

Research Article

Root System Architecture and Physiological Characteristics of Soybean (*Glycine max* L.) Seedlings in Response to PEG6000-Simulated Drought Stress

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The production of soybean is restricted in sub-Saharan Africa by several stress conditions, including drought because its production is exclusively rain-fed. Identifying drought resistant varieties is of paramount importance. Thus, the objectives of this work were to (i) evaluate the effect of polyethylene glycol 6000 (PEG6000) on soybean at the seedling stage, (ii) determine the root system architecture and physiological characters to water deficit stress, and (iii) establish the correlation among the quantitative variables responsible for drought tolerance in soybean varieties. Twenty soybean accessions (G1 to G20) were subjected to 10% PEG6000 concentration at seedling stages under a controlled environment using a randomized complete block design with 3 replicates. Vegetative growth data were collected. Highly significant differences ($P < 0.0001$) of proline, carotenoid, chlorophyll a, and chlorophyll b contents were recorded among the 20 accessions in response to PEG application. G16 and G19 had the highest carotenoid, highest chlorophyll a, and chlorophyll b. The highest dry weight was observed in G16 and G10, while the number of leaves was recorded in G19 and G17. G4, G9, G10, and G13 demonstrated the highest dry weight. The PEG-simulated drought stress reduced the average root diameters and the number of lateral roots of all 20 accession plants. G1, G3, G4, G8, G9, and G15 had the longest roots than the control plants as a mechanism to withstand drought stress by seeking water in the deep. Number of leaves was significantly and positively correlated with shoot dry weight, root dry weight, and root diameter but was significantly and negatively correlated with canopy wilting. Proline content was significantly and positively correlated with carotenoid, chlorophyll content, chlorophyll a, and chlorophyll b. G10, G19, G9, G6, G16, G17, G20, G16, and G18 are the tolerant cultivars to drought stress on the basis of growth, physiological, and root system architecture.

1. Introduction

Soybean is an important crop worldwide due to its protein, oil, fatty acids, fibre, and nutrient content essential for health in humans and animals [1–3]. Processed soybean products include soy cheese, soy cake, soy milk, soy mustard, soy yogurt, and infant foods associated with high protein, which is cheaper when compared with fish and meat [4–6]. However, the production of soybean is limited in sub-Saharan Africa by numerous stresses, including drought because its production is exclusively rain-fed.

Drought conditions often reduce yield at multiple stages of development, including flowering, pollination, and the grain-filling stage [7–9]. The decline in yields due to drought calls for identification of drought tolerance crops to meet up with the demand of crops including soybeans in the global markets [10]. There are tolerant soybean varieties among the existing genotypes in the world gene banks. Exploiting the existing collection of soybean germplasm will help come up with soybeans resistant to drought conditions. Therefore, there is an imperative need to screen important crops such as soybean for tolerance to drought

stress so as to enhance its productivity and yields. Such results would substantially benefit many small farmers, whose life is challenged by constant food insecurity and hunger. IPCC [11, 12] reported that the poorest countries will suffer the most from the consequences of climate change because abiotic stress will reduce yields drastically in the most economically important crops in tropical and subtropical regions, and food production in Africa will severely be compromised. Three main mechanisms are employed by plants to deal with water deficit stress, and these include drought escape and avoidance and tolerance to drought and use various mechanisms to cope with drought stress [13]. In drought escape, plants shorten and complete their life cycle before the commencement of drought stress or complete the critical stages of its development before the onset of water deficit in the soil, which allows the plant to produce some yields instead of total crop failure. Avoidance of drought is the second mechanism, which involves reducing water loss from aerial parts by using cuticle wax or by having the ability to extract moisture from the soil efficiently and the plants continue to keep high water status during periods of stress. The third mechanism is tolerance to drought stress, during periods of drought stress plants continue to maintain turgor by delaying stomatal closure, keeping chloroplast volume, and reducing leaf wilting and plants continue metabolic reactions even at low water potential by synthesizing osmoprotectants, osmolytes, or compatible solutes or by protoplasmic tolerance [14]. This tolerance to internal water deficits allows to prolong the functioning of photosynthesis. Carbon products can then be used for both osmotic adjustment and root growth. Another consequence of maintaining carbon metabolism is a decrease in the frequency of photoinhibition episodes. At the cellular level, osmotic adjustment plays a key role in maintaining turgor at low leaf water potentials.

Methods including mannitol, sorbitol, polyethylene glycol (PEG), and water withholding have been used to impose drought stress in the field or greenhouse or laboratories to understand the mechanisms of plant tolerance to drought stress. PEG-simulated drought stress and water withholding have been the forefront methods of studying drought stress. PEG 6000 is a natural polymer, which is nontoxic and nonionic. PEG 6000 induces osmotic stress in plants by reducing water potential as it is observed during the shortage of rainfall [15, 16].

Some studies have used PEG to simulate drought stress in crops [17–22], while the stress study by water withholding is highly reported in the tropics [23, 24]. PEG induces drought in the plant root systems and causes the inaccessibility of available water in the soil to the growing plants. Thus, the present research was conducted to (i) evaluate the effect of PEG on soybean at the seedling stage, (ii) determine the root system architecture and physiological characters due to water deficit, and (iii) establish the correlation among the quantitative variables responsible for drought tolerance in soybean varieties.

2. Materials and Methods

2.1. Study Area and Plant Materials. This study was carried out in the greenhouse at Bowen University, Iwo, Osun State, Nigeria, located between 7°38' Nord and 4°11' East longitude with an altitude of 322 m above sea level.

A total of 20 soybean accessions used in the present study were obtained from the International Institute of Tropical Agriculture (IITA), Ibadan Nigeria (Table 1).

2.2. Experimental Design and Drought Conditions. The study was conducted from June to August 2022 in a randomized complete block design with 3 replicates and twenty soybean accessions. The growing medium for soybean production consisted of topsoil and sawdust in the ratio 2 : 1. Eleven (11) kg of the substrate were filled in each experimental bag. Two factors, namely, accession and PEG6000-simulated drought stress, were studied.

Twenty-one (21) days after sowing, drought treatments were imposed to soybean seedlings thinned to three plants per experimental bag. Two treatments were applied including control and simulation of drought with 10% polyethylene glycol 6000 (10% PEG-6000) for 14 days. 200 ml of 10% PEG was applied to designated seedlings, while 200 ml of water was applied to plants which served as control. Pesticides and fertilizers were not applied during the experiment.

2.3. Data Collection

2.3.1. The Growth Parameters. Measured were plant height (cm), number of leaves, leaf length (cm), leaf width (cm), and above ground wilting, which was recorded using a modified wilting scale of 0–5 [21, 25]. The leaf wilting was rated in stressed plants visually on a scale of 0 to 5. The wilting scale of 0 represented no wilting. 1 represented the unifoliate wilting. 2 represented the 1st wilting of trifoliate leaves. 3 represented the first 2 trifoliate, 4 represented the first 3 trifoliate leaves wilting together. 5 represented the whole plant wilting.

2.3.2. Root System Architecture. On the last day of the experiment, the uprooted, treated, and control plants were meticulously washed with tap water to get rid of debris and soil particles. Then, root length, number of lateral roots, and root diameter were measured as morphological root system architecture traits.

2.3.3. Biomass Yield. Biomass yield was measured after 24 hours of drying below and above ground samples in an oven set at 80°C.

2.3.4. Physiological Parameters. On the 14th day of drought imposition, fresh leaves were sampled for the measurement of carotenoid, chlorophyll a, chlorophyll b, and proline content. These photosynthetic pigments were evaluated using a modified Arnon [26] procedure. Carotenoid,

TABLE 1: Twenty (20) soybean accessions used.

S/N	Accession
G1	TGm-50
G2	TGm-95
G3	TGm-112
4	TGm-263
5	TGm-422
6	TGm-665
7	TGm-946
8	TGm-951
9	TGm-1326
10	TGm-1678
11	TGm-3972
12	TGm-4004
13	TGm-4400
14	TGm-4414
15	TGm-4144
16	TGm-4015
17	TGm-4499
18	TGm-4500
19	TGm-4502
20	TGm-4022

chlorophyll a, and chlorophyll b contents were computed using the equation of Porra [27], whereas proline was assessed using Bates et al. [28] procedure. For the measurement of chlorophyll content using SPAD meter, the average of triplicate readings was recorded on the fully expanded leaflet of each plant. SPAD meter is based on the ratio of transmission of near infrared to red wavelengths. The measurement of chlorophyll content using SPAD meter is a nondestructive method unlike in the case of chlorophyll a and chlorophyll b where the destructive method was applied for the determination of chlorophyll a and b using the equations.

2.4. Statistical Analysis. The morphological, root architecture, biomass yield, and physiological data collected were subjected to analyses of variance by using the R statistical package version R-4.0.5. Fischer's least significant difference (F-LSD) was used for the separation of means at a probability level of 5%. PCA was run using the FactoMineR and factoextra packages, and Pearson correlation was done using corr. Functions in R. A hierarchical cluster analysis was performed using the cluster factoextra package in R.

3. Results

3.1. ANOVA for Genotypes, Peg-Simulated Drought, and Genotypes by Environment Interaction. The Analysis of the Variance indicated that PEG6000-simulated drought stress by variety interaction had a very highly significant effect ($P < 0.01$) on root length ($P \leq 0.05$), carotenoid ($P \leq 0.0001$), proline ($P \leq 0.0001$), and chlorophyll a root diameter (Table 2).

The ANOVA (Table 2) also revealed that variety had a significant effect on root architecture such as the number of lateral roots ($P \leq 0.05$), root diameter ($P \leq 0.0001$), and root length ($P \leq 0.05$); physiological parameters such as

carotenoid ($P \leq 0.0001$), proline ($P \leq 0.0001$), chlorophyll a ($P \leq 0.0001$), and chlorophyll b ($P \leq 0.0001$); and morphological traits including plant height ($P \leq 0.0001$) and the number of leaves ($P \leq 0.0001$).

PEG6000-simulated drought stress had each highly significant ($P \leq 0.0001$) influence on all the morphological root architecture, physiological, and morphological characters except for the number of leaves where a significant difference ($P \leq 0.05$) was observed. There was no significant difference in chlorophyll content (Table 2).

3.2. Effect of PEG-Simulated Drought Stress on Canopy Wilting. Canopy wilting was recorded in all accessions, although the degree of wilting differed from one accession to the other (Figure 1). The highest wilting was observed in G3 and G10, while the lowest canopy wilting was in G19. No wilting was observed in well-watered plants.

3.3. Effect of PEG-Simulated Drought Stress on Morphological Traits. The analysis of variance of the number of leaves revealed a highly significant difference among varieties ($P < 0.0022$) (Table 3). Also, there were significant differences in leaf length among the varieties. The average leaf length ranged from 8 to 12 under drought stress, and the highest value was observed in G20. The highest value of plant height was recorded in G6 and G14, and highly significant differences were observed among varieties subjected to simulated drought stress. There was a significant difference between the varieties for leaf width under simulated drought stress. PEG-simulated drought stress inhibited the morphological traits when compared to the control plants.

3.4. Effect of PEG-Simulated Drought Stress on the Morphological Architecture of Root System and Biomass Yield. There were significant differences ($P < 0.001$) in the average root length among soybean accessions under normal and drought stress simulated by PEG 6000 (Table 4 and Figure 2). The results showed that there was a reduction of root length in G5, G6, G10, G11, G12, G13, G14, G17, G18, and G19, while G1, G3, G4, G8, G9 and G15 after PEG-6000 simulated drought stress had their root lengths higher than the control plants as a mechanism to seek for water in soil. The PEG-simulated drought stress reduced the average root diameters and the number of lateral roots of all 20 accession plants. Under normal watering conditions and drought stress, there was a significant difference for the above dry matter yield, and the highest value was recorded in G11, G16, and G8 under normal conditions and G16 and G10 under PEG-simulated drought stress (Table 4).

The morphological architecture of root system under (A) control, (B) drought, and (C) PEG-simulated drought stress are revealed in Figure 2. We observed that the main roots of seedling under drought (B) and PEG-simulated drought stress (C) are longer than those of under well-watered plants. The PEG-simulated drought stress has caused the change in the root architecture by extending the main roots and reducing the number of lateral roots.

TABLE 2: Significance of soybean root system architecture, morphological, and physiological variables under levels of drought stress.

Variables	Varieties	PEG	Varieties * PEG	Min	Max
Number of lateral roots	170*	12100***	151 ^{ns}	4	65
Root diameter	0.36***	4.92***	0.17*	1.02	2.99
Above dry weight	0.064 ^{ns}	1.4301***	0.06 ^{ns}	0.1	1.2
Below dry weight	0.021 ^{ns}	1.160***	0.032 ^{ns}	0.1	1.2
Root length	455*	3681***	694***	27	142
Carotenoid	137.1***	544.4***	105.7***	2.759	30.83
Proline	0.273***	2.33***	0.34***	0.05	1.25
Chl a	50.23***	189.37***	47.46***	3.519	22.63
Chl b	18.98***	82.44***	13.83***	0.083	10.662
Leaf length	2.83 ^{ns}	65.86***	2.10 ^{ns}	4.60	15.40
Leaf width	2.47 ^{ns}	52.37***	1.24 ^{ns}	5.50	11.60
No. of leaves	0.699***	1.408*	0.25 ^{ns}	2	5
Plant height	16.84***	59.03***	4.26 ^{ns}	8	20.60
Chl content	18.136 ^{ns}	1.65 ^{ns}	14.91 ^{ns}	21.50	39.10
Canopy wilting	0.37 ^{ns}	104.53***	0.29 ^{ns}	0	5

Values specify the mean square. *, **, and *** show significance levels at $P < 0.05$, $P < 0.01$, and $P < 0.001$, respectively. ^{ns}Indicates nonsignificant. Treatments with common letters for genotype, treatment, and genotype \times treatment interaction are not statistically significant at $P < 0.05$.

Figure 3 shows the below dry biomass yield of the 20 soybean accessions. The PEG-simulated drought stress decreased below dry biomass yield in all accessions. The best below dry weight was recorded with G10 followed by G9, G4, G6, and G13.

3.5. Effect of PEG-Simulated Drought Stress on Physiological Traits. Highly significant differences ($P < 0.0001$) were recorded with accessions regarding carotenoid, chlorophyll a, and chlorophyll b contents in their responses to PEG application (Table 5). There was an increase in carotenoids, chlorophyll a, and chlorophyll B for some varieties and a decrease for others. Increase in carotenoid and chlorophyll a was recorded in G1, G2, G6, G7, G8, G10, G11, G12, G14, G15, G16, G17, and G19 under drought stress. G16 and G19 drought recorded the highest values of 29.46 and 30.13, respectively, for carotenoid; 19.96 and 21.46, respectively, for chlorophyll a; and 10.64 and 9.38, respectively, for chlorophyll b (Table 5). There was no significant difference in the average chlorophyll contents under well-watered plants and water-stressed plants.

Figure 4 presents the concentration of proline under normal and water deficit conditions. A highly significant ($P < 0.0001$) increase in proline concentration was observed in the leaves of all genotypes except for G1 and G3 under water stress. G6, G2, G16, and G7 accumulated more proline than others.

3.6. Principal Component Analysis. The relationships among physiological, morphological, root architecture system, and biomass yield variables were established using the principal component analysis (Figures 5–7). A total of 10 axes were generated explaining the total variability (Figure 5). The first two dimensions accounted for 48.14% of the variance culminating in a moderate contribution to the total variation (Figures 5 and 6). But the combination of the first five dimensions explained up to 80.54% of the variance in the data, resulting in a very strong contribution to the total variation

(Figures 5–7). Carotenoid, proline, chlorophyll content, chlorophyll a and chlorophyll b, root diameter, and shoot dry weight significantly contributed to the formation of Dim1, whereas plant height, leaf length, leaf width, root length, and root diameter were significantly and positively correlated with Dim2, and the number of lateral roots was significantly and negatively correlated with Dim2. Canopy wilting and the number of lateral roots showed a significant and positive correlation with Dim3, but the number of leaves was significantly and negatively correlated with Dim3. Root dry weight, shoot dry weight, and the number of leaves were significantly and positively correlated with Dim4 (Figures 5 and 6).

In Figure 6, both variables and accessions were loaded at the same time indicating the relationship among traits and the distances between genotypes. The nearer the vector to the axis, the stronger the correlation. TGm-112, TGm-263, TGm-422, TGm-3972, TGm-4015, and TGm-4502 contributed significantly to Dim1, while TGm-50, TGm-95, TGm-665, TGm-946, TGm-4500, and TGm-4022 significantly contributed to Dim2. TGm-4004 and TGm-4144 are the two significant accessions for the formation of Dim3. Dim4 consisted of TGm-1678, TGm-4400, and TGm-4414, while only TGm-951 significantly contributed to Dim5 (Figure 6).

TGm-1678 (G10) did well for number of leaves, leaf length, root length, above dry weight, below dry weight, chlorophyll a, and chlorophyll b. TGm-4502 (G19) did well for no. of leaves, leaf width, root diameter, carotenoids, chlorophyll a, chlorophyll b, and chlorophyll content. TGm-1326 (G9) did well for leaf length, leaf width, root length, and root diameter. TGm-665 (G6) did well for leaf length, leaf width, root diameter, carotenoids, chlorophyll a, and chlorophyll content. TGm-4015 (G16) did well for root length, number of lateral roots, above dry weight, carotenoids, chlorophyll a, chlorophyll b, and chlorophyll content. TGm-4499 (G17) did well for no. of leaves, above dry weight, carotenoids, chlorophyll a, and chlorophyll b. TGm-4022 (G20) did well for leaf width, plant height, root length,

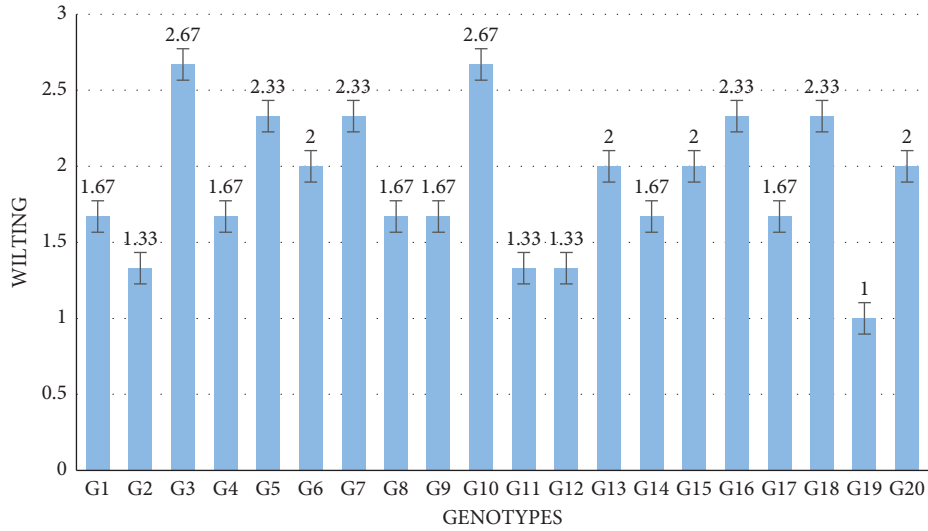


FIGURE 1: Canopy wilting of leaves of 20 accessions under PEG6000-simulated drought stress.

TABLE 3: Effect of PEG6000-simulated drought stress on morphological parameters.

VAR	No of leaves		Leaf length		Leaf width		Plant height	
	Control	PEG6000	Control	PEG6000	Control	PEG6000	Control	PEG6000
G1	3.33bcd	3.33bc	10.83	10.58ab	10.07	9.6ab	14.93abcde	12.33cdef
G2	3cd	3cd	9.8d	8.32d	8.77	8bc	13.47cde	12.1def
G3	3.33bcd	3cd	11.13	10.33abc	9.4	8.95abc	14.8abcde	12.03def
G4	3.66abc	3.33bc	8.733	8.47cd	8.56	7.5c	16.13abcd	12.73cde
G5	3cd	2.66d	10.76	10.17abcd	9.06	8.77abc	13.6cde	12.13def
G6	3.33bcd	3cd	11.03	11.43ab	9.36	9.9a	16.7abc	17.2a
G7	3.33bcd	2.66d	10.73	9.72bcd	9.03	8.32abc	12.33e	10.13f
G8	3.66abc	3.33bc	11.06	10.83ab	9.7	9.17ab	14.56bcde	14.4bcd
G9	3.33bcd	3.33bc	10.66	10.47ab	10.4	9.72a	17.23ab	14.63bc
G10	4ab	3.66ab	11.43	10.8ab	10.06	9.82a	17.96a	14.63bc
G11	4.33a	3cd	12.3	10.78ab	10.16	9.15ab	14.6bcde	11.36ef
G12	2.66d	3.33bc	11.53	11.03ab	9.73	9.13ab	12.93de	11.73ef
G13	3.33bcd	3.33bc	11.3	10.35abc	10.36	9.6ab	12.63e	13.3cde
G14	3cd	3cd	10.33	9.72bcd	8.93	8.37abc	15.53abcde	18.03a
G15	3cd	3cd	10.5	10.72ab	9.33	9.08abc	13.2de	12.46cdef
G16	3.66abc	3cd	12.13	10.8ab	9.96	8.93abc	15.26abcde	11.6ef
G17	4ab	4a	11.43	9.95abcd	10.46	9.05abc	13.96bcde	11.96ef
G18	3cd	2.66d	11.13	10.52ab	8.9	8.38abc	13.36de	11.46ef
G19	3.66abc	4a	12.76	11.13ab	11.16	9.6ab	15.33abcde	13.1cde
G20	3.33bcd	3cd	12.96	11.67a	10.23	9.38ab	14.86abcde	16.36ab
LSD	0.87	0.63	2.20	1.92	1.74	2.03	3.31	2.37
CV	15.50	12.09	11.97	16.09	10.90	15.65	13.63	10.84
Pr (<F)	0.05	0.0022**	0.108 ^{ns}	1.21e - 06***	0.248 ^{ns}	9.71e - 08***	0.015**	2.89e - 07***

Values specify mean. *, **, and ***, show significance levels at $P < 0.05$, $P < 0.01$, and $P < 0.001$, respectively. nsIndicates nonsignificant. Treatments with common letters for genotype are not statistically significant at $P < 0.05$.

root diameter, carotenoids, and chlorophyll b. G6 did well for leaf length and root length. TGm-951 (G8) did well for leaf length, root length, and chlorophyll a.

3.7. Relationship between Variable Analysis. Proline content was significantly and positively correlated with the following characters: carotenoid ($r = 0.67$, $P < 0/001$), chlorophyll content ($r = 0.74$, $P < 0.0002$), chlorophyll a ($r = 0.69$, $P < 0.0008$), and chlorophyll b ($r = 0.57$, $P < 0.009$)

(Figure 8). There was significant and positive correlation between carotenoid and chlorophyll a ($r = 0.97$, $P < 2.64E - 12$), carotenoid and chlorophyll b ($r = 0.85$, $P < 0.2.10E - 06$), and carotenoid and chlorophyll content ($r = 0.45$, $P < 0.046$). Shoot dry weight was positively correlated with proline ($r = 0.216$, $P < 0.360$), root diameter, root length, root dry weight, number of lateral root, leaf length, chlorophyll content, chlorophyll a, chlorophyll b, positively and significantly correlated with ($r = 0.448$, $P < 0.04$). Plant height was significantly and positively

TABLE 4: Effect of PEG6000-simulated drought stress on the morphological architecture of root systems and dry matter yield.

VAR	Root length		Root diameter		Number of lateral roots		Above dry weight	
	Control	PEG6000	Control	PEG6000	Control	PEG6000	Control	PEG6000
G1	52.2jkl	52.77cd	2.81ab	2.50ab	39defg	27.5bc	0.4e	0.5bc
G2	45.7m	54.77cd	2.30efg	2.09bcd	49.67abcd	36.67a	0.57cde	0.6abc
G3	54ijk	57.77cd	2.37cdef	2.02cde	41bcdefg	30.67bc	0.73abcde	0.63abc
G4	38.67n	43.67cd	2.29efg	2.05de	47.67abcde	35.17ab	0.5de	0.42c
G5	71b	47.77cd	2.72abcd	2.18bcd	38.33defgh	28.83bc	0.57cde	0.42c
G6	78.3a	56.57cd	2.96a	2.66a	54abc	37a	0.77abcde	0.63abc
G7	49.3l	45.1cd	2.013fg	1.87de	25h	23.5cd	0.7abcde	0.57abc
G8	60ef	93.33a	2.25efg	2.09bcd	37defgh	27.17bc	0.9abc	0.7ab
G9	51.67kl	55.33cd	2.17efg	2.09bcd	56a	37.33a	0.67abcde	0.58abc
G10	64.2d	53.67cd	2.47bcde	2.26abcd	48.67abcd	36.67a	0.67abcde	0.7ab
G11	55hij	44.67cd	2.83ab	2.36abc	40.33cdefg	29.5bc	1a	0.68abc
G12	73.87b	50.67cd	1.91gh	1.85de	46.33abcdef	30bc	0.87abc	0.72ab
G13	58.33fg	42.77cd	2.11efg	1.97cde	25h	22.67cd	0.93ab	0.72ab
G14	60.73ef	50.77cd	2.77abc	2.27abcd	32.67fgh	23.83cd	0.83abcd	0.63abc
G15	56.2ghi	64.33cd	2.11efg	1.94cde	54.33ab	38.83a	0.67abcde	0.57abc
G16	62de	61.83cd	2.03fg	1.97cde	34.33efgh	29.83bc	0.93ab	0.8a
G17	58fgh	37.23d	1.95fgh	1.89de	38defgh	28.83bc	0.6bcde	0.65abc
G18	67.3c	46.9cd	1.55h	1.58e	31.67gh	31.17ab	0.5de	0.5bc
G19	52.5jk	38.5d	2.35def	2.26abcd	32.67fgh	23.17cd	0.8abcd	0.65abc
G20	58.7fg	87.67ab	2.11efg	2.15bcd	31gh	23.5cd	0.77abcd	0.57abc
LSD	3.00	25.77	0.42	0.445	13.83	16.91	0.35	0.277
CV	3.11		11.03	18.49	20.85	49.04	29.33	39.26
Pr (<F)	0.000307***	0.00718**	9.68e - 07***	1.39e - 10***	0.000229***	< 2e - 16***	0.0439*	5.96e - 08***

Values specify mean. *, **, and ***, show significance levels at $P < 0.05$, $P < 0.01$, and $P < 0.001$, respectively. ^{ns}Indicates nonsignificant. Treatments with common letters for genotype are not statistically significant at $P < 0.05$.



FIGURE 2: Morphological architecture of the root system under (a) control, (b) drought, and (c) PEG-simulated drought stress.

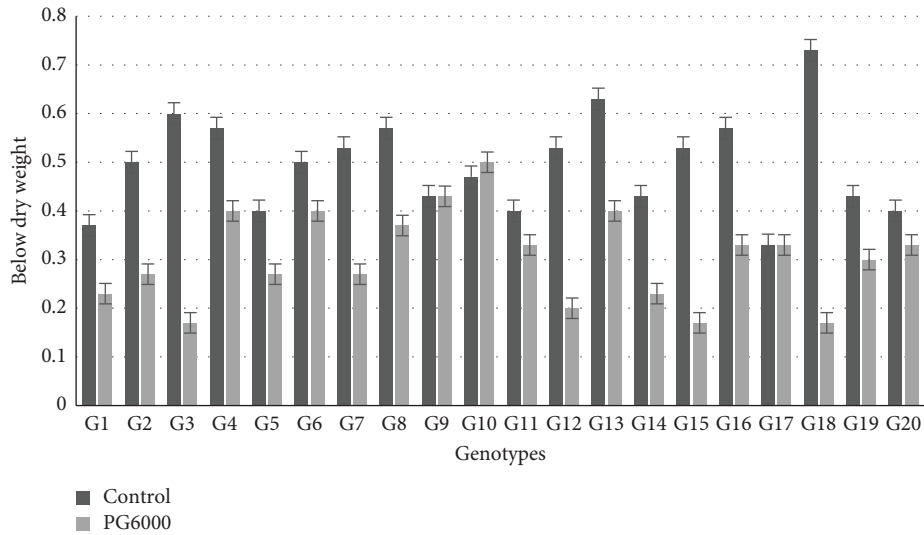


FIGURE 3: Effect of drought stress on the below dry weight of soybean genotypes.

TABLE 5: Effect of PEG6000-simulated drought stress on physiological characters.

GEN	Carotenoid ($\mu\text{g}\cdot\text{mL}^{-1}$)		Chla ($\mu\text{g}\cdot\text{mL}^{-1}$)		Chlb ($\mu\text{g}\cdot\text{mL}^{-1}$)		Chl content ($\mu\text{mol}\cdot\text{m}^{-2}$)	
	Control	PEG6000	Control	PEG6000	Control	PEG6000	Control	PEG6000
G1	10.71efg	11.671i	10.79fgh	12.33f	0.4219hi	0.6409kl	29.46	25.26
G2	4.86h	18.53e	6.129j	16.51d	1.304efghi	2.614hi	32.56	33.53
G3	12.23de	11.95hi	12.79ef	12.16f	0.5544ghi	0.2459l	30.8	25.16
G4	18.23c	6.60k	15.12cd	8.56h	3.308bc	1.997i	28.33	26.86
G5	10.67efg	7.34k	12.14fg	9.63g	1.504efgh	2.365i	33.53	26.6
G6	18.07c	22.49d	14.62de	19.14bc	3.646b	3.573fg	31.3	32.56
G7	2.76h	21.50d	3.52k	18.70c	0.7912ghi	3.020gh	30.66	29.4
G8	4.33h	6.96k	6.17j	8.15h	1.912def	1.227jk	28.16	32.83
G9	13.80d	9.96j	11.61fg	10.26g	2.323cde	0.300l	28.43	28.16
G10	9.74fg	18.24e	8.96hi	15.43e	0.8442fghi	3.059gh	26.93	25.96
G11	11.83def	13.15gh	11.66fg	12.47f	0.2164i	0.7912jkl	27.33	26.56
G12	9.10g	16.46f	10.27gh	14.54e	1.20fghi	2.073i	32.43	30.86
G13	20.65b	14.21g	17.78ab	12.31f	3.07bc	2.032i	27.03	28.7
G14	8.89g	18.02e	7.01ij	12.66f	1.153fghi	1.997i	28.5	29.5
G15	16.20c	18.49e	16.20bcd	15.40e	0.703ghi	3.700f	30.2	28.2
G16	12.68de	29.46a	11.14fg	19.96b	1.643efg	10.64a	28.26	32.76
G17	16.20c	26.17b	16.92abc	19.95b	1.432efgh	6.567d	27.9	29.56
G18	17.48c	14.11g	15.37cd	12.87f	2.833bcd	1.352j	29.9	30.73
G19	12.56de	30.13a	11.43fg	21.46a	1.212fghi	9.380b	26.63	31.83
G20	23.92a	23.83c	18.48a	16.45d	5.747a	7.785c	29.4	28
Grand mean	12.78	16.96	51.46	46.23	5.514	27.289	11.99	21.06
LSD	2.23	1.21	2.15	0.89	1.09	0.63	6.09	5.94
CV	10.55	3.39	10.92	3.77	36.93	11.21	12.55	12.34
Pr (<F)	< 2e - 16***	< 2e - 16***	< 2e - 16***	< 2e - 16***	4.78e - 11***	< 2e - 16***	0.607 ^{ns}	0.099 ^{ns}

Values specify mean. *, **, and *** show significance levels at $P < 0.05$, $P < 0.01$, and $P < 0.001$, respectively. ^{ns}Indicates nonsignificant. Treatments with common letters for genotype are not statistically significant at $P < 0.05$.

correlated with leaf width ($r=0.50$, $P < 0.024$), root dry weight ($r=0.46$, $P < 0.040$), and root diameter ($r=0.51$, $P < 0.021$). The number of leaves was significantly and positively correlated with shoot dry weight ($r=0.45$, $P < 0.047$), root dry weight ($r=0.47$, $P < 0.038$), and root

diameter ($r=0.51$, $P < 0.021$) but was significantly and negatively correlated with canopy wilting ($r=-0.45$, $P < 0.048$). Leaf width was significantly and positively correlated with leaf length ($r=0.81$, $P < 1.19E - 05$), root length ($r=0.45$, $P < 0.047$), and root diameter ($r=0.50$, $P < 0.024$).

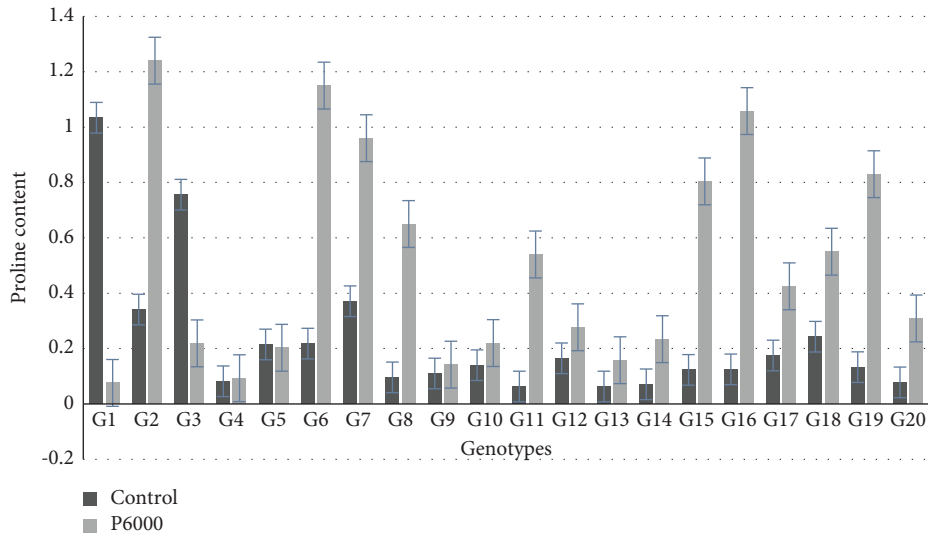


FIGURE 4: Effect of drought stress on proline concentration ($\mu\text{g}\cdot\text{mL}^{-1}\cdot\text{FW}$) of soybean genotypes.

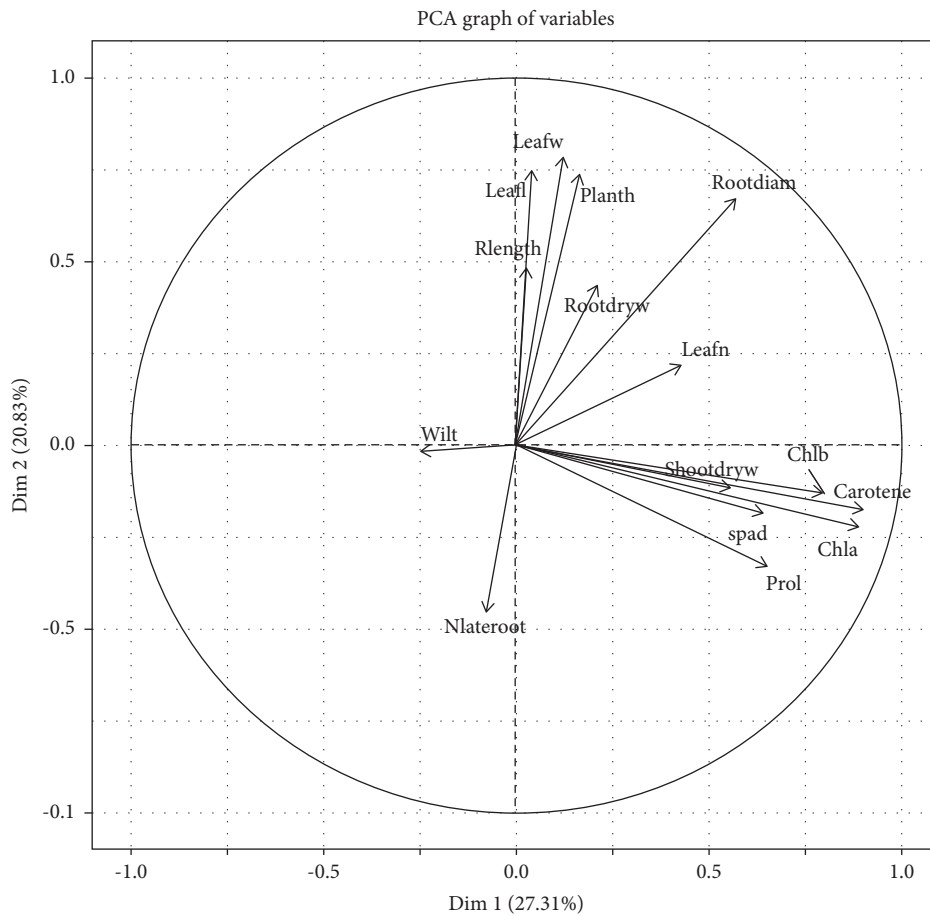


FIGURE 5: Correlation between physiological root system architecture and morphological traits per axis. Prol = proline, leafn = leaf number, planth = plant height, leafw = leaf width, chla = chlorophyll a, chlb = chlorophyll b, carotene = carotenoids, Rlength = root length, shootdryw = shoot dry weight, rootdryw = root dry weight, Nlateroot = number of lateral root, and Rootdiam = root diameter.

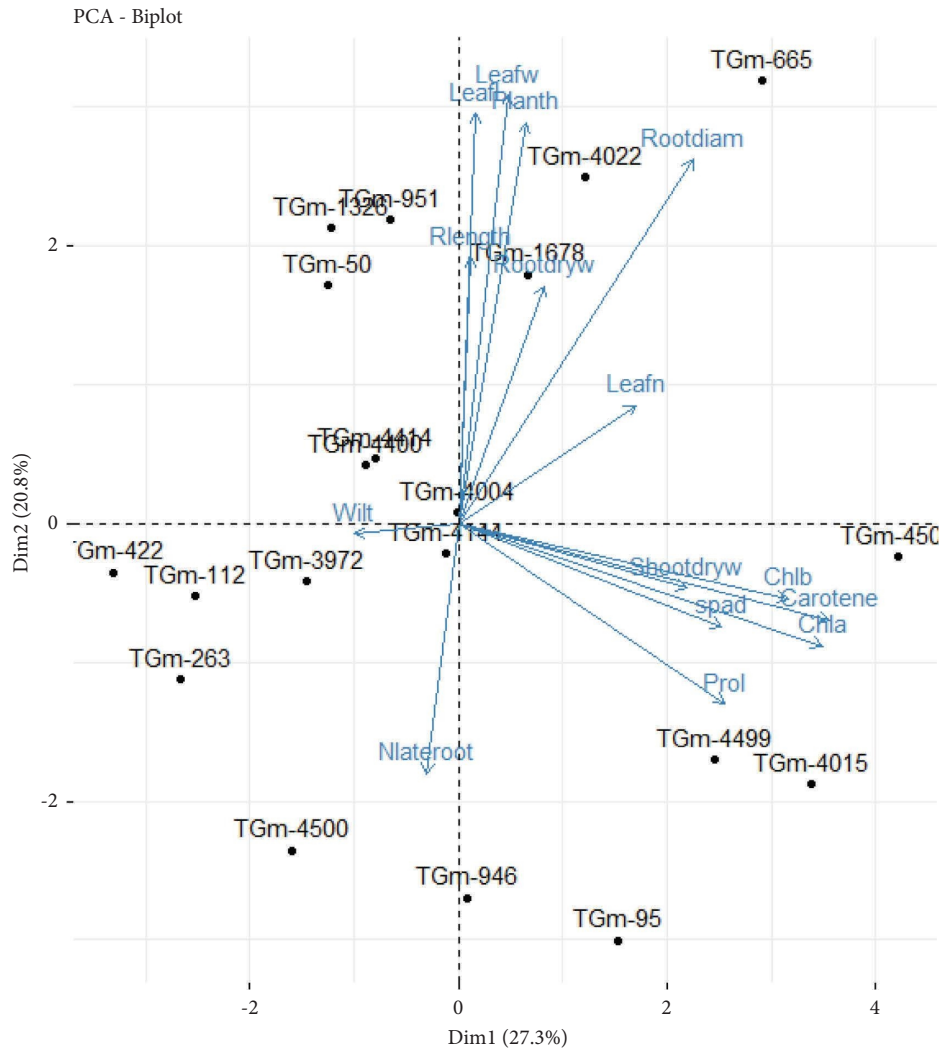


FIGURE 6: Correlation between and amongst genotypes and physiological, root system architecture, and morphological traits under drought stress. Prol = proline, leafn = leaf number, planth = plant height, leafw = leaf width, chla = chlorophyll a, chlb = chlorophyll b, carotene = carotenoids, Rlength = root length, shootdryw = shoot dry weight, rootdryw = root dry weight, Nlaterroot = number of lateral root, and Rootdiam = root diameter.

4. Discussion

Soybean is one of the main sources of protein and vegetable oil contributing to the world economy, and its production is faced with the challenges of change in climate, especially being sensitive to drought stress. It is crucial to develop resistant soybean varieties to drought for food security reasons. The first step toward the realization of this is the screening and then the identification of resistant genotypes, which can be used as planting materials or gene reservoirs for soybean breeding programs. There are many mechanisms that plants put in place to lessen the effect of drought stress. Yan et al. [29] suggested that there should be more screening of soybean germplasm for vital characters such as high heritability and stable traits such as drought tolerance in soybean. Thus, this study has been carried out to identify drought-tolerant soybean varieties because it is really a concern for

farmers and the world at large to have the yields of soybean compete with other crops such as cereals.

The synthesis of chlorophyll is fundamental to light interception for efficient photosynthesis [30, 31]. Modifying pigmentation could lead to a decrease in photosynthesis and an increase in photooxidation and photoinhibition resulting in more rapid yellowing and death of the leaves [32, 33]. This study showed an increase in carotenoids, chlorophyll a, chlorophyll b, and free proline contents in most of the genotypes, while there was a significant decline in other genotypes under PEG-simulated drought stress. The same observations were made by Wijewardana et al. [34] who reported that carotenoids increased linearly in soybean plants with declining soil moisture content. Basal et al. [22] observed that chlorophyll a and chlorophyll b declined as PEG concentration increased at all stages, but the reduction was insignificant at vegetative stages and significant at reproductive stages. Similar results were obtained by Zhang

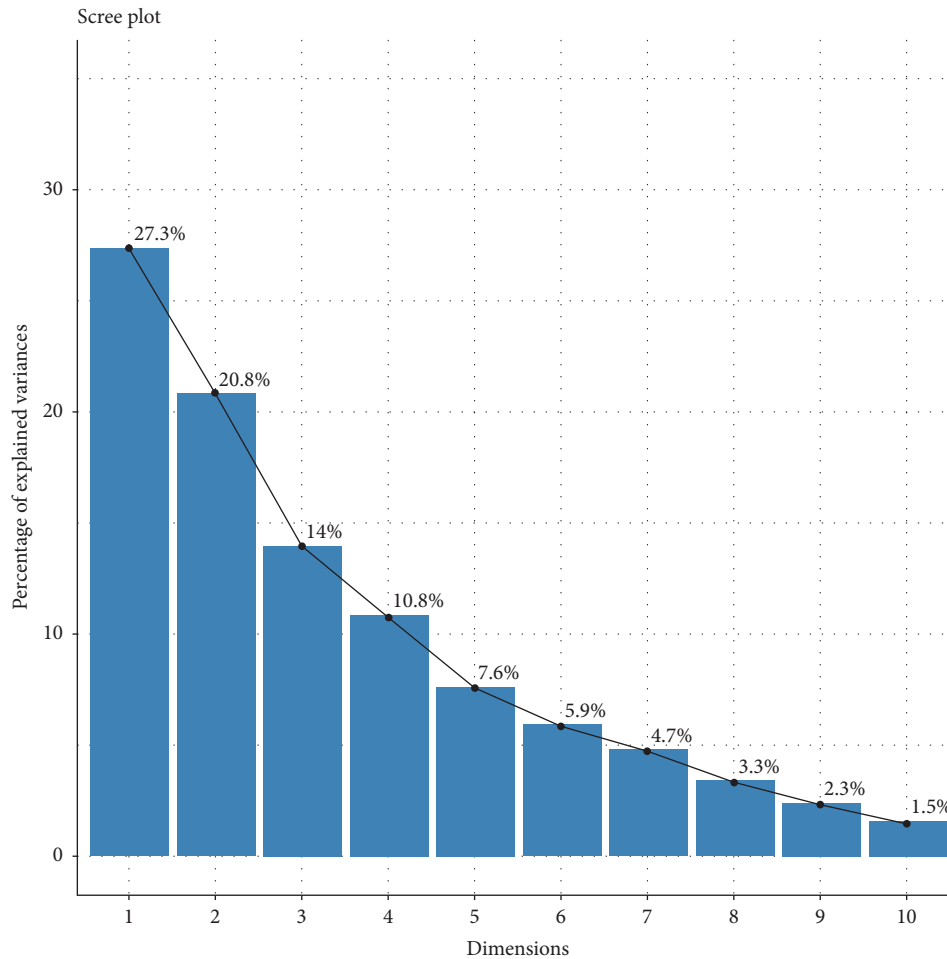


FIGURE 7: Contribution percentage of explained variances of 10 dimensions of physiological, root system architecture, and morphological traits under drought stress.

et al. [35] who observed that soybean plants under water deficit stress showed lower chlorophyll a, chlorophyll b, and carotenoid content. The concentration of free proline decreased in G1 and G3, whereas there was a sharp and remarkable increase in free proline for the other 18 genotypes under water stress. Our results corroborate those obtained with cowpea [36] and beans [37]. The chlorophyll contents were not affected by drought stress after 10 days of drought imposition. Gunes et al. [38] and Silva et al. [40], who worked, respectively, on chickpea cultivars and sugarcane genotypes, considered chlorophyll content, relative water content, and ascorbic acid as secondary indicators for choosing drought-tolerant genotypes and yield as a key characteristic for measuring drought tolerance under water deficit.

PEG-simulated drought stress in this study triggered reductions in plant height, leaf length, leaf width, and the number of leaves. The decline was more pronounced in plant height, leaf length, and leaf width than in the number of leaves. The reasons could be due to their sensitivity to lack of water and the significant decline in metabolic reactions for food production and nutrient transportation, which are essential for plant growth. Similar results were obtained by

previous studies [29, 40]. Yan et al. [29] reported that plant height and shoot growth of soybean varieties declined with PEG-simulated drought stress imposition. The above-mentioned results are consistent with those of [41–43] who showed that significant differences in variety and drought stress for plant height, leaf area index, shoot biomass, root biomass, and total root length.

Morphological root system architecture plays a crucial role in the productivity and survival of plants subjected to either flooding or drought conditions [29, 44, 45]. The root diameters and the number of lateral roots of all 20 accession plants subjected to drought stress were smaller than those of control plants. This is consistent with the results of [29], who recorded a decrease in root diameter but an increase in the number of lateral roots with the PEG-simulated drought stress. We observed variability in root length produced by genotypes as their response to drought stress. The root lengths of G5, G6, G10, G11, G12, G13, G14, G17, G18, and G19 were reduced due to PEG-simulated drought, while under the same drought conditions, G1, G3, G4, G8, G9, and G15 responded differently by increasing their root lengths as a mechanism of tolerance to drought stress. Battisti and Sentelhas [46] reported that soybean varieties that can

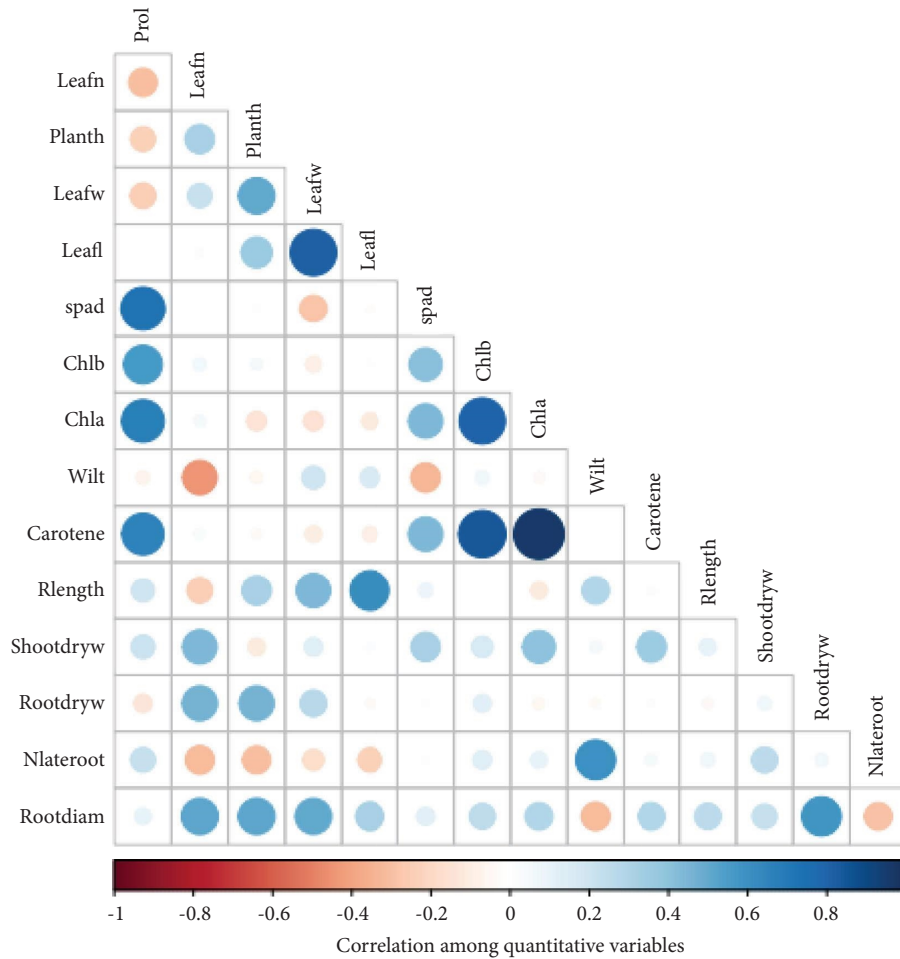


FIGURE 8: Correlations among physiological, root system architecture, and morphological traits under drought stress; Pearson's rank correlation matrix indicating the correlation among variables of soybean accessions grown under water deficit stress. Prol = proline, leafn = leaf number, planth = plant height, leafw = leaf width, chla = chlorophyll a, chlb = chlorophyll b, carotene = carotenoids, Rlength = root length, shootdryw = shoot dry weight, rootdryw = root dry weight, Nlateroot = number of lateral root, and Rootdiam = root diameter.

produce deeper root depth under drought stress are to be considered as resilient to climate change because they can elongate their taproots to search for water deep in the soil.

The PEG-simulated drought stress decreased below dry biomass yield in almost all the accessions. Thus, the below dry biomass yield is more affected than the above dry matter yield by drought stress. This is consistent with the results of [21, 29, 47–49] who reported that under water stress, there is a notable decrease in shoot dry biomass and root dry biomass at growth phases.

Plants of the accessions wilted as their response to PEG-simulated drought stress, and the degree of canopy wilting differed from one accession to the other. The same results about soybean wilting under drought stress were observed by previous researchers [21, 29, 50].

Positive and significant correlations among physiological, morphological, and root architecture were observed in this study as shown in the Results section. Liu et al. [51] in their study observed significant and positive correlations between drought resistance and dry weight, root volume, total length, and the number of lateral roots in soybean. We observed the same results

with Yan et al. [29] who demonstrated that the wilting index was intensely and negatively correlated with root length, root surface area, number of lateral roots, and root volume.

Under PEG-simulated drought stress, the principle components analysis is used to detect the quantitative characters that are positively and significantly correlated with each component of the plot. In the biplot, the correlation between cultivars and quantitative traits was depicted at each principal component. In addition, PC is for grouping of inherently related cultivars into the same class based on the performance under drought stress. G10 recorded the best no. of leaves, leaf length, root length, above dry weight, below dry weight, chlorophyll a, and chlorophyll b. G19 had the best no. of leaves, leaf width, root diameter, carotenoids, chlorophyll a, chlorophyll b, and chlorophyll content. G9 performed well for leaf length, leaf width, root length, and root diameter. G6 performed well for leaf length, leaf width, root diameter, carotenoids, chlorophyll a, and chlorophyll content. G16 revealed better root length, number of lateral root, above dry weight, carotenoids, chlorophyll a, chlorophyll b, and

chlorophyll content. G17 TGM-4499 recorded better no. of leaves, above dry weight, carotenoids, chlorophyll a, and chlorophyll b. G20 TGM-4022 demonstrated better leaf width, plant height, root length, root diameter, carotenoids, and chlorophyll b. G8 showed better leaf length, root length, and chlorophyll a. The soybean cultivars with best physiological, above, and below ground as described above were the best and tolerant cultivars under PEG-simulated drought stress.

5. Conclusion

This study shows remarkable changes in physiology, root system architecture, and dry matter yields of the 20 accessions under PEG-simulated drought stress. G16 and G19 recorded the highest carotenoid, highest chlorophyll a, and chlorophyll b. The highest dry weight was observed with G16 and G10, while the number of leaves was recorded in G19 and G17. G4, G9, G10, and G13 demonstrated the highest dry weight. For chlorophyll content, G6, G8, G16, G19, and G18 were the best. G10, G19, G9, G6, G16, G17, G20, G16, and G18 are the tolerant cultivars to drought stress on the basis of growth, physiological, and root system architecture.

Data Availability

The data used to support the findings of this study are available from the corresponding author upon reasonable request.

Conflicts of Interest

The authors declare that they have no conflicts of interest.

References

- [1] L. Hao, Y. Wang, J. Zhang et al., "Coronatine enhances drought tolerance via improving antioxidative capacity to maintaining higher photosynthetic performance in soybean," *Plant Science*, vol. 210, pp. 1–9, 2013.
- [2] R. N. Mutava, S. J. K. Prince, N. H. Syed et al., "Understanding abiotic stress tolerance mechanisms in soybean: a comparative evaluation of soybean response to drought and flooding stress," *Plant Physiology and Biochemistry*, vol. 86, pp. 109–120, 2015.
- [3] P. Cerezini, B. Kuwano, M. B. dos Santos, F. Terassi, M. Hungria, and M. A. Nogueira, "Strategies to promote early nodulation in soybean under drought," *Field Crops Research*, vol. 196, pp. 160–167, 2016.
- [4] P. Kumar, M. K. Chatli, N. Mehta, P. Singh, O. P. Malav, and A. K. Verma, "Meat analogues: health promising sustainable meat substitutes," *Critical Reviews in Food Science and Nutrition*, vol. 57, no. 5, pp. 923–932, 2017.
- [5] K. Zaheer and M. Humayoun Akhtar, "An updated review of dietary isoflavones: nutrition, processing, bioavailability and impacts on human health," *Critical Reviews in Food Science and Nutrition*, vol. 57, no. 6, pp. 1280–1293, 2017.
- [6] G. Rizzo and L. Baroni, "Soy, soy foods and their role in vegetarian diets," *Nutrients*, vol. 10, no. 1, p. 43, 2018.
- [7] R. Dolferus, X. Ji, and R. A. Richards, "Abiotic stress and control of grain number in cereals," *Plant Science*, vol. 181, no. 4, pp. 331–341, 2011.
- [8] R. Dolferus, N. Powell, X. Ji et al., "The physiology of reproductive-stage abiotic stress tolerance in cereals," in *Molecular Stress Physiology of Plants*, G. R. Rout and A. B. Das, Eds., pp. 193–216, Springer, New Delhi, India, 2013.
- [9] N. Senapati, P. Stratonovitch, M. J. Paul, and M. A. Semenov, "Drought tolerance during reproductive development is important for increasing wheat yield potential under climate change in Europe," *Journal of Experimental Botany*, vol. 70, no. 9, pp. 2549–2560, 2018.
- [10] R. V. Martin, R. Washington, and T. E. Downing, "Seasonal maize forecasting for South Africa and Zimbabwe derived from an agro-climatological model," *Journal of Applied Meteorology*, vol. 39, no. 9, pp. 1473–1479, 2000.
- [11] Ipcc, *Climate Change 2001: The Scientific Basis. Contribution of Working Group I to the Third Assessment Report of the Intergovernmental Panel on Climate Change*, J. T. Houghton, Y. Ding, D. J. Griggs et al., Eds., Ipcc, Cambridge, UK, 2001.
- [12] Ipcc, *Climate Change 2007: The Physical Science Basis. Contribution of Working Group I to the Fourth Assessment Report of the Intergovernmental Panel on Climate Change*, S. Solomon, D. Qin, M. Manning et al., Eds., Cambridge University Press, Cambridge, UK, 2007.
- [13] N. C. Turner, G. C. Wright, and K. H. M. Siddique, "Adaptation of grain legumes (pulses) to water limited environments," *Advances in Agronomy*, vol. 71, pp. 193–123, 2001.
- [14] H. T. Nguyen, R. C. Babu, and A. Blum, "Breeding for drought resistance in rice: physiology and molecular genetics considerations," *Crop Science*, vol. 37, no. 5, pp. 1426–1434, 1997.
- [15] A. Muscolo, M. Sidari, U. Anastasi, C. Santonoceto, and A. Maggio, "Effect of PEG-induced drought stress on seed germination of four lentil genotypes," *Journal of Plant Interactions*, vol. 9, no. 1, pp. 354–363, 2014.
- [16] V. Ishola Esan, T. Ayanniyin Ayanbamiji, J. Omoyemi Adeyemo, and S. Oluwafemi, "Effect of drought on seed germination and early seedling of tomato genotypes using polyethylene glycol 6000," *International Journal of Sciences*, vol. 4, no. 02, pp. 36–43, 2018.
- [17] D. W. Lawlor, "Absorption of polyethylene glycols by plants and their effects on plant growth," *New Phytologist*, vol. 69, no. 2, pp. 501–513, 1970.
- [18] M. Hamayun, E. Sohn, S. A. Khan, Z. K. Shinwari, A. L. Khan, and I. Lee, "Silicon alleviates the adverse effects of salinity and drought stress on growth and endogenous plant growth hormones of soybean (*Glycine max L.*)," *Pakistan Journal of Botany*, vol. 42, no. 3, pp. 1713–1722, 2010.
- [19] S. Hajhashemi and A. Sofo, "The effect of polyethylene glycol-induced drought stress on photosynthesis, carbohydrates and cell membrane in *Stevia rebaudiana* grown in greenhouse," *Acta Physiologiae Plantarum*, vol. 40, no. 8, p. 142, 2018.
- [20] C. Zhang, S. Shi, B. Wang, and J. Zhao, "Physiological and biochemical changes in different drought-tolerant alfalfa (*Medicago sativa L.*) varieties under PEG-induced drought stress," *Acta Physiologiae Plantarum*, vol. 40, no. 2, p. 25, 2018.
- [21] G. Wang, Q. Zhou, M. He, X. Zhong, and G. Tang, "Wilting index and root morphological characteristics used as drought-tolerance variety selection at the seedling stage in soybean (*Glycine max L.*)," *Plant Growth Regulation*, vol. 92, no. 1, pp. 29–42, 2020.
- [22] O. Basal, A. Szabó, and S. Veres, "Physiology of soybean as affected by PEG-induced drought stress," *Current Plant Biology*, vol. 22, pp. 100135–100138, 2020.
- [23] I. Alam, S. A. Sharmin, K. H. Kim, J. K. Yang, M. S. Choi, and B. H. Lee, "Proteome analysis of soybean roots subjected to

- short-term drought stress," *Plant and Soil*, vol. 333, no. 1-2, pp. 491-505, 2010.
- [24] C. V. Ha, D. T. Le, R. Nishiyama et al., "Characterization of the newly developed soybean cultivar DT2008 in relation to the model variety W82 reveals a new genetic resource for comparative and functional genomics for improved drought tolerance," *BioMed Research International*, vol. 2013, Article ID 759657, 8 pages, 2013.
- [25] S. M. Pathan, J. D. Lee, D. A. Sleper et al., "Two soybean plant introductions display slow leaf wilting and reduced yield loss under drought," *Journal of Agronomy and Crop Science*, vol. 200, no. 3, pp. 231-236, 2014.
- [26] D. I. Arnon, "Copper enzymes in isolated chloroplasts. Polyphenoloxidase in *Beta vulgaris*," *Plant Physiology*, vol. 24, no. 1, pp. 1-15, 1949.
- [27] R. J. Porra, "The chequered history of the development and use of simultaneous equations for the accurate determination of chlorophylls a and b," *Photosynthesis Research*, vol. 73, no. 1-3, pp. 149-156, 2002.
- [28] L. S. Bates, R. P. Waldren, and I. D. Teare, "Rapid determination of free proline for water-stress studies," *Plant and Soil*, vol. 39, no. 1, pp. 205-207, 1973.
- [29] C. Yan, S. Song, W. Wang et al., "Screening diverse soybean genotypes for drought tolerance by membership function value based on multiple traits and drought-tolerant coefficient of yield," *BMC Plant Biology*, vol. 20, no. 321, pp. 1-15, 2020.
- [30] J. Bornman, T. Vogelmann, and G. Martin, "Measurement of chlorophyll fluorescence within leaves using a fibre-optic microprobe," *Plant, Cell and Environment*, vol. 14, no. 7, pp. 719-725, 1991.
- [31] L. L. Eggink, H. Park, and J. K. Hooper, "The role of chlorophyll b in photosynthesis: hypothesis," *BMC Plant Biology*, vol. 1, no. 2, pp. 2-7, 2001.
- [32] H. Upadhyaya, M. H. Khan, and S. K. Panda, "Hydrogen peroxide induces oxidative stress in detached leaves of *Oryza sativa* L.," *General and Applied Plant Physiology*, vol. 33, pp. 83-95, 2007.
- [33] H. Zgallai, K. Steppe, and R. Lemeur, "Photosynthetic, physiological and biochemical responses of tomato plants to polyethylene glycol-induced water deficit," *Journal of Integrative Plant Biology*, vol. 47, no. 12, pp. 1470-1478, 2005.
- [34] C. Wijewardana, F. A. Alsajri, J. T. Irby et al., "Physiological assessment of water deficit in soybean using midday leaf water potential and spectral features," *Journal of Plant Interactions*, vol. 14, no. 1, pp. 533-543, 2019.
- [35] J. Zhang, J. Liu, C. Yang, S. Du, and W. Yang, "Photosynthetic performance of soybean plants to water deficit under high and low light intensity," *South African Journal of Botany*, vol. 105, pp. 279-287, 2016.
- [36] V. Ezin, A. G. C. Tosse, I. B. Chabi, and A. Ahanchede, "Adaptation of cowpea (*vigna unguiculata* (L.) walp.) to water deficit during vegetative and reproductive phases using physiological and agronomic characters," *International Journal of Agronomy*, vol. 2021, Article ID 9665312, 12 pages, 2021.
- [37] M. L. Praba, J. E. Cairns, R. C. Babu, and H. R. Lafitte, "Identification of physiological traits underlying cultivar differences in drought tolerance in rice and wheat," *Journal of Agronomy and Crop Science*, vol. 195, no. 1, pp. 30-46, 2009.
- [38] A. Gunes, A. Inal, M. S. Adak, E. G. Bagci, N. Cicek, and F. Eraslan, "Effect of drought stress implemented at pre- or post-anthesis stage on some physiological parameters as screening criteria in chickpea cultivars," *Russian Journal of Plant Physiology*, vol. 55, no. 1, pp. 59-67, 2008.
- [39] M. A. Silva, J. L. Jifon, V. Sharma et al., "Use of physiological parameters in screening drought tolerance in sugarcane genotypes," *Sugar Technology*, vol. 13, no. 19, pp. 1-7, 2011.
- [40] Y. Ohashi, N. Nakayama, H. Saneoka, and K. Fujita, "Effects of drought stress on photosynthetic gas exchange, chlorophyll fluorescence and stem diameter of soybean plants," *Biologias Plantarum*, vol. 50, no. 1, pp. 138-141, 2006.
- [41] M. Zaifnejad, R. B. Clark, and C. Y. Sullivan, "Aluminum and water stress effects on growth and proline of sorghum," *Journal of Plant Physiology*, vol. 150, no. 3, pp. 338-344, 1997.
- [42] L. C. Purcell, J. T. Edwards, and K. R. Brye, "Soybean yield and biomass responses to cumulative transpiration: questioning widely held beliefs," *Field Crops Research*, vol. 101, no. 1, pp. 10-18, 2007.
- [43] D. Li, H. Liu, Y. Qiao et al., "Effects of elevated CO₂ on the growth, seed yield, and water use efficiency of soybean (*Glycine max* L. Merr.) under drought stress," *Agricultural Water Management*, vol. 129, pp. 105-112, 2013.
- [44] L. P. Manavalan, S. K. Guttikonda, V. T. Nguyen, J. G. Shannon, and H. T. Nguyen, "Evaluation of diverse soybean germplasm for root growth and architecture," *Plant and Soil*, vol. 330, no. 1-2, pp. 503-514, 2010.
- [45] S. Jogaiah, S. R. Govind, and L. S. P. Tran, "Systems biology-based approaches toward understanding drought tolerance in food crops," *Critical Reviews in Biotechnology*, vol. 33, no. 1, pp. 23-39, 2013.
- [46] R. Battisti and P. C. Sentelhas, "Improvement of soybean resilience to drought through deep root system in Brazil," *Agronomy Journal*, vol. 109, no. 4, pp. 1612-1622, 2017.
- [47] D. J. Sammons, D. B. Peters, and T. Hymowitz, "Screening soybeans for tolerance to moisture stress: a field procedure," *Field Crops Research*, vol. 3, pp. 321-335, 1980.
- [48] D. S. Harris, W. T. Schapaugh, and E. T. Kanemasu, "Genetic diversity in soybeans for leaf canopy temperature and the association of leaf canopy temperature and yield ¹," *Crop Science*, vol. 24, no. 5, pp. 839-842, 1984.
- [49] K. Jumrani and V. S. Bhatia, "Impact of combined stress of high temperature and water deficit on growth and seed yield of soybean," *Physiology and Molecular Biology of Plants*, vol. 24, no. 1, pp. 37-50, 2018.
- [50] X. Liu, "Drought," in *Research on Tolerance to Stresses in Chinese Soybean*, H. M. Lam, R. Chang, G. Shao, and Z. Liu, Eds., China agricultural press, Beijing, China, 2009.
- [51] F. Liu, M. N. Andersen, S. E. Jacobsen, and C. R. Jensen, "Stomatal control and water use efficiency of soybean (*Glycine max* L. Merr.) during progressive soil drying," *Environmental and Experimental Botany*, vol. 54, no. 1, pp. 33-40, 2005.