster 2 including Acc 151, Acc 82, Acc 179, Acc 175, Acc 131, and Acc 61 showed higher values for key agronomic traits such as tolerance index (TOL), leaf length (LL), and petiole length (PL). Cluster 1 comprised accessions such as Acc 25, Acc 55, Acc 95, Acc 200, Acc 100, Acc 117, Acc 184, and Acc 97, which exhibited strong performance across yield-related indices including yield stability index (YSI), stress tolerance index (STI), geometric mean productivity (GMP), mean productivity (MP), seed yield (SY), and leaf width (LW). In contrast, cluster 3 (Acc 121, Acc 105, Acc 96, and Acc 150) was characterized by higher values for stress susceptibility index (SSI), number of petioles per plant (NPP), and plant height (PH). Under non-stress conditions (Fig. 5b), cluster 1 included accessions such as Acc 121, Acc 105, Acc 150, Acc 25, and Acc

**Table 6**  
Means of leaf gas exchange and chlorophyll fluorescence parameters of Bambara groundnut accessions under non-stress condition.

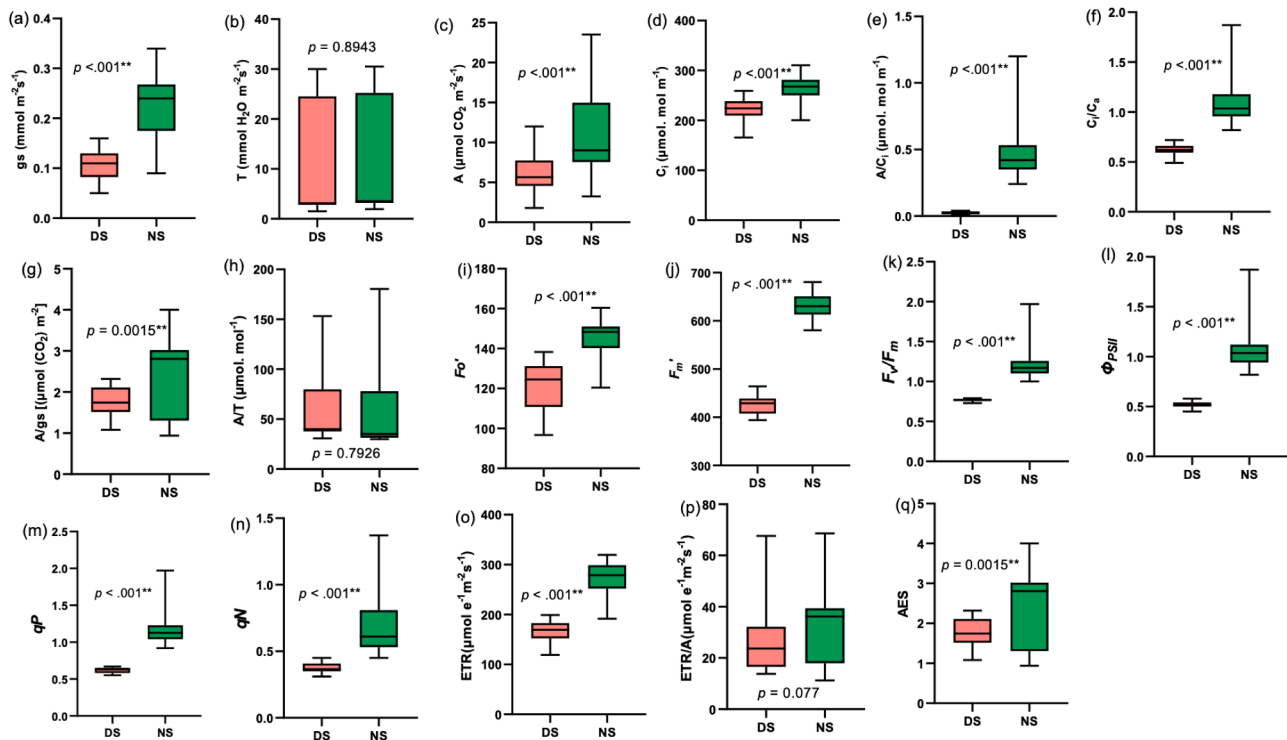
| Accessions | Leaf gas exchange parameters |                    |                    |                     |                    |                    |                     |                     |                     | Chlorophyll fluorescence parameters |                    |                    |                    |                    |                     |                    |                    |
|------------|------------------------------|--------------------|--------------------|---------------------|--------------------|--------------------|---------------------|---------------------|---------------------|-------------------------------------|--------------------|--------------------|--------------------|--------------------|---------------------|--------------------|--------------------|
|            | gs                           | T                  | A                  | Ci                  | A/Ci               | Ci/Ca              | WUEi                | WUEinst             | Fo'                 | Fm'                                 | Fv/Fm              | ΦPSII              | qP                 | qN                 | ETR                 | ETR/A              | AES                |
| Acc 25     | 0.24                         | 4.17               | 9.67               | 281.17              | 1.20 <sup>a</sup>  | 1.87 <sup>a</sup>  | 4.00 <sup>a</sup>   | 35.17               | 151.17              | 651.17                              | 1.97 <sup>a</sup>  | 1.87 <sup>a</sup>  | 1.97 <sup>a</sup>  | 1.37 <sup>a</sup>  | 299.17 <sup>a</sup> | 36.23              | 4.00 <sup>a</sup>  |
| Acc 55     | 0.22                         | 3.08               | 6.48               | 260.28              | 0.31               | 0.93               | 2.50                | 31.28               | 140.28              | 620.28                              | 1.06               | 0.93               | 1.03               | 0.53 <sup>ab</sup> | 276.28              | 44.80              | 2.50               |
| Acc 61     | 0.25                         | 3.49               | 8.59               | 270.59              | 0.62 <sup>ab</sup> | 1.26 <sup>ab</sup> | 3.35 <sup>ab</sup>  | 35.37               | 145.59              | 630.59                              | 1.38 <sup>ab</sup> | 1.27 <sup>ab</sup> | 1.36 <sup>ab</sup> | 0.82 <sup>ab</sup> | 289.59              | 36.72              | 3.35 <sup>ab</sup> |
| Acc 78     | 0.19                         | 3.04               | 6.94               | 250.44              | 0.46 <sup>ab</sup> | 1.06 <sup>ab</sup> | 2.94                | 36.55               | 135.44              | 610.44                              | 1.22 <sup>ab</sup> | 1.07 <sup>ab</sup> | 1.16 <sup>ab</sup> | 0.72 <sup>ab</sup> | 268.44              | 41.67              | 2.94               |
| Acc 82     | 0.26                         | 3.55               | 9.35               | 290.35              | 0.38 <sup>ab</sup> | 1.08 <sup>ab</sup> | 3.16 <sup>abc</sup> | 34.97               | 155.35              | 660.35                              | 1.16 <sup>ab</sup> | 1.07 <sup>ab</sup> | 1.16 <sup>ab</sup> | 0.54 <sup>ab</sup> | 306.35              | 34.35              | 3.16 <sup>ab</sup> |
| Acc 87     | 0.34 <sup>a</sup>            | 4.39               | 10.39              | 310.39 <sup>a</sup> | 0.42 <sup>ab</sup> | 1.17 <sup>ab</sup> | 2.89                | 31.64               | 160.39 <sup>a</sup> | 680.39 <sup>a</sup>                 | 1.21 <sup>ab</sup> | 1.14 <sup>ab</sup> | 1.24 <sup>ab</sup> | 0.54 <sup>ab</sup> | 319.39              | 32.29              | 2.89               |
| Acc 95     | 0.10                         | 1.94               | 3.24               | 220.44              | 0.46 <sup>ab</sup> | 0.99               | 2.31                | 35.44               | 120.44              | 580.44                              | 1.20 <sup>ab</sup> | 0.89               | 1.04               | 0.84 <sup>ab</sup> | 191.44              | 68.65 <sup>a</sup> | 2.31               |
| Acc 96     | 0.09                         | 3.32               | 8.02               | 275.52              | 0.55 <sup>ab</sup> | 1.21 <sup>ab</sup> | 3.20 <sup>ab</sup>  | 34.61               | 148.52              | 640.52                              | 1.31 <sup>ab</sup> | 1.20 <sup>ab</sup> | 1.30 <sup>ab</sup> | 0.74 <sup>ab</sup> | 289.52              | 39.05              | 3.20 <sup>ab</sup> |
| Acc 97     | 0.10                         | 2.22               | 5.22               | 240.22              | 0.24               | 0.82               | 2.72                | 33.55               | 130.22              | 600.22                              | 1.00               | 0.82               | 0.92               | 0.52 <sup>ab</sup> | 255.22              | 51.22              | 2.72               |
| Acc 100    | 0.32 <sup>ab</sup>           | 3.77               | 9.77               | 300.27              | 0.30               | 1.02 <sup>ab</sup> | 2.98                | 31.93               | 158.27              | 670.27                              | 1.08               | 1.00 <sup>ab</sup> | 1.09               | 0.45               | 311.27              | 33.01              | 2.98               |
| Acc 105    | 0.29                         | 3.48               | 8.68               | 265.38              | 0.41 <sup>ab</sup> | 1.04 <sup>ab</sup> | 3.05                | 30.02               | 152.38              | 650.38                              | 1.18 <sup>ab</sup> | 1.09 <sup>ab</sup> | 1.18 <sup>ab</sup> | 0.58 <sup>ab</sup> | 302.38              | 36.77              | 3.05               |
| Acc 117    | 0.27                         | 3.31               | 8.31               | 280.31              | 0.34 <sup>ab</sup> | 1.01 <sup>ab</sup> | 2.97                | 31.08               | 150.31              | 640.31                              | 1.11 <sup>ab</sup> | 1.01 <sup>ab</sup> | 1.10 <sup>ab</sup> | 0.52 <sup>ab</sup> | 298.31              | 37.56              | 2.97               |
| Acc 121    | 0.25                         | 3.07               | 7.67               | 260.27              | 0.30               | 0.92               | 2.91                | 29.87               | 148.27              | 630.27                              | 1.06               | 0.95               | 1.05               | 0.49               | 289.27              | 39.32              | 2.91               |
| Acc 131    | 0.26                         | 3.49               | 8.59               | 275.39              | 0.42 <sup>ab</sup> | 1.07 <sup>ab</sup> | 3.03                | 37.66               | 150.39              | 640.39                              | 1.18 <sup>ab</sup> | 1.08 <sup>ab</sup> | 1.16 <sup>ab</sup> | 0.62 <sup>ab</sup> | 293.39              | 36.12              | 3.03               |
| Acc 150    | 0.23                         | 3.32               | 7.52               | 250.32              | 0.35 <sup>ab</sup> | 0.95               | 2.72                | 34.61               | 145.32              | 630.32                              | 1.10 <sup>ab</sup> | 0.98               | 1.08               | 0.56 <sup>ab</sup> | 281.32              | 39.35              | 2.72               |
| Acc 151    | 0.30 <sup>abc</sup>          | 3.92               | 9.62               | 285.32              | 0.35 <sup>ab</sup> | 1.03 <sup>ab</sup> | 2.90                | 31.32               | 155.32              | 660.32                              | 1.13 <sup>ab</sup> | 1.06 <sup>ab</sup> | 1.15 <sup>ab</sup> | 0.49               | 314.32              | 34.08              | 2.90               |
| Acc 175    | 0.24                         | 3.12               | 7.02               | 265.32              | 0.35 <sup>ab</sup> | 0.98               | 2.71                | 30.78               | 140.32              | 620.32                              | 1.10 <sup>ab</sup> | 0.94               | 1.06               | 0.58 <sup>ab</sup> | 264.32              | 39.72              | 2.71               |
| Acc 177    | 0.17                         | 25.82              | 15.82              | 250.82              | 0.88 <sup>ab</sup> | 1.44 <sup>ab</sup> | 1.42                | 100.82              | 140.82              | 610.82                              | 1.60 <sup>ab</sup> | 1.40 <sup>ab</sup> | 1.47 <sup>ab</sup> | 1.17 <sup>ab</sup> | 247.82              | 17.29              | 1.42               |
| Acc 179    | 0.14                         | 23.42              | 12.42              | 200.42              | 0.48 <sup>ab</sup> | 0.92               | 0.94                | 100.42              | 135.42              | 600.42                              | 1.18 <sup>ab</sup> | 0.97               | 1.04               | 0.80 <sup>ab</sup> | 234.42              | 19.92              | 0.94               |
| Acc 184    | 0.25                         | 27.53              | 20.53              | 260.53              | 0.61 <sup>ab</sup> | 1.18 <sup>ab</sup> | 1.27                | 80.53               | 145.53              | 630.53                              | 1.31 <sup>ab</sup> | 1.13 <sup>ab</sup> | 1.25 <sup>ab</sup> | 0.81 <sup>ab</sup> | 255.53              | 13.28              | 1.27               |
| Acc 190    | 0.30 <sup>abc</sup>          | 28.33              | 21.33              | 290.33              | 0.41 <sup>ab</sup> | 1.06 <sup>ab</sup> | 1.08                | 70.33               | 150.33              | 650.33                              | 1.12 <sup>ab</sup> | 0.95               | 1.06               | 0.60 <sup>ab</sup> | 264.33              | 12.90              | 1.08               |
| Acc 197    | 0.11                         | 26.37              | 18.37              | 220.37              | 0.46 <sup>ab</sup> | 0.92               | 1.07                | 180.37 <sup>a</sup> | 140.37              | 600.37                              | 1.14 <sup>ab</sup> | 0.93               | 1.00               | 0.74 <sup>ab</sup> | 238.37              | 13.59              | 1.07               |
| Acc 199    | 0.24                         | 29.34              | 22.34              | 270.34              | 0.42 <sup>ab</sup> | 1.02 <sup>ab</sup> | 1.10                | 100.34              | 150.34              | 640.34                              | 1.12 <sup>ab</sup> | 0.93               | 1.04               | 0.64 <sup>ab</sup> | 251.34              | 11.75              | 1.10               |
| Acc 200    | 0.19                         | 30.50 <sup>a</sup> | 23.50 <sup>a</sup> | 280.50              | 0.58 <sup>ab</sup> | 1.20 <sup>ab</sup> | 1.26                | 128.27              | 148.50              | 630.50                              | 1.27 <sup>ab</sup> | 1.08 <sup>ab</sup> | 1.18 <sup>ab</sup> | 0.82 <sup>ab</sup> | 247.50              | 11.24              | 1.26               |
| LSD        | 0.05                         | 0.872              | 0.872              | 0.872               | 0.872              | 0.872              | 0.872               | 0.872               | 0.872               | 0.872                               | 0.872              | 0.872              | 0.872              | 0.872              | 0.872               | 0.872              | 0.872              |
| CV %       | 34.09                        | 107.13             | 51.03              | 9.66                | 102.81             | 44.55              | 40.35               | 70.84               | 6.22                | 3.79                                | 39.55              | 45.16              | 41.32              | 71.02              | 42.68               | 42.68              | 40.35              |
| p value    | <.001                        | <.001              | <.001              | <.001               | 0.979              | 0.969              | <.001               | <.001               | <.001               | <.001                               | 0.983              | 0.972              | 0.975              | 0.959              | <.001               | <.001              | <.001              |

Different upper-case letters within a column indicate significant difference among accessions. gs; (mmol m<sup>-2</sup> s<sup>-1</sup>), T; transpiration rate (mmol H<sub>2</sub>O m<sup>-2</sup> s<sup>-1</sup>), A; (μmol CO<sub>2</sub> m<sup>-2</sup> s<sup>-1</sup>), A/Ci; (μmol. mol<sup>-1</sup>), Ci; (μmol. mol<sup>-1</sup>), WUEi; [(μmol (CO<sub>2</sub>) m<sup>-2</sup>); WUEinst, (μmol. mol<sup>-1</sup>), Fv/Fm; (ratio); ΦPSII, the effective quantum efficiency of PSII photochemistry; qP, photochemical quenching; qN, non-photochemical quenching; ETR, (μmol e<sup>-1</sup> m<sup>-2</sup> s<sup>-1</sup>); ETR/A, (μmol e μmol<sup>-1</sup> CO<sub>2</sub>); AES, alternative electron sinks.

**Table 7**  
Means of leaf gas exchange and chlorophyll fluorescence parameters of Bambara groundnut accessions under drought-stress condition.

| Accessions | Leaf gas exchange parameters |                 |                 |                      |                       |                   |                   |                     | Chlorophyll fluorescence parameters |                       |                      |                     |                     |                    |                       |                    |                   |
|------------|------------------------------|-----------------|-----------------|----------------------|-----------------------|-------------------|-------------------|---------------------|-------------------------------------|-----------------------|----------------------|---------------------|---------------------|--------------------|-----------------------|--------------------|-------------------|
|            | gs                           | T               | A               | Ci                   | A/Ci                  | Ci/Ca             | WUEi              | WUEinst             | Fo'                                 | Fm'                   | Fv/Fm                | ΦPSII               | qP                  | qN                 | ETR                   | ETR/A              | AES               |
| Acc 25     | 0.13                         | 3.00            | 5.50            | 243.33 <sup>ab</sup> | 0.017                 | 0.65              | 2.11              | 38.31               | 138.33 <sup>a</sup>                 | 448.33 <sup>abc</sup> | 0.776                | 0.539               | 0.65                | 0.346              | 193.33 <sup>ab</sup>  | 33.18              | 2.11              |
| Acc 55     | 0.09                         | 2.79            | 3.79            | 226.67               | 0.012                 | 0.61              | 1.58              | 32.99               | 131.67 <sup>a</sup>                 | 431.67                | 0.763                | 0.510               | 0.62                | 0.363              | 166.67                | 35.35              | 1.58              |
| Acc 61     | 0.10                         | 2.89            | 5.19            | 221.67               | 0.015                 | 0.61              | 1.78              | 43.26               | 124.67 <sup>a</sup>                 | 429.33                | 0.768                | 0.518               | 0.63                | 0.348              | 169.67 <sup>abc</sup> | 26.75              | 1.78              |
| Acc 78     | 0.06                         | 2.57            | 4.17            | 184.67               | 0.019                 | 0.55              | 1.59              | 44.64               | 99.67 <sup>a</sup>                  | 396.33                | 0.735                | 0.495               | 0.57                | 0.375              | 128.67                | 24.76              | 1.59              |
| Acc 82     | 0.12                         | 3.18            | 6.18            | 235.33 <sup>ab</sup> | 0.011                 | 0.66              | 2.29              | 39.61               | 125.33 <sup>a</sup>                 | 438.67                | 0.773                | 0.550               | 0.65                | 0.313              | 185.33 <sup>ab</sup>  | 23.48              | 2.29              |
| Acc 87     | 0.15 <sup>ab</sup>           | 3.99            | 6.99            | 259 <sup>a</sup>     | 0.015                 | 0.72 <sup>a</sup> | 2.18              | 38.74               | 134.00 <sup>a</sup>                 | 464 <sup>a</sup>      | 0.791 <sup>a</sup>   | 0.570 <sup>a</sup>  | 0.671 <sup>a</sup>  | 0.311              | 199 <sup>a</sup>      | 23.86              | 2.18              |
| Acc 95     | 0.05                         | 1.50            | 1.80            | 169.00               | 0.011                 | 0.50              | 1.20              | 30.67               | 104.00 <sup>a</sup>                 | 394.00                | 0.742                | 0.445               | 0.55                | 0.452 <sup>a</sup> | 119.00                | 67.66 <sup>a</sup> | 1.20              |
| Acc 96     | 0.11                         | 2.80            | 4.80            | 226.33               | 0.020                 | 0.63              | 1.71              | 35.97               | 131.33 <sup>a</sup>                 | 428.00                | 0.770                | 0.511               | 0.62                | 0.380              | 171.33 <sup>abc</sup> | 21.96              | 1.71              |
| Acc 97     | 0.08                         | 2.00            | 3.00            | 188.33               | 0.011                 | 0.55              | 1.36              | 32.16               | 108.33 <sup>a</sup>                 | 401.67                | 0.747                | 0.503               | 0.58                | 0.417              | 128.33                | 41.00              | 1.36              |
| Acc 100    | 0.15 <sup>ab</sup>           | 3.50            | 6.50            | 252.67 <sup>ab</sup> | 0.020                 | 0.70              | 2.32 <sup>a</sup> | 41.66               | 132.67 <sup>a</sup>                 | 452.67 <sup>ab</sup>  | 0.787 <sup>abc</sup> | 0.554               | 0.66                | 0.337              | 187.67 <sup>ab</sup>  | 20.08              | 2.32 <sup>a</sup> |
| Acc 105    | 0.14                         | 3.10            | 5.80            | 224.00               | 0.022                 | 0.62              | 2.32 <sup>a</sup> | 38.43               | 129.00 <sup>a</sup>                 | 439.00                | 0.778                | 0.548               | 0.65                | 0.348              | 174 <sup>abc</sup>    | 25.81              | 2.32 <sup>a</sup> |
| Acc 117    | 0.13                         | 3.00            | 5.50            | 236 <sup>ab</sup>    | 0.018                 | 0.65              | 2.11              | 37.97               | 131.00 <sup>a</sup>                 | 441.00                | 0.777                | 0.543               | 0.65                | 0.347              | 171 <sup>abc</sup>    | 23.12              | 2.11              |
| Acc 121    | 0.12                         | 2.80            | 4.80            | 224.00               | 0.017                 | 0.61              | 1.96              | 37.66               | 132.00 <sup>a</sup>                 | 436.67                | 0.767                | 0.538               | 0.64                | 0.357              | 169 <sup>abc</sup>    | 24.25              | 1.96              |
| Acc 131    | 0.11                         | 3.10            | 5.20            | 228.00               | 0.019                 | 0.64              | 2.03              | 42.94               | 128.00 <sup>a</sup>                 | 438.00                | 0.779                | 0.535               | 0.64                | 0.359              | 168 <sup>abc</sup>    | 33.17              | 2.03              |
| Acc 150    | 0.10                         | 3.00            | 4.50            | 209.33               | 0.020                 | 0.59              | 1.91              | 40.33               | 124.33 <sup>a</sup>                 | 424.33                | 0.771                | 0.536               | 0.63                | 0.371              | 154.33                | 33.50              | 1.91              |
| Acc 151    | 0.15 <sup>ab</sup>           | 3.60            | 6.80            | 239.33 <sup>ab</sup> | 0.024                 | 0.69              | 2.27              | 38.66               | 124.33 <sup>a</sup>                 | 444.33 <sup>abc</sup> | 0.789 <sup>ab</sup>  | 0.569 <sup>ab</sup> | 0.669 <sup>ab</sup> | 0.329              | 182.33 <sup>ab</sup>  | 22.73              | 2.27              |
| Acc 175    | 0.11                         | 2.80            | 4.50            | 224.00               | 0.016                 | 0.62              | 1.80              | 36.24               | 122.00 <sup>a</sup>                 | 423.33                | 0.758                | 0.498               | 0.61                | 0.388              | 152.00                | 29.23              | 1.80              |
| Acc 177    | 0.07                         | 25.00           | 8.00            | 211.00               | 0.032                 | 0.61              | 1.14              | 111.29              | 111.00 <sup>a</sup>                 | 407.67                | 0.759                | 0.468               | 0.58                | 0.419              | 156.00                | 15.42              | 1.14              |
| Acc 179    | 0.06                         | 23.00           | 6.50            | 165.67               | 0.033                 | 0.49              | 1.08              | 105.33              | 110.67 <sup>a</sup>                 | 397.33                | 0.740                | 0.473               | 0.55                | 0.450 <sup>a</sup> | 150.67                | 21.69              | 1.08              |
| Acc 184    | 0.13                         | 27.00           | 11.00           | 222.00               | 0.041 <sup>ab</sup>   | 0.62              | 1.40              | 82.28               | 122.00 <sup>a</sup>                 | 428.67                | 0.767                | 0.520               | 0.63                | 0.367              | 182.00 <sup>ab</sup>  | 15.09              | 1.40              |
| Acc 190    | 0.16 <sup>a</sup>            | 28.00           | 12 <sup>a</sup> | 250 <sup>ab</sup>    | 0.0406 <sup>abc</sup> | 0.70              | 1.50              | 72.33               | 125.00 <sup>a</sup>                 | 435.00                | 0.778                | 0.512               | 0.64                | 0.358              | 190 <sup>ab</sup>     | 14.33              | 1.50              |
| Acc 197    | 0.06                         | 26.00           | 9.50            | 166.67               | 0.043 <sup>a</sup>    | 0.52              | 1.58              | 153.33 <sup>a</sup> | 96.67 <sup>a</sup>                  | 396.67                | 0.758                | 0.484               | 0.58                | 0.418              | 146.67                | 14.67              | 1.58              |
| Acc 199    | 0.11                         | 29.00           | 11.00           | 217.00               | 0.042 <sup>ab</sup>   | 0.66              | 1.55              | 97.33               | 102.00 <sup>a</sup>                 | 408.67                | 0.770                | 0.496               | 0.60                | 0.400              | 162.00                | 13.76              | 1.55              |
| Acc 200    | 0.09                         | 30 <sup>a</sup> | 11.50           | 245 <sup>ab</sup>    | 0.039 <sup>abc</sup>  | 0.67              | 1.61              | 123.11              | 123.00 <sup>a</sup>                 | 424.33                | 0.767                | 0.580               | 0.59                | 0.407              | 183.00 <sup>ab</sup>  | 14.76              | 1.61              |
| LSD        | 0.014                        | 0.014           | 0.014           | 48.586               | 0.009                 | 0.014             | 0.014             | 7.586               | 48.586                              | 48.586                | 0.014                | 0.014               | 0.014               | 0.014              | 48.586                | 15.754             | 0.014             |
| CV %       | 29.89                        | 111.45          | 42.12           | 16.38                | 50.8                  | 9.8               | 21.46             | 59.12               | 22.3                                | 5.86                  | 2.12                 | 7.82                | 5.81                | 10.38              | 19.28                 | 53.81              | 21.46             |
| p value    | <.001                        | <.001           | <.001           | 0.004                | <.001                 | <.001             | <.001             | <.001               | 0.966                               | 0.024                 | <.001                | <.001               | <.001               | <.001              | 0.105812              | <.001              | <.001             |

Different upper-case letters within a column indicate significant difference among accessions. gs; (mmol m<sup>-2</sup> s<sup>-1</sup>), T; transpiration rate (mmol H<sub>2</sub>O m<sup>-2</sup> s<sup>-1</sup>), A; (μmol CO<sub>2</sub> m<sup>-2</sup> s<sup>-1</sup>), A/Ci; (μmol. mol<sup>-1</sup>), Ci; (μmol. mol<sup>-1</sup>), WUEi; [(μmol (CO<sub>2</sub>) m<sup>-2</sup>); WUEinst, (μmol. mol<sup>-1</sup>), Fv/Fm; (ratio); ΦPSII, the effective quantum efficiency of PSII photochemistry; qP, photochemical quenching; qN, non-photochemical quenching; ETR, (μmol e<sup>-1</sup> m<sup>-2</sup> s<sup>-1</sup>); ETR/A, (μmol e μmol<sup>-1</sup> CO<sub>2</sub>); AES, alternative electron sinks.



**Fig. 3.** Effect of drought stress in light adapted leaves of Bambara groundnut landrace accessions: (a) stomatal conductance (gs), (b) transpiration rate (T), (c) net CO<sub>2</sub> assimilation rate (A), (d) intercellular CO<sub>2</sub> concentration (Ci), (e) CO<sub>2</sub> assimilation rate/intercellular CO<sub>2</sub> concentration (A/Ci) (f) ratio of intercellular and atmospheric CO<sub>2</sub> (Ci/Ca), (g) intrinsic water use efficiency (A/gs), (h) instantaneous water-use efficiency (A/T), (i) minimum fluorescence (Fo'), (j) maximum fluorescence (Fm'), (k) maximum quantum efficiency of photosystem II photochemistry (Fv/Fm), (l) effective quantum efficiency of PSII photochemistry (ΦPSII), (m) photochemical quenching (qP), (n) non-photochemical quenching (qN), (o) electron transport rate (ETR), (p) relative measure of electron transport to oxygen molecules (ETR/A), (q) alternative electron sink (AES).

96, which had high values for petiole length (PL), leaf length (LL), and stress susceptibility index (SSI). Cluster 2 comprising Acc 95, Acc 97, Acc 184, Acc 78, Acc 177, Acc 100, Acc 117, Acc 200, Acc 87, and Acc 55 showed superior performance for yield stability index (YSI), stress tolerance index (STI), geometric mean productivity (GMP), mean productivity (MP), seed yield (SY), and leaf length (LL). Cluster 3 included accessions such as Acc 61, Acc 131, Acc 179, Acc 197, Acc 82, Acc 151, Acc 190, Acc 175, and Acc 199, which were notable for high values in tolerance (TOL), total biomass (TB), and plant height (PH). For physiological responses under drought (Fig. 5c), cluster 1 comprising Acc 25, Acc 82, Acc 105, Acc 117, Acc 87, Acc 100, and Acc 151 exhibited strong physiological adaptation. These accessions showed high values for stomatal conductance (gs), maximum quantum efficiency (Fv/Fm), maximum fluorescence (Fm'), photochemical quenching (qP), electron transport rate (ETR), intercellular CO<sub>2</sub> concentration (Ci), Ci/Ca ratio, minimum fluorescence (Fo'), net photosynthetic rate (A), water use efficiency (WUE<sub>inst</sub> and WUE<sub>i</sub>), ETR/A, ΦPSII, and alternative electron sink (AES). Cluster 2, including Acc 95 and Acc 78, had the lowest values across these traits, indicating poor physiological performance. Cluster 3 displayed reduced or negative values for key photosynthetic parameters such as gs, Fv/Fm, Fm', qP, ETR, and Ci, reflecting high sensitivity to drought. Under non-stress conditions (Fig. 5d), Acc 25 cluster 1 stood out for high values of Ci/Ca, ΦPSII, qN, A/Ci, Fv/Fm, and qP, indicative of efficient photosynthesis and photoprotection. Cluster 2 comprising accessions such as Acc 55, Acc 175, Acc 121, Acc 150, Acc 177, Acc 179, Acc 197, Acc 190, Acc 199, Acc 184, and Acc 200 exhibited moderate to high values for WUE<sub>i</sub> and Fv'/Fm', suggesting good water-use efficiency and photochemical performance. Cluster 3, which included Acc 61, Acc 96, Acc 117, Acc 105, Acc 131, Acc 87, Acc 100, Acc 82, and Acc 151, showed high gs, ETR, Ci, Fo', and Fm', reflecting enhanced gas exchange and chlorophyll fluorescence traits under optimal growing conditions.

### 3.6. Correlation among morphological, physiological, and fluorescence traits

Pearson correlation analysis was conducted to uncover key relationships among traits influencing drought tolerance mechanisms in Bambara groundnut accessions. Under drought stress conditions (Fig. 6a), strong and significant positive correlations were observed among agronomic traits and drought tolerance indices. Leaf length (LL) and petiole length (PL) showed a very high correlation ( $r = 0.90$ ), while total biomass (TB) was strongly associated with seed yield (SY) ( $r = 0.90$ ). Stress tolerance index (STI) exhibited higher correlations with both yield stability index (YSI) and geometric mean productivity (GMP) ( $r = 1.00$  and  $r = 0.90$ , respectively). Similarly, mean productivity (MP) was perfectly correlated with GMP and YSI ( $r = 1.00$  for both), underscoring their shared influence on yield performance under stress. Under non-stress conditions (Fig. 6b), LL and PL remained highly correlated ( $r = 0.80$ ). Leaf width (LW) displayed strong associations with plant height (PH) ( $r = 0.90$ ), seed yield (SY) ( $r = 0.70$ ), total biomass (TB) ( $r = 0.70$ ), and number of petioles per plant (NPP) ( $r = 0.70$ ). Stress stability index (STI) continued to exhibit strong correlations with tolerance index (TOL), mean productivity (MP), and geometric mean productivity (GMP) ( $r = 0.90$ ,  $1.00$ , and  $1.00$ , respectively), indicating its consistent role in productivity assessment. Physiological trait correlations under drought stress (Fig. 6c) revealed significant positive relationships among photosynthetic and water-use parameters. Net photosynthetic rate (A) was strongly correlated with A/Ci ( $r = 0.90$ ), while stomatal conductance (gs) correlated with the Ci/Ca ratio ( $r = 0.90$ ). Transpiration rate (T) also showed a strong association with A/Ci ( $r = 0.90$ ), and intercellular CO<sub>2</sub> concentration (Ci) correlated with electron transport rate (ETR) ( $r = 0.90$ ). Additionally, Ci/Ca was positively associated with ETR ( $r = 0.90$ ), and intrinsic water-use efficiency (WUE<sub>i</sub>) exhibited a perfect correlation with apparent electron sink (AES) ( $r = 1.00$ ), highlighting

Table 8

Summary of factor loadings, eigenvalues, Kaiser-Meyer-Olkin measure of sampling adequacy, percent and cumulative variation for physiological parameters.

| Trait         | Drought stress |        |        |        |        | Non stress |        |        |        |        |
|---------------|----------------|--------|--------|--------|--------|------------|--------|--------|--------|--------|
|               | PC1            | PC2    | PC3    | PC4    | PC5    | PC1        | PC2    | PC3    | PC4    | PC5    |
| LL            | 0.044          | -0.264 | -0.007 | 0.283  | -0.216 | -0.113     | 0.006  | 0.047  | 0.215  | -0.129 |
| LW            | 0.010          | -0.116 | 0.158  | -0.063 | -0.344 | -0.266     | -0.071 | 0.058  | -0.050 | 0.150  |
| PL            | 0.026          | -0.274 | -0.075 | 0.292  | -0.180 | -0.069     | 0.006  | -0.009 | 0.262  | -0.401 |
| PH            | -0.032         | -0.100 | 0.170  | -0.370 | -0.451 | -0.245     | -0.124 | 0.035  | -0.007 | 0.310  |
| NPP           | -0.058         | -0.014 | 0.052  | -0.361 | -0.511 | -0.217     | -0.085 | 0.096  | 0.012  | 0.264  |
| TB            | -0.043         | -0.018 | 0.381  | -0.153 | 0.139  | -0.251     | -0.059 | 0.116  | -0.047 | 0.215  |
| SY            | -0.075         | -0.068 | 0.400  | -0.078 | 0.137  | -0.303     | -0.025 | 0.109  | 0.064  | 0.114  |
| STI           | -0.025         | -0.232 | 0.331  | 0.053  | 0.099  | -0.186     | -0.012 | 0.081  | 0.385  | -0.087 |
| SSI           | -0.047         | 0.177  | 0.260  | -0.300 | 0.189  | 0.173      | 0.055  | -0.044 | 0.278  | -0.316 |
| TOL           | 0.044          | -0.305 | -0.115 | 0.252  | -0.157 | -0.280     | -0.037 | 0.102  | -0.119 | 0.171  |
| MP            | -0.013         | -0.325 | 0.178  | 0.174  | -0.046 | -0.284     | -0.011 | 0.101  | 0.223  | 0.047  |
| GMP           | -0.041         | -0.244 | 0.323  | 0.073  | 0.055  | -0.201     | -0.006 | 0.065  | 0.389  | -0.056 |
| YSI           | -0.025         | -0.232 | 0.331  | 0.053  | 0.099  | -0.186     | -0.012 | 0.081  | 0.385  | -0.087 |
| gs            | 0.284          | -0.034 | 0.014  | -0.112 | 0.018  | -0.026     | -0.208 | 0.256  | -0.126 | -0.184 |
| T             | -0.102         | -0.285 | -0.172 | -0.216 | 0.190  | -0.249     | 0.076  | -0.195 | -0.156 | -0.211 |
| A             | 0.040          | -0.300 | -0.179 | -0.240 | 0.158  | -0.251     | -0.005 | -0.124 | -0.205 | -0.273 |
| Ci            | 0.284          | -0.099 | 0.060  | 0.000  | 0.099  | -0.046     | -0.280 | 0.248  | -0.032 | -0.147 |
| A/Ci          | -0.066         | -0.252 | -0.222 | -0.293 | 0.116  | -0.029     | -0.253 | -0.325 | 0.050  | 0.021  |
| Ci/Ca         | 0.270          | -0.145 | 0.030  | -0.037 | 0.039  | -0.016     | -0.334 | -0.221 | 0.061  | 0.000  |
| WUEi          | 0.271          | 0.096  | 0.031  | -0.043 | -0.194 | 0.231      | -0.211 | 0.146  | 0.109  | 0.157  |
| WUEinst       | -0.135         | -0.248 | -0.189 | -0.193 | 0.085  | -0.211     | 0.120  | -0.222 | -0.152 | -0.100 |
| Fo'           | 0.265          | 0.062  | 0.065  | 0.035  | 0.146  | -0.044     | -0.262 | 0.202  | -0.205 | -0.255 |
| Fm'           | 0.302          | -0.002 | 0.057  | 0.025  | 0.078  | -0.021     | -0.276 | 0.241  | -0.124 | -0.233 |
| Fv/Fm         | 0.286          | -0.050 | -0.019 | -0.096 | 0.028  | 0.001      | -0.284 | -0.292 | 0.073  | 0.036  |
| ΦPSII         | 0.247          | -0.003 | 0.011  | -0.169 | 0.045  | 0.027      | -0.342 | -0.211 | 0.039  | 0.018  |
| qP            | 0.303          | 0.034  | 0.007  | -0.013 | -0.004 | 0.031      | -0.340 | -0.212 | 0.050  | 0.013  |
| qN            | -0.290         | -0.028 | -0.018 | -0.073 | 0.034  | -0.034     | -0.162 | -0.377 | 0.089  | 0.075  |
| ETR           | 0.272          | -0.127 | -0.050 | -0.072 | 0.121  | 0.083      | -0.265 | 0.258  | -0.093 | -0.082 |
| ETR/A         | -0.090         | 0.244  | 0.180  | 0.209  | -0.038 | 0.241      | 0.045  | 0.096  | 0.259  | 0.241  |
| AES           | 0.271          | 0.096  | 0.031  | -0.043 | -0.194 | 0.231      | -0.211 | 0.146  | 0.109  | 0.157  |
| Eigenvalue    | 10.310         | 6.790  | 5.230  | 2.330  | 1.570  | 8.990      | 6.330  | 5.460  | 3.610  | 2.070  |
| Variability % | 34.360         | 22.620 | 17.440 | 7.780  | 5.230  | 29.970     | 21.100 | 18.190 | 12.040 | 6.910  |
| Cumulative %  | 34.360         | 56.980 | 74.420 | 82.200 | 87.430 | 29.970     | 51.070 | 69.260 | 81.310 | 88.220 |

LL; leaf length, LW; leaf width, PH; plant height, PL; petiole length, SY; seed yield, TB; total biomass, NPP; number of petioles per plant, STI; stress tolerance index, SSI; stress susceptibility index, TOL; tolerance, MP; mean productivity index, GMP; geometric mean productivity, YSI; yield stability index gs; (mmol m<sup>-2</sup> s<sup>-1</sup>), T; transpiration rate (mmol H<sub>2</sub>O m<sup>-2</sup> s<sup>-1</sup>), A; net CO<sub>2</sub> assimilation rate (μmol CO<sub>2</sub> m<sup>-2</sup> s<sup>-1</sup>), A/Ci; CO<sub>2</sub> assimilation rate/intercellular CO<sub>2</sub> concentration (μmol. mol m<sup>-1</sup>), Ci; intercellular CO<sub>2</sub> concentration (μmol. mol m<sup>-1</sup>), WUEi; intrinsic water use efficiency [(μmol (CO<sub>2</sub>) m<sup>-2</sup>)]' WUEinst; instantaneous water-use efficiency (μmol. mol<sup>-1</sup>), Fv/Fm; maximum quantum efficiency of photosystem II photochemistry (ratio), ΦPSII; the effective quantum efficiency of PSII photochemistry the effective quantum efficiency of PSII photochemistry, qP; photochemical quenching, qN; non-photochemical quenching, ETR; electron transport rate (μmol e<sup>-1</sup> m<sup>-2</sup> s<sup>-1</sup>), ETR/A; relative measure of electron transport to oxygen molecules (μmol e μmol<sup>-1</sup> CO<sub>2</sub>), AE; alternative electron sinks.

the tight coordination between photosynthetic efficiency and water-use dynamics under stress. Under non-stress conditions (Fig. 6d), intercellular CO<sub>2</sub> concentration (Ci) correlated strongly with maximum fluorescence (Fm') (r = 0.90), and ETR was also highly correlated with Fm' (r = 0.90). Non-photochemical quenching (qN) showed a perfect correlation with A/Ci (r = 1.00). Moreover, WUEi and minimum fluorescence (Fo') were strongly associated with AES and ETR (r = 1.00 and 0.80, respectively), illustrating the functional integration of gas exchange and chlorophyll fluorescence traits under favourable growing conditions.

## 4. Discussion

### 4.1. Impact of drought stress on growth and yield performance of Bambara Groundnut Accessions

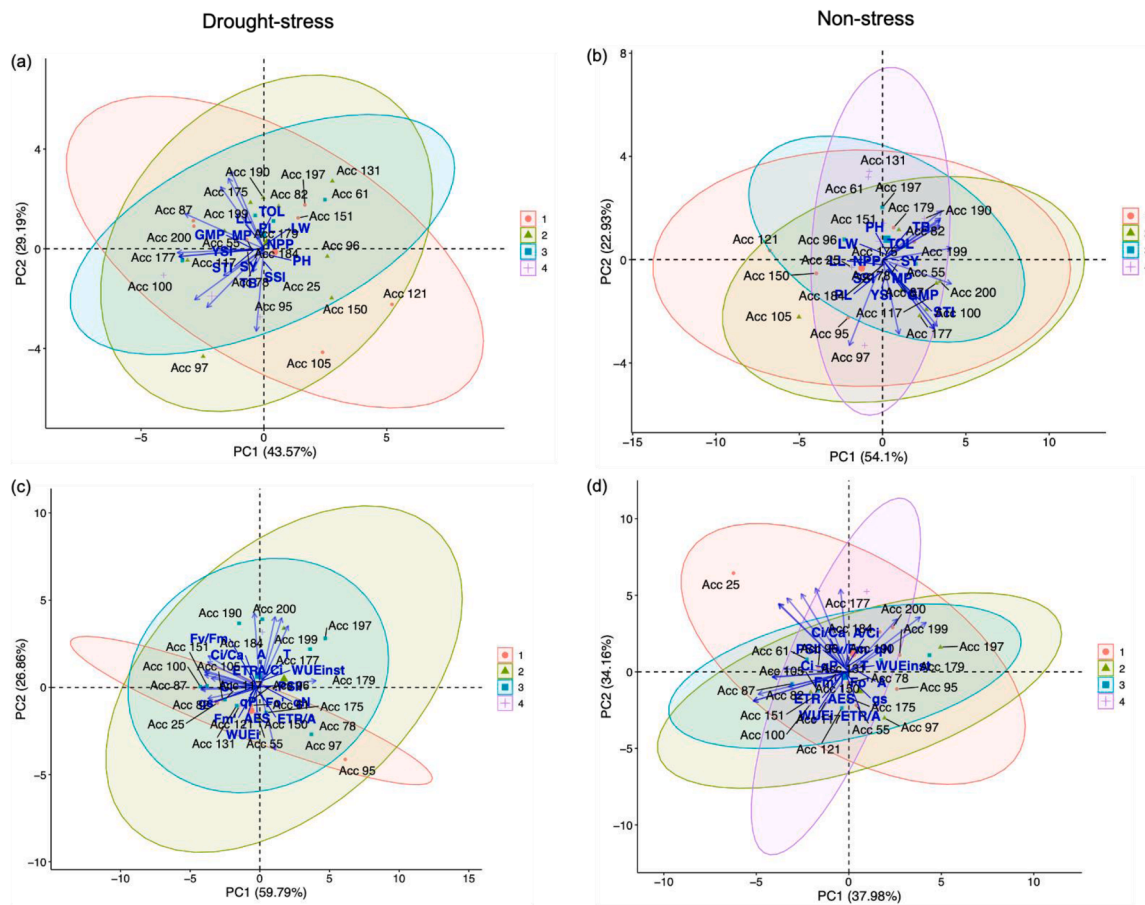
Drought stress significantly affected the growth and yield of Bambara groundnut accessions, with variations in several traits under both stressed and non-stressed conditions. The analysis revealed that leaf length, total biomass, and seed yield were all impacted by drought stress. Leaf length, for example, showed significant variation between conditions, with Acc 199, Acc 151, and Acc 175 exhibiting the longest leaves under stress, suggesting that these accessions may possess some level of drought tolerance (Lateef & Tahir, 2024; Young et al., 2004). However, the total biomass and seed yield were more drastically reduced under drought, indicating a substantial reduction in overall

plant productivity (Dietz et al., 2021).

The violin plots further illustrated the effects of drought stress on growth and yield traits. Under drought conditions, traits like leaf length, leaf width, petiole length, and plant height exhibited wider distributions, reflecting greater variability among accessions, possibly due to the differential tolerance levels to water scarcity (Zahedi et al., 2025). Total biomass was most severely affected, as shown by the leftward shift in its distribution, highlighting the inhibitory impact of drought on plant growth and development. In contrast, under non-stress conditions, the distributions were more centralized, indicating a more uniform response across accessions. These findings emphasize the significant negative impact of drought on Bambara groundnut, but also suggest potential for identifying drought-tolerant accessions with better resilience under water-limited conditions (Kunene et al., 2022).

### 4.2. Assessment of Bambara groundnut accessions' performance under drought stress using drought indices

The variability observed among Bambara groundnut accessions in drought tolerance indices highlights the diverse adaptive responses within the germplasm (Khan et al., 2021; Mayes et al., 2019). Notably, accessions such as Acc 200, Acc 190, Acc 175, and Acc 87, which exhibited high tolerance index (TOL) values and high performance under both stress and non-stress conditions, are promising candidates for dual environment productivity. Acc 200, in particular, combined the highest TOL (20.00) with a high yield stability index (YSI = 0.47),



**Fig. 4.** Biplots of principal component analysis (PCA) showing phenotypic diversity among Bambara groundnut accessions. (a) and (b) show the PCA of agronomic traits and stress tolerance indices under drought-stress and non-stress conditions, respectively. (c) and (d) show PCA based on leaf gas exchange and chlorophyll fluorescence parameters under drought-stress and non-stress conditions, respectively. The axes represent the first two principal components (PC1 and PC2), with percentages indicating the amount of total variance explained. Accessions are grouped based on clustering patterns, and confidence ellipses represent the variability within each group. Arrows indicate the contribution and direction of each trait to the principal components.

indicating strong adaptability and productivity potential (Farshadfar et al., 2013). On the other hand, accessions like Acc 177, Acc 95, and Acc 97, which recorded high yields under stress ( $Y_s$ ), strong stress tolerance index (STI), and stable YSI values, reflect intrinsic drought resilience traits. This aligns with findings by Mandizvo et al. (2022), who emphasized the STI as a useful index to identify genotypes that perform well under both stress and non-stress conditions. The low stress susceptibility index (SSI) values observed in Acc 197, Acc 190, Acc 175, and Acc 82 further underscore their superior drought tolerance, as lower SSI values denote less yield reduction under stress (Ali et al., 2013). Moreover, accessions such as Acc 55, Acc 78, Acc 82, Acc 87, Acc 97, and Acc 200 demonstrated high mean productivity (MP) and geometric mean productivity (GMP), suggesting stable and consistent yield performance across environments. This trait is particularly desirable in drought-prone regions, where yield consistency is critical for food security (Abebe et al., 2020). The combination of high GMP, low SSI, and favourable STI in several accessions emphasizes their suitability for inclusion in drought tolerance breeding programs aimed at improving tolerance and yield stability under climate variability (Mohammadi & Geravandi, 2024).

#### 4.3. Adaptive mechanisms and genotypic variability underlying drought tolerance in Bambara groundnut

Drought stress significantly influenced key physiological traits in Bambara groundnut, as reflected by decrease in gas exchange and chlorophyll fluorescence parameters. The significant effects of accession

and treatment, along with the interaction for traits such as stomatal conductance ( $g_s$ ), photosynthetic rate ( $A$ ), and water-use efficiency (WUE), underscore genotypic variability in drought response mechanisms (Haghpanah et al., 2024; Roche, 2015). The marked declines in  $g_s$ ,  $A$ , and  $A/C_i$  across accessions indicate both stomatal and non-stomatal limitations to photosynthesis under drought, a common stress response in legumes (Pilon et al., 2018). Accessions such as Acc 25 and Acc 61 maintained higher  $A$  and WUEi values under stress, suggesting effective stomatal regulation and sustained carbon assimilation key traits for drought adaptation (Zahedi et al., 2025). Chlorophyll fluorescence traits, particularly  $F_v/F_m$  and  $\Phi_{PSII}$ , also declined under drought, reflecting impaired PSII photochemistry (Mihaljević et al., 2021). However, moderate reductions in these parameters in Acc 25, Acc 61, and Acc 87 indicate enhanced photoprotection and better stress tolerance (Sofo, 2011). The relative stability of ETR and ETR/A in these accessions suggests efficient electron transport maintained under limited carbon fixation, possibly through alternative sinks such as photorespiration or the water cycle (Perera-Castro & Flexas, 2023). The significant reduction in AES among drought-sensitive accessions further highlights their limited capacity to manage excess excitation energy (Qiao et al., 2024). Overall, these findings demonstrate clear physiological differentiation among accessions, with some maintaining higher gas exchange and PSII efficiency under stress.

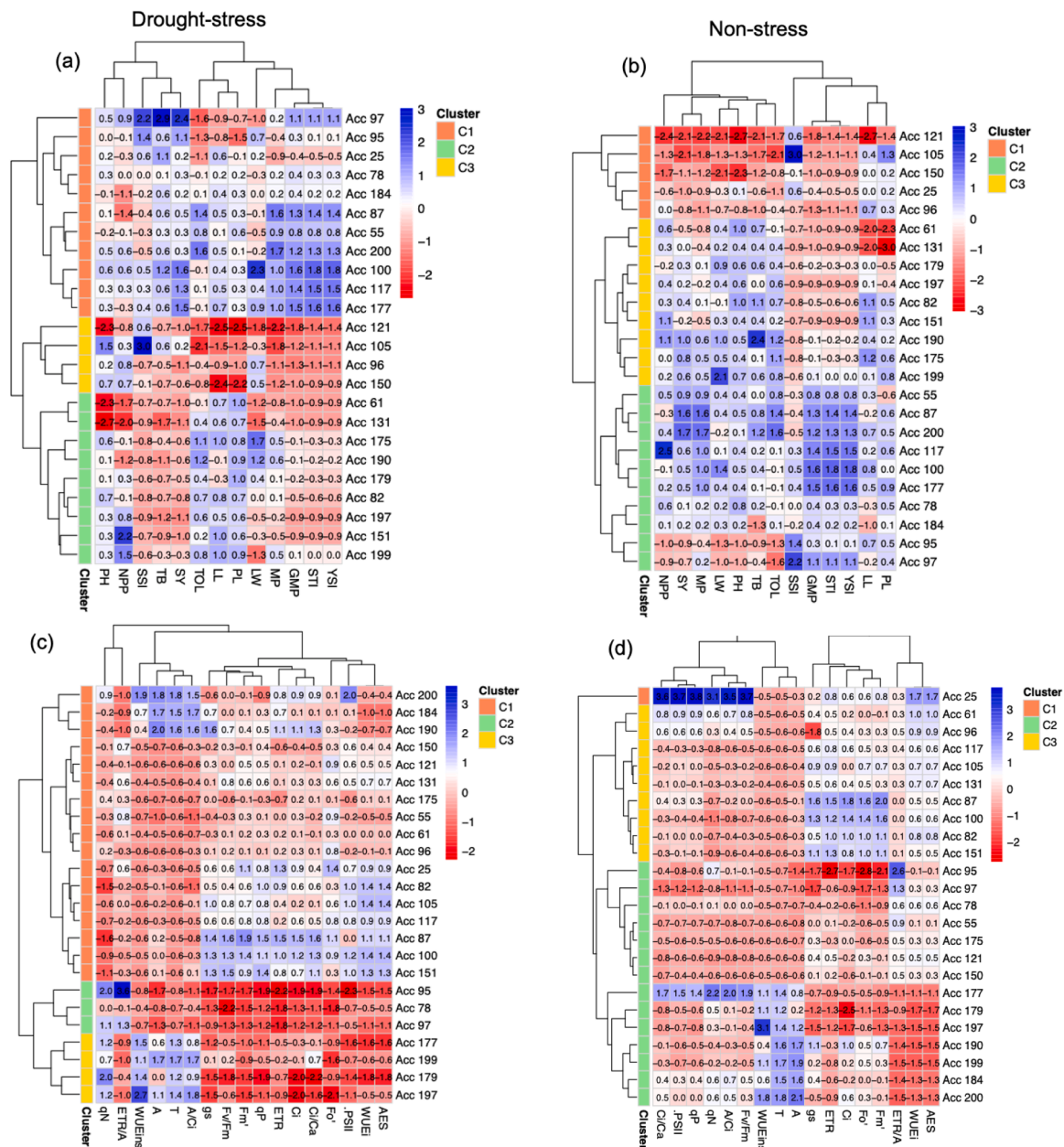


Fig. 5. Hierarchical clustering heatmaps illustrating phenotypic variation among Bambara groundnut accessions. (a) and (b) show agronomic traits and stress tolerance indices under drought-stress and non-stress conditions, (c) and (d) show the clustering under drought-stress and non-stress conditions based on leaf gas exchange and chlorophyll fluorescence parameters. Traits are represented by standardized values (Z-scores), with blue indicating higher performance and red indicating lower performance.

#### 4.4. Trait dynamics and adaptive mechanisms in Bambara groundnut under stress and non-stress conditions

The PCA results revealed distinct trait clusters associated with drought tolerance and physiological adaptability in Bambara groundnut, underscoring the multivariate complexity of drought response. The high contribution of gas exchange and chlorophyll fluorescence traits (e.g., *gs*, *Ci*, *ETR*, *Fv/Fm*) to PC1 under drought conditions suggests that photosynthetic efficiency and electron transport are critical determinants of stress adaptation (Badr & Brüggemann, 2020). This also aligns with findings in other leguminous crops where efficient photochemical processes contribute to drought resilience (Behzadi et al., 2023). The separation of yield-related traits (*TB*, *SY*) in PC3 and morphological attributes in PC4 and PC5 implies that while morphology may influence drought response, physiological and metabolic traits play

a more central role in maintaining productivity under stress (Anjum et al., 2011). Accessions like Acc 200 and Acc 177, which clustered with indices such as *STI* and *GMP*, exhibited a more integrated drought-tolerant profile, balancing yield stability with stress physiology. Notably, traits such as *WUEinst*, *qN*, and *AES* were key contributors to variation in physiological adaptation, suggesting their potential as reliable markers in drought screening (Cai et al., 2020). Under non-stress conditions, the broader dispersion of accessions reflects the crop's inherent phenotypic plasticity, but the strong association of traits like *MP* and *GMP* with high-yielding accessions indicates that these indices retain predictive value even in optimal environments (Mandizvo et al., 2022). These patterns reinforce the utility of multivariate approaches in identifying superior genotypes with stable performance across environments (Kunene et al., 2025).

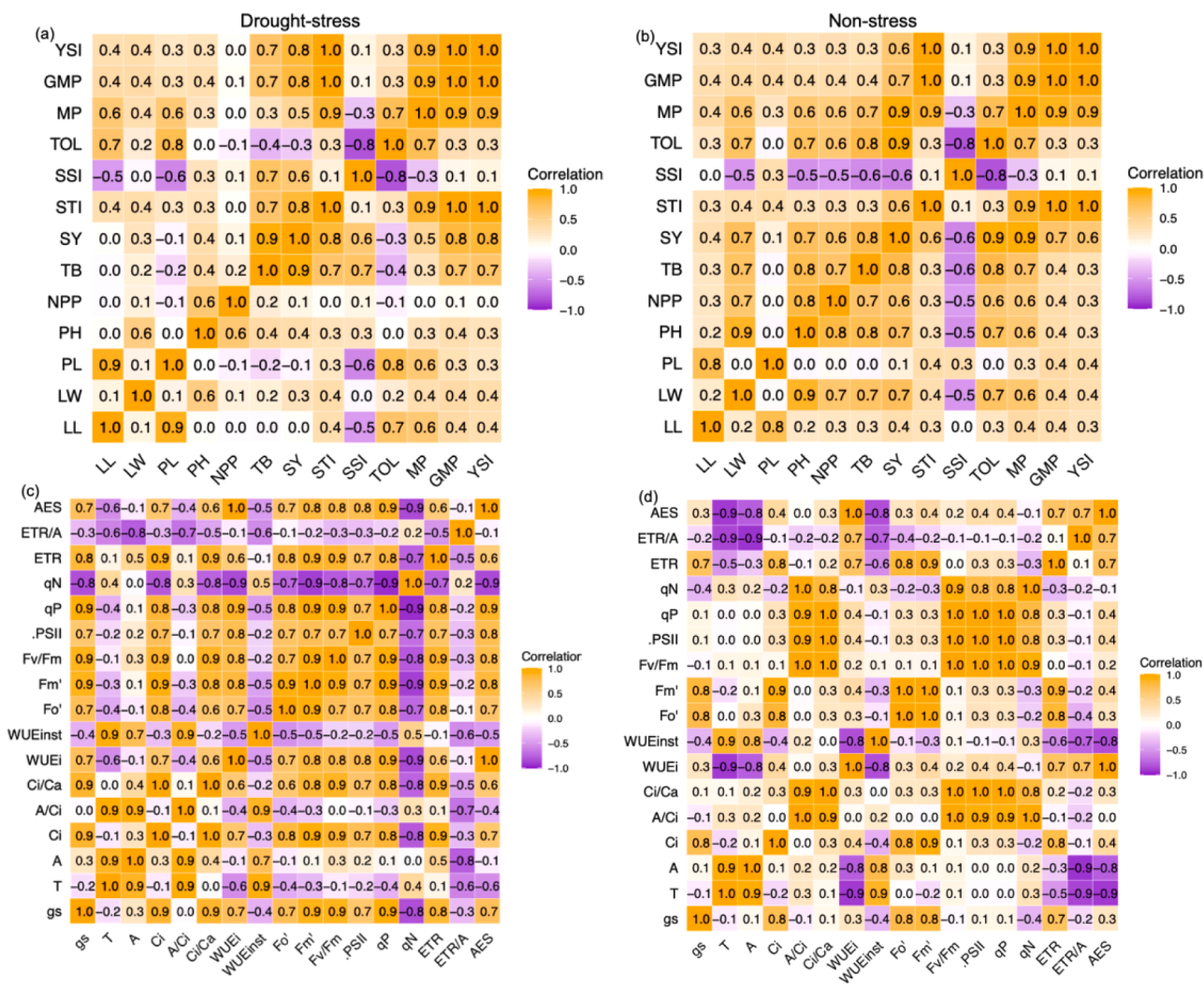


Fig. 6. Correlation matrices illustrating relationships among agronomic traits and drought tolerance indices (a-b) as well as leaf gas exchange and chlorophyll fluorescence parameters (c-d) under drought-stress and non-stress conditions.

4.5. Trait-based grouping of accessions for improved drought tolerance and yield potential performance

The hierarchical clustering analysis provided clear evidence of phenotypic divergence among Bambara groundnut accessions under both drought-stress and non-stress conditions, reflecting distinct adaptation strategies and potential for targeted selection. Under drought stress, cluster 1 accessions (e.g., Acc 200, Acc 100, Acc 184) were consistently associated with high yield-related indices such as STI, GMP, MP, and SY, underscoring their resilience and stable performance under water deficit (Mohammadi & Geravandi, 2024). In contrast, cluster 2 accessions exhibited larger vegetative structures (e.g., LL, PL) and higher TOL, suggesting a growth-oriented drought escape mechanism that may be less efficient under prolonged drought (Salekdeh et al., 2002). Accessions in cluster 3 showed high SSI and NPP but poor yield performance, indicating higher drought susceptibility, which is consistent with stress-sensitive phenotypes where growth is not efficiently translated into reproductive success (Chattopadhyay et al., 2018). Under non-stress conditions, a shift in clustering revealed genotype-environment interaction effects (Matonger et al., 2023). High-yielding accessions remained consistent in cluster 2, showing their stability across environments (Mishra et al., 2024). Meanwhile, accessions in clusters 1 and 3 varied in their trait associations, indicating the influence of optimal conditions on morphological and physiological

expression (Chhetri et al., 2019). These results emphasize the importance of multi-environment testing to accurately classify accessions for breeding purposes (Pandey et al., 2020). Physiological clustering further delineated accessions with superior photosynthetic efficiency and water-use traits (de Miguel et al., 2014). Under drought, cluster 1 accessions (e.g., Acc 25, Acc 100, Acc 151) exhibited high  $g_s$ , Fv/Fm, qP, ETR, and Ci, suggesting robust photochemical capacity and stomatal regulation key mechanisms in sustaining photosynthesis under stress (Chaves et al., 2009; Lawson et al., 2011). These physiological profiles, coupled with efficient water-use (WUEinst, WUEi), highlight their potential for drought-resilient productivity. Accessions like Acc 25 exhibited high photochemical and photoprotective traits, including  $\Phi$ PSII, qN, and A/Ci, pointing to efficient energy use and dissipation mechanisms in non-stress environments (Baroni et al., 2024). Meanwhile, accessions in clusters 2 and 3 displayed diverse combinations of gas exchange and fluorescence traits, with some (e.g., cluster 3) maintaining high  $g_s$  and ETR, potentially reflecting genotypic differences in photosynthetic regulation even under favourable conditions (Jiang et al., 2006).

4.6. Trait interactions and their relevance to stress tolerance and phenotypic stability

The Pearson correlation analysis highlighted strong

interrelationships among agronomic and physiological traits critical to drought adaptation in Bambara groundnut, showing the complex yet coordinated nature of stress response mechanisms (Gao et al., 2023). Under drought stress, agronomic traits such as leaf length and petiole length ( $r = 0.90$ ), and seed yield with total biomass ( $r = 0.90$ ), demonstrated tightly linked growth and productivity traits, while indices like STI, GMP, MP, and YSI were perfectly correlated ( $r = 1.00$ ), confirming their collective reliability for identifying high-yielding, drought-resilient genotypes (Sofi et al., 2021; Zhao et al., 2019). Physiologically, net photosynthesis was strongly aligned with stomatal and electron transport traits, including A/Ci and ETR ( $r = 0.90$ ), while water-use efficiency (WUEi) was perfectly associated with the apparent electron sink (AES) ( $r = 1.00$ ), showing the important role of gas exchange, photochemistry, and drought tolerance (Buezo et al., 2019; Singh et al., 2022). Under non-stress conditions, similar trait coordination persisted, particularly between fluorescence indicators (Fm', Fo') and photosynthetic efficiency metrics such as ETR and qN, highlighting the genotype-dependent maintenance of photosynthetic integrity even under favourable environments (Gomes et al., 2020).

## 5. Conclusion

This study demonstrates that drought stress have a significant influence on growth, yield, physiological, and photochemical traits in Bambara groundnut. Significant genotypic variation was observed across evaluated traits, confirming the presence of both sensitive and resilient accessions within the conditions. Accessions such as Acc 25, Acc 61, Acc 87, Acc 97, and Acc 200 consistently exhibited superior physiological performance and yield stability under drought stress, indicating their potential as candidates for developing drought-resilient cultivars. The integration of morphological, physiological, and drought index-based assessments enabled the identification of key traits such as water-use efficiency, photosynthetic rate, and biomass accumulation that contribute to drought adaptation. The use of principal component analysis further emphasized the centrality of these traits in driving phenotypic diversity. Taken together, these findings provide critical insights for breeding programs and underline the adaptive potential of Bambara groundnut as a climate-resilient legume for sustainable agriculture in water-limited regions.

## Data availability

The data that supports the findings of this study are available from the corresponding author upon reasonable request.

## CRedit authorship contribution statement

**Sithembile Kunene:** Conceptualization, Formal analysis, Writing – original draft, Investigation, Validation, Data curation, Methodology. **Abe Shegro Gerrano:** Data curation, Writing – review & editing, Project administration, Conceptualization, Supervision. **Alfred Oduor Odindo:** Formal analysis, Writing – review & editing, Methodology, Conceptualization, Supervision.

## Declaration of competing interest

The authors declare that there are no conflicts of interest related to this research. This study was conducted without any financial support or external influence from commercial entities. All authors have contributed equally to the research and writing of this manuscript. There are no affiliations, funding sources, or personal relationships that could have appeared to influence the research presented in this paper.

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## References

- Abebe, T., Belay, G., Tadesse, T., Keneni, G., 2020. Selection efficiency of yield based drought tolerance indices to identify superior sorghum [*Sorghum bicolor* (L.) Moench] genotypes under two-contrasting environments. *Afr J Agric Res* 15 (3), 379–392.
- Abu, H.B., Buah, S.S.J., 2011. Characterization of bambara groundnut landraces and their evaluation by farmers in the Upper West Region of Ghana. *J. Dev. Sustain. Agric.* 6 (1), 64–74.
- Ali, S., Gautam, R., Mahajan, R., Krishnamurthy, S., Sharma, S., Singh, R., Ismail, A., 2013. Stress indices and selectable traits in SALTOL QTL introgressed rice genotypes for reproductive stage tolerance to sodicity and salinity stresses. *Field Crops Res.* 154, 65–73.
- Anjum, S.A., Xie, X.Y., Wang, L.C., Saleem, M.F., Man, C., Lei, W., 2011. Morphological, physiological and biochemical responses of plants to drought stress. *Afr J Agric Res* 6 (9), 2026–2032.
- Ayed, S., Othmani, A., Bouhaouel, I., Teixeira da Silva, J.A., 2021. Multi-environment screening of durum wheat genotypes for drought tolerance in changing climatic events. *Agronomy* (5), 11. <https://doi.org/10.3390/agronomy11050875>.
- Badr, A., Brüggemann, W., 2020. Comparative analysis of drought stress response of maize genotypes using chlorophyll fluorescence measurements and leaf relative water content. *Photosynthetica* 58 (2), 38–645.
- Baroni, D.F., de Souza, G.A., Bernardo, W.d.P., Santos, A.R., Barcellos, L.C.d.S., Barcelos, L.F., Correia, L.Z., de Almeida, C.M., Verdin Filho, A.C., Rodrigues, W.P., 2024. Stomatal and non-Stomatal leaf responses during two sequential water stress cycles in young coffee canephora plants. *Stresses* 4 (3), 575–597.
- Beebe, S.E., Rao, I.M., Blair, M.W., Acosta-Gallegos, J.A., 2013. Phenotyping common beans for adaptation to drought. *Front Physiol* 4, 35.
- Behzadi, Y., Salehi, A., Dehnavi, M.M., Balouchi, H., 2023. Improvement of biochemical and antioxidant responses of borage (*Borago officinalis* L.) under drought stress conditions with the use of vermicompost and zinc sulfate. *Physiol. Mol. Biol. Plants* 29 (12), 1881–1896.
- Belay, G.A., Zhang, Z., Xu, P., 2021. Physio-morphological and biochemical trait-based evaluation of Ethiopian and Chinese wheat germplasm for drought tolerance at the seedling stage. *Sustainability* (9), 13. <https://doi.org/10.3390/su13094605>.
- Buezo, J., Sanz-Saez, A., Moran, J.F., Soba, D., Aranjuelo, I., Esteban, R., 2019. Drought tolerance response of high-yielding soybean varieties to mild drought: physiological and photochemical adjustments. *Physiol. Plant* 166 (1), 88–104.
- Cai, K., Chen, X., Han, Z., Wu, X., Zhang, S., Li, Q., Nazir, M.M., Zhang, G., Zeng, F., 2020. Screening of worldwide barley collection for drought tolerance: the assessment of various physiological measures as the selection criteria. *Front Plant Sci.* 11, 1159.
- Chai, H.H., Massawe, F., Mayes, S., 2016. Effects of mild drought stress on the morpho-physiological characteristics of a bambara groundnut segregating population. *Euphytica* 208, 225–236.
- Chattopadhyay, K., Reddy, J., Pradhan, S., Patnaik, S., Marndi, B., Swain, P., Nayak, A., Anandan, A., Chakraborty, K., Sarkar, R., 2018. Genetic improvement of Rice for multiple stress tolerance in unfavorable rainfed ecology. *Rice Res. Enhancing Product. Profitab. Clim. Resil.* 542, 122.
- Chaves, M.M., Flexas, J., Pinheiro, C., 2009. Photosynthesis under drought and salt stress: regulation mechanisms from whole plant to cell. *Ann. Bot* 103 (4), 551–560.
- Chhetri, H.B., Macaya-Sanz, D., Kainer, D., Biswal, A.K., Evans, L.M., Chen, J.G., Collins, C., Hunt, K., Mohanty, S.S., Rosenstiel, T., 2019. Multitrait genome-wide association analysis of *Populus trichocarpa* identifies key polymorphisms controlling morphological and physiological traits. *New Phytol.* 223 (1), 293–309.
- Chibarabada, T.P., Modi, A.T., Mabhauthi, T., 2014. Seed quality characteristics of a bambara groundnut (*Vigna subterranea* L.) landrace differing in seed coat colour. *S. Afr. J. Plant Soil* 31 (4), 219–226.
- de Miguel, M., Cabezas, J.-A., de María, N., Sánchez-Gómez, D., Guevara, M.-Á., Vélez, M.-D., Sáez-Laguna, E., Díaz, L.-M., Mancha, J.-A., Barbero, M.-C., 2014. Genetic control of functional traits related to photosynthesis and water use efficiency in *Pinus pinaster* Ait. Drought response: integration of genome annotation, allele association and QTL detection for candidate gene identification. *BMC Genom.* 15, 1–19.
- Dietz, K.J., Zörb, C., Geilfus, C.M., 2021. Drought and crop yield. *Plant Biol.* 23 (6), 881–893.
- Ekbc, E., Cagran, C., Korkmaz, K., Kose, M.A., Aras, V., 2017. Assessment of watermelon accessions for salt tolerance using stress tolerance indices. *Ciênc. Agrotecnologia* 41 (6), 616–625.
- Evans, J.R., 2009. Potential errors in electron transport rates calculated from chlorophyll fluorescence as revealed by a multilayer leaf model. *Plant Cell Physiol.* 50 (4), 698–706.
- Farshadfar, E., Mohammadi, R., Farshadfar, M., Dabiri, S., 2013. Relationships and repeatability of drought tolerance indices in wheat-rye disomic addition lines. *Aust. J. Crop. Sci.* 7 (1), 130–138.
- Finch-Savage, W.E., Bassel, G.W., 2016. Seed vigour and crop establishment: extending performance beyond adaptation. *J. Exp. Bot.* 67 (3), 567–591.
- Gao, X., Chai, H.H., Ho, W.K., Mayes, S., Massawe, F., 2023. Deciphering the molecular basis for photosynthetic parameters in Bambara groundnut (*Vigna subterranea* L. Verdc) under drought stress. *BMC Plant Biol.* 23 (1), 287.

- Gomes, A.M., Rodrigues, A.P., António, C., Rodrigues, A.M., Leitão, A.E., Batista-Santos, P., Nhandumbo, N., Massinga, R., Ribeiro-Barros, A.I., Ramalho, J.C., 2020. Drought response of cowpea (*Vigna unguiculata* (L.) Walp.) Landraces at leaf physiological and metabolite profile levels. *Env. Exp. Bot.* 175, 104060.
- Grzesiak, S., Hordyńska, N., Szczyrek, P., Grzesiak, M.T., Noga, A., Szechyńska-Hebda, M., 2018. Variation among wheat (*Triticum easativum* L.) genotypes in response to the drought stress: I – selection approaches. *J. Plant Interact.* 14 (1), 30–44. <https://doi.org/10.1080/17429145.2018.1550817>.
- Haghpanah, M., Hashemipetroudi, S., Arzani, A., Araniti, F., 2024. Drought tolerance in plants: physiological and molecular responses. *Plants* 13 (21), 2962.
- Jiang, Q., Roche, D., Monaco, T., Hole, D., 2006. Stomatal conductance is a key parameter to assess limitations to photosynthesis and growth potential in barley genotypes. *Plant Biol.* 8 (04), 515–521.
- Khan, M.M.H., Rafii, M.Y., Ramlee, S.I., Jusoh, M., Al Mamun, M., Halidu, J., 2021. DNA fingerprinting, fixation-index (Fst), and admixture mapping of selected Bambara groundnut (*Vigna subterranea* [L.] Verdc.) accessions using ISSR markers system. *Sci. Rep.* 11 (1), 14527.
- Khan, M.M.H., Rafii, M.Y., Ramlee, S.I., Jusoh, M., Al-Mamun, M., 2021. Bambara groundnut (*Vigna subterranea* L. Verdc): A crop for the new millennium, its genetic diversity, and improvements to mitigate future food and nutritional challenges. *Sustainability* 13 (10), 5530.
- Kitao, M., Lei, T.T., Koike, T., Tobita, H., Maruyama, Y., 2003. Higher electron transport rate observed at low intercellular CO<sub>2</sub> concentration in long-term drought-acclimated leaves of Japanese mountain birch (*Betula ermanii*). *Physiol Plant* 118 (3), 406–413.
- Kunene, S., Gerrano, A.S., Odindo, A.O., 2025. Stability and performance of Bambara groundnut (*Vigna subterranea* (L.) verdc.) Genotypes in different South African environments. *S. Afr. J. Plant Soil* 1–12.
- Kunene, S., Odindo, A.O., Gerrano, A.S., Mandizvo, T., 2022. Screening Bambara groundnut (*Vigna subterranea* L. Verdc) genotypes for drought tolerance at the germination stage under simulated drought conditions. *Plants* 11 (24), 3562.
- Lateef, D. D., & Tahir, N. A.-r. (2024). Genetic diversity of barley accessions and their response under abiotic stresses using different approaches. *arXiv preprint arXiv: 2403.14181*.
- Lawson, T., von Caemmerer, S., Baroli, I., 2011. Photosynthesis and stomatal behaviour. *Prog. Bot.* 72, 265–304.
- Liu, F., Jensen, C.R., Andersen, M.N., 2004. Drought stress effect on carbohydrate concentration in soybean leaves and pods during early reproductive development: its implication in altering pod set. *Field Crops Res* 86 (1), 1–13.
- Mabhaudhi, T., Modi, A., Beletse, Y., 2013. Growth, phenological and yield responses of a bambara groundnut (*Vigna subterranea* L. Verdc) landrace to imposed water stress: II. Rain shelter conditions. *Water Sa* 39 (2), 191–198.
- Mandizvo, T., Odindo, A.O., 2019. Seed coat structural and imbibitional characteristics of dark and light coloured Bambara groundnut (*Vigna subterranea* L.) landraces. *Heliyon* 5 (2), e01249.
- Mandizvo, T., Odindo, A.O., Mashilo, J., Magwaza, L.S., 2022. Drought tolerance assessment of citron watermelon (*Citrullus lanatus* var. *citroides* (LH Bailey) mansf. Ex greb.) Accessions based on morphological and physiological traits. *Plant Physiol. Biochem.* 180, 106–123.
- Massawe, F., Mwale, S., Azam-Ali, S., Roberts, J., 2005. Breeding in bambara groundnut (*Vigna subterranea* (L.) Verdc.): strategic considerations. *Afr. J. Biotechnol* 4 (6), 463–471.
- Matongera, N., Ndhlela, T., van Biljon, A., Labuschagne, M., 2023. Genotype x environment interaction and yield stability of normal and biofortified maize inbred lines in stress and non-stress environments. *Cogent Food Agric.* 9 (1), 2163868.
- Mayes, S., Ho, W.K., Chai, H.H., Gao, X., Kundy, A.C., Mateva, K.I., Zahrlakmal, M., Hahiree, M.K.I.M., Kendabie, P., Licea, L., 2019. Bambara groundnut: an exemplar underutilised legume for resilience under climate change. *Planta* 250 (3), 803–820.
- Medrano, H., Tomás, M., Martorell, S., Flexas, J., Hernández, E., Rosselló, J., Pou, A., Escalona, J.-M., Bota, J., 2015. From leaf to whole-plant water use efficiency (WUE) in complex canopies: limitations of leaf WUE as a selection target. *Crop J* 3 (3), 220–228.
- Mihaljević, I., Viljevac Vuletić, M., Tomaš, V., Horvat, D., Zdunić, Z., Vuković, D., 2021. PSII photochemistry responses to drought stress in autochthonous and modern sweet cherry cultivars. *Photosynthetica* 59 (4), 517–528.
- Mishra, S., Koundinya, A., Aghora, T., 2024. Stability analysis to identify improved lines of cluster bean (*Cyamopsis tetragonoloba* L. Taub.). *Plant Genet. Resour.* 22 (3), 173–180.
- Mohammadi, R., Geravandi, M., 2024. Multi-trait selection for agronomic performance and drought tolerance among durum wheat genotypes evaluated under rainfed and irrigated environments. *Exp. Agric.* 60, e3.
- Muhammad, I., Rafii, M.Y., Ramlee, S.I., Nazli, M.H., Harun, A.R., Oladosu, Y., Musa, I., Arolo, F., Chukwu, S.C., Sani Haliru, B., 2020. Exploration of bambara groundnut (*Vigna subterranea* (L.) Verdc.), an underutilized crop, to aid global food security: varietal improvement, genetic diversity and processing. *Agronomy* 10 (6), 766.
- Muhammad, Y.Y., Massawe, F., Mayes, S., 2016. Effects of short-term water deficit stress on physiological characteristics of Bambara groundnut (*Vigna subterranea* (L.) Verdc.). *S. Afr. J. Plant Soil* 33 (1), 51–58.
- Mwale, S., Azam-Ali, S., Massawe, F., 2007. Growth and development of bambara groundnut (*Vigna subterranea*) in response to soil moisture: 1. Dry matter and yield. *Eur. J. Agron.* 26 (4), 345–353.
- Ort, D.R., Baker, N.R., 2002. A photoprotective role for O<sub>2</sub> as an alternative electron sink in photosynthesis? *Curr. Opin. Plant Biol.* 5 (3), 193–198.
- Pandey, M.K., Chaudhari, S., Jarquin, D., Janila, P., Crossa, J., Patil, S.C., Sundravandana, S., Khare, D., Bhat, R.S., Radhakrishnan, T., 2020. Genome-based trait prediction in multi-environment breeding trials in groundnut. *Theor. Appl. Genet.* 133, 3101–3117.
- Perera-Castro, A.V., Flexas, J., 2023. The ratio of electron transport to assimilation (ETR/AN): underutilized but essential for assessing both equipment's proper performance and plant status. *Planta* 257 (2), 29.
- Pilon, C., Snider, J.L., Sobolev, V., Chastain, D.R., Sorensen, R.B., Meeks, C.D., Massa, A. N., Walk, T., Singh, B., Earl, H.J., 2018. Assessing stomatal and non-stomatal limitations to carbon assimilation under progressive drought in peanut (*Arachis hypogaea* L.). *J. Plant Physiol.* 231, 124–134.
- Pour-Aboughadareh, A., Yousefian, M., Moradkhani, H., Moghaddam Vahed, M., Pocza, P., Siddique, K.H.M., 2019. iPASTIC: an online toolkit to estimate plant abiotic stress indices. *Appl. Plant Sci.* 7 (7), e11278. <https://doi.org/10.1002/aps3.11278>.
- Prasad, P., Staggengberg, S., Ristic, Z., 2008. Impacts of drought and/or heat stress on physiological, developmental, growth, and yield processes of crop plants. Response Crops Ltd. water: Underst. Model. Water stress eff. Plant growth process. 1, 301–355.
- Qiao, M., Hong, C., Jiao, Y., Hou, S., Gao, H., 2024. Impacts of drought on photosynthesis in major food crops and the related mechanisms of plant responses to drought. *Plants* 13 (13), 1808.
- Rahmah, N., Ilyas, S., Setiawan, A., 2020. Evaluation of bambara groundnut (*Vigna subterranea* L. Verdc.) genotypes for drought tolerance at germination stage. *Sabrao J. Breed. Genet.* 52, 45–63.
- Roche, D., 2015. Stomatal conductance is essential for higher yield potential of C3 crops. *CRC Crit. Rev. Plant Sci.* 34 (4), 429–453.
- Salekdeh, G.H., Siopongco, J., Wade, L.J., Ghareyazie, B., Bennett, J., 2002. Proteomic analysis of rice leaves during drought stress and recovery. *PROTEOM: Int. Ed.* 2 (9), 1131–1145.
- Sánchez-Virosta, Á., Sadras, V.O., Sánchez-Gómez, D., 2021. Phenotypic plasticity in relation to inter-cultivar variation of garlic (*Allium sativum* L.) functional performance and yield-stability in response to water availability. *Sci. Hortic.* 285. <https://doi.org/10.1016/j.scienta.2021.110128>.
- Singh, G., Goldberg, S., Schaefer, D., Zhang, F., Sharma, S., Mishra, V., Xu, J., 2022. Biochemical, gas exchange, and chlorophyll fluorescence analysis of maize genotypes under drought stress reveals important insights into their interaction and homeostasis. *Photosynthetica* 60 (3), 376.
- Sofi, P.A., Shafi, S., Singh, B., Jaiswal, J., Mishra, V., Mir, R., 2021. Combined selection for productivity and resilience through modified stress tolerance indices in a HUW-234 X HUW-468 derived wheat (*Triticum aestivum* L.) RIL mapping population for heat stress. *Electron. J. Plant Breed.* 12 (3), 612–622.
- Sofa, A., 2011. Drought stress tolerance and photoprotection in two varieties of olive tree. *Acta Agric. Scand. B-Soil Plant Sci.* 61 (8), 711–720.
- Unigwe, A.E., Gerrano, A.S., Adebola, P., Pillay, M., Monrovia, L., 2016. Morphological variation in selected accessions of Bambara groundnut (*Vigna subterranea* L. Verdc) in South Africa. *J. Agric. Sci.* 8 (11), 69–80.
- Wei, Y., Jin, J., Jiang, S., Ning, S., Liu, L., 2018. Quantitative response of soybean development and yield to drought stress during different growth stages in the Huaibei Plain, China. *Agronomy* 8 (7), 97.
- Young, T.E., Meeley, R.B., Gallie, D.R., 2004. ACC synthase expression regulates leaf performance and drought tolerance in maize. *Plant J.* 40 (5), 813–825.
- Zahedi, S.M., Karimi, M., Venditti, A., Zahra, N., Siddique, K.H., Farooq, M., 2025. Plant adaptation to drought stress: the role of anatomical and morphological characteristics in maintaining the water status. *J. Soil Sci. Plant Nutr.* 25 (1), 409–427.
- Zhao, Z., He, K., Feng, Z., Li, Y., Chang, L., Zhang, X., Xu, S., Liu, J., Xue, J., 2019. Evaluation of yield-based low nitrogen tolerance indices for screening maize (*Zea mays* L.) inbred lines. *Agronomy* 9 (5), 240.