Screening of Bangladesh Tea (*Camellia sinensis* (L.) Kuntze) Genotypes for Drought Tolerance

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Abstract. Drought stress is a major abiotic factor limiting tea (*Camellia sinensis* (L.) Kuntze) productivity worldwide, particularly in Bangladesh, where climatic variability is increasing. This study aimed to screen a diverse collection of tea genotypes for drought tolerance under nursery and field conditions to identify the promising one(s) suitable for cultivation in drought-prone areas of the country. Five tea genotypes (A/17/7, Mz/39, Sh/9/65, B/8/93 and A/8/8) along with a control variety (BT2) were evaluated based on morphological, physiological, and biochemical parameters such as plant height, root length, root volume, vertical length of root, dry matter, relative water content (RWC), proline accumulation, total polyphenol content, reducing sugar, etc. Four treatments, *viz.* T_0 (maintaining 36% soil moisture content) as a control with three drought treatments: T_1 (maintaining 30% soil moisture content), T_2 (maintaining 24% soil moisture content), and T_3 (maintaining 18% soil moisture content), were used for assessing drought tolerance both under nursery and field conditions. Clustering analysis indicated clear variations among the genotypes, where B/8/93 was distinguished as the most drought-tolerant and A/8/8 as the most sensitive. Mz/39 exhibited wide adaptability under field conditions, while the remaining cultivars (BT2, Sh/9/65, and A/17/7) were grouped as having intermediate tolerance. The study provides a foundation for further genetic improvement of drought resilience in tea, contributing to the long-term sustainability of the Bangladesh tea industry.

Keywords: Tea, Drought, Morpho-physiological, Water Relation, Biochemical Traits.

1 Introduction

Tea (*Camellia sinensis* (L.) Kuntze is a globally important beverage crop cultivated across tropical and subtropical regions, with significant economic, social, and environmental value (Zohora & Arefin, 2022). In Bangladesh, tea cultivation has a long history,

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dating back to the British colonial period, and continues to be a key sector in the agricultural economy. Currently, tea is grown primarily in the Sylhet, Moulvibazar, Chattogram hill tracts, and northern regions, covering over 168 tea estates and supporting the livelihoods of more than 300,000 people directly and indirectly (Arefin et al., 2024). Bangladesh is both a producer and consumer of tea, and the demand for higher yield with better quality is increasing day by day to meet domestic needs and export targets (Arefin et al., 2020).

However, tea cultivation is highly sensitive to environmental factors, particularly water availability. Drought stress has emerged as a critical constraint to tea production, especially in recent years due to the growing impact of climate change (Das et al., 2021). Irregular rainfall, extended dry seasons, and rising temperatures have contributed to increased frequency and intensity of droughts in many tea-growing areas of Bangladesh. Drought affects tea plants at multiple levels, including reduced leaf growth, lower photosynthetic activity, impaired nutrient uptake, and oxidative damage, ultimately leading to reduced yield and quality (Hossain et al., 2017). This situation poses a serious threat to the sustainability of tea cultivation in Bangladesh, where irrigation infrastructure is limited.

Genotypic variation plays a crucial role in how plants respond to drought (Maleki et al., 2021). Identifying and selecting drought-tolerant germplasm is essential for developing resilient tea cultivars that can maintain productivity under water-limited conditions. Various physiological, morphological, and biochemical traits, such as relative water content (RWC), leaf wilting, chlorophyll content, stomatal conductance, proline accumulation, and antioxidant enzyme activity, have been widely used as indicators for drought tolerance in tea (Langaroudi et al., 2023) and other crops (Saini et al., 2021). While some progress has been made globally in screening tea germplasm for drought tolerance, there remains a significant knowledge gap in the context of Bangladesh. Most tea cultivars in the country have not been systematically evaluated for their drought response, limiting the ability of breeders and growers to make proper decisions about cultivation in drought prone areas.

In this context, the present study was conducted to assess the drought tolerance of six tea germplasms collected from different tea estates of Bangladesh. The objectives of this study were (1) to compare the drought response of six selected tea germplasms under stress conditions; (2) to identify drought-tolerant test clones based on a combination of morpho-physiological and biochemical traits under both nursey and field conditions. The findings of current investigation will provide valuable insights for developing sustainable tea production systems in Bangladesh to minimize the adverse effects of climate variability on this crucial crop.

2 Materials and Methods

The experiment was conducted using five test clones (A/17/7, Mz/39, Sh/9/65, B/8/93, and A/8/8), previously selected from the seedling sections of various tea estates based on specific selection parameters (Arefin et al., 2024), along with the widely cultivated clone BT2 as the control, to compare the performance of the test clones. The whole research program was carried out under both nursery and field conditions at Botany Division, Bangladesh Tea Research Institute, Sreemangal, Moulvibazar during the period from 2014 to 2019. To evaluate drought tolerance, four soil moisture regimes were maintained as treatments. The control (T_0) was kept at 36% soil moisture content, while three drought levels were imposed as T_1 (30% soil moisture), T_2 (24% soil moisture), and T_3 (18% soil moisture). The moisture levels were carefully monitored and maintained throughout the experimental period to ensure consistent drought stress conditions by creating 'Rainout Shelter' (Fig. 1 and Fig. 2).

2.1 Data Collection Parameters under Nursery Condition

Under nursery condition, the experiment was set up using plastic bag of 30 cm height and 10 cm diameter, each filled with a uniform soil–cow dung–sand mixture (2:1:1 v/v) and planted with healthy, uniform tea saplings of each genotype by following complete block design (CRD) with five replications. After a 30-day establishment period under well-watered conditions, drought stress was imposed and maintained by monitoring soil moisture with a portable soil moisture meter. Soil moisture levels were recorded daily at 20 cm depth, and irrigation was regulated to maintain the required stress levels (Fig. 1). The following parameters regarding morphological, physiological and water relation traits were collected during drought assessment under nursery conditions after 30, 60, 120, 150 and 180 days of drought imposition (DDI):

1. Number of roots per plant: At harvest, the plants were carefully uprooted from their bags to avoid root damage. The roots were then meticulously washed under running tap water to remove all adhering soil particles. Excess water was blotted with paper towels. Then root-related parameters were collected, such as the number of roots, which were counted numerically.
2. Total root length: Primary roots of each genotype were separated by above mentioned method, and total length were calculated in centimetres (cm).
3. Root dry weight: Root dry weight was measured after oven drying of the root at 80±2°C for 72 hours.
4. Shoot dry weight: Shoot dry weight was measured after oven drying of the shoot at 80±2°C for 72 hours.
5. Vertical depth of root: Roots were collected from each sample, and the longest primary roots were identified. Then the lengths were measured in centimetres (cm) as the vertical depth of roots.
6. Plant height: The height was measured from the soil surface to stem apex in centimetres (cm).
7. Leaf area: It was computed by the 'Leaf Area Measurement Machine' named LI-3100 Area Meter (LI-COR Inc., USA) in centimetre square (cm^2).
8. Total dry matter: Total dry matter per plant (g) was calculated by the addition of the dry weight of the shoot and root.
9. Chlorophyll stability index (CSI): CSI (expressed as %) was calculated by the standard method (Kaloyereas, 1958).

10. Relative water content (RWC): RWC (expressed as %) of leaf was determined by the turgid weight and dry weight (Barrs & Weatherley, 1962).
11. Proline content: Proline content of leaf (expressed as µ mol g^{-1} fresh weight) was determined by the sulfosalicylic acid and ninhydrin reagents (Bates et al., 1973).

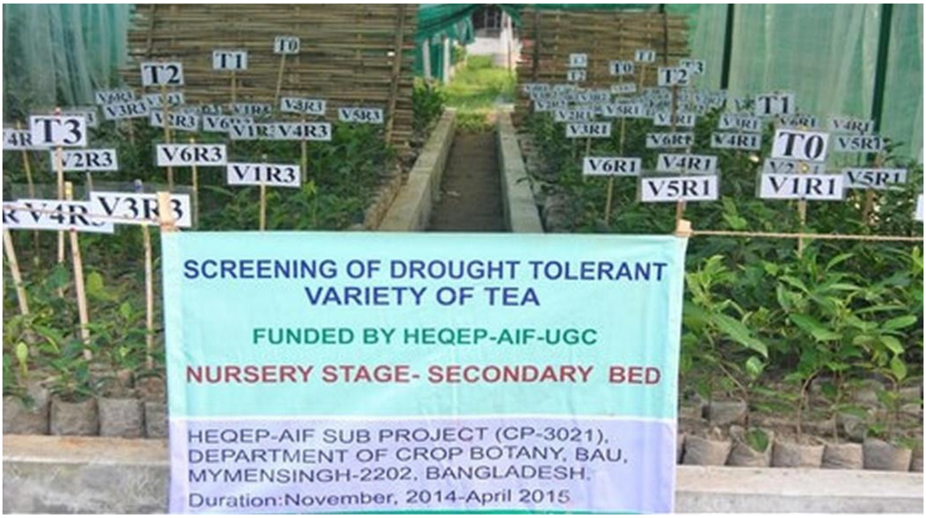

Fig. 1. Screening of drought-tolerant germplasm at the nursery stage in the 'Rainout shelter'.

2.2 Data Collection Parameters under Field Condition

The field experiment was conducted using 18 months old healthy and uniform tea saplings of each genotype, which were planted in randomized complete block designated plots with five replications. To separate each replication and prevent lateral movement of soil moisture, drains were constructed between the replications. After six months of establishment, decentering pruning was done at 20 cm plant height during the monsoon. Once the plants reached the tipping height of 45 cm, drought treatments were imposed. A rainout shelter was installed over the treatment plots (T_0, T_1, T_2 and T_3) to prevent external moisture interference during the drought period (Fig. 2). Drought conditions were maintained continuously for 30 days, and soil moisture status was strictly observed and regulated by partial irrigation, which was monitored by a neutron probe moisture meter. After 60 days of drought imposition (60 DDI), a total of 9 parameters about morphological, physiological, biochemical, and water relation traits were collected for the evaluation of drought tolerance under field conditions:

1. Root volume: For root collection, a large pit was carefully dug around the selected plant to expose the root system. The plant, along with the adhering soil, was gently uplifted to minimize root breakage. Roots were then separated by carefully washing off the soil under running water. After cleaning, the roots were blotted dry using a soft towel. The water displacement method was adapted to measure root volume in cm^3 (cc).

2. Plant height: A uniform tipping height of 45 cm was maintained for each plant before the imposition of drought. Treatment-wise plant height was measured after 60 DDI from the soil surface to the shoot apex and expressed in centimetres (cm).
3. Base diameter: Base diameter (cm) was measured at the collar region of the plants, *i.e.* one centimetre above soil.
4. Shoot length: Pluckable shoots above the tipping height (45 cm) were collected and average length was measured in centimetres (cm).
5. Number of branches: The number of branches from the main stem was measured on a numerical scale.
6. Green leaf per plant: Pluckable shoots, *i.e.* green leaf of each plant above the tipping height (45 cm) were collected and weighed on a weighing scale (g).
7. Total polyphenol content: It was expressed as per cent gallic acid equivalents (ppm as % GAE) (Sarwar et al., 2023).
8. Catechin: It was expressed as per cent catechin equivalents (ppm as % CE) (Swain & Hillis, 1959).
9. Reducing Sugar: Reducing sugar (expressed as ppm) was calculated by the standard protocol (Hawkins, 1929).

Fig. 2. Screening of drought-tolerant germplasm under field conditions in the ‘Rainout shelter’.

2.3 Statistical Analysis

The collected data on various characteristics under study were statistically analyzed to find out the statistical significance of the experimental results. The means were calculated, and the analysis of variance for all the characters was performed using the statistical package SPSS. The mean differences were evaluated by Duncan's New Multiple Range Test. The radar-plot graph and clustering heatmaps were made by the statistical software OriginPro 2025.

3 Results and Discussion

3.1 Effect of Drought on Tea Plants under Nursery Conditions

The effect of drought on morpho-physiological and water relations traits in six tea genotypes under nursery conditions is given below against different parameters with respective Tables:

Number of Roots Per Plant. Drought stress increases root number as an adaptive response to enhance water absorption from deeper soil. The clonal response on the number of roots to drought conditions appeared significant compared to the control condition. After 180 days of drought imposition (DDI), test clone B/8/93 showed maximum number of roots (20.83), followed by A/17/7 (18.07) and Mz/39 (17.88), which were statistically similar. Moderate number of roots were detected in Sh/9/65 (16.21) and standard BT2 (15.13), while A/8/8 showed minimum (15.38) number of roots (Table 1). Numerous lateral root formation was detected under drought condition and *PtrABR1* genes were responsible for increased root production (Sun et al., 2023). This result fairly agreed with the findings of the current experiment.

Table 1. Treatment-wise and genotypic effect on the number of roots under nursery conditions.

Treatments	30 DDI	60 DDI	90 DDI	120 DDI	150 DDI	180 DDI
T_0	10.09 a	10.20 b	11.23 c	13.49 c	14.78 d	16.46 d
T_1	9.95bc	10.57 a	11.63 b	13.92 b	16.27 a	17.96 a
T_2	9.97b	10.57 a	11.83 a	14.09 ab	15.85 b	17.42 b
T_3	9.90d	10.63 a	11.94 a	14.19 a	14.99 c	17.17 c
LSD $_{(0.05)}$	0.21	0.14	0.12	0.21	0.17	0.19
CV (%)	3.27	3.04	2.65	2.27	3.66	2.67
Genotypes						
A/17/7	10.32 b	11.22 a	12.19 b	14.51 b	16.38 b	18.07 b
Mz/39	10.26 b	11.23 a	12.65 a	14.58 b	16.18 b	17.88 b
Sh/9/65	9.517 c	10.08 bc	11.30 c	13.52 c	14.58 d	16.21 c
BT2	8.825 e	10.16 bc	11.27 c	11.51 e	12.91 e	15.13 e
B/8/93	11.85 a	12.1 b	12.26 b	17.49 a	17.73 a	20.83 a
A/8/8	9.108 d	9.992 c	10.27 d	11.93 d	15.06 c	15.38 d
LSD $_{(0.05)}$	0.26	0.17	0.15	0.25	0.21	0.23
CV (%)	3.27	3.04	2.65	2.27	3.66	2.67

T_0= 36% soil moisture content, T_1= 30% soil moisture content, T_2= 24% soil moisture content, T_3= 18% soil moisture content. DDI: Days of drought imposition. Mean values in the column having similar letter(s) do not differ significantly at the 5% level of probability.

Total Root Length. Tolerant plants accumulate more abscisic acid (ABA) in roots to increase the root length under low water stress condition (Aslam et al., 2022). This morphological adjustment helps plants explore a deeper soil to access limited moisture. It also increases survival chances under prolonged water deficit by optimizing water and nutrient uptake. Another study found that, the highest root length (15.69 cm) in CNS 352 cultivar out of 60 varieties was observed after drought imposition (Thiep et al., 2015). Similar observations are also observed in current study. Drought conditions significantly affected the total root length in all genotypes. Cultivar B/8/93 showed the longest roots (163.7 cm), followed by Mz/39 (139.6 cm) and A/17/7 (138.7 cm), whereas cultivar A/8/8 showed the minimum total root length (101.1 cm) at 180 DDI (Table 2).

Table 2. Treatment-wise and genotypic effect on total root length (cm) under nursery conditions.

Treatments	30 DDI	60 DDI	90 DDI	120 DDI	150 DDI	180 DDI
T_0	37.52 b	44.74 d	57.52 c	72.82 d	94.40 d	122.5 d
T_1	38.15 a	46.95 c	59.91 b	76.71 c	105.4 c	131.2 c
T_2	38.21 a	47.10 b	59.99 b	77.26 b	106.7 b	132.1 b
T_3	38.17 a	47.26 a	60.39 a	77.73 a	117.2 a	132.9 a
LSD $_{(0.05)}$	0.11	0.07	0.19	0.10	0.32	0.10
CV (%)	2.43	3.22	3.50	3.20	3.46	3.12
Genotypes						
A/17/7	38.22 c	47.40 c	61.80 c	78.00c	108.8 c	138.7 c
Mz/39	39.13 b	48.61 b	63.39 b	80.13b	106.6 d	139.6 b
Sh/9/65	35.09 d	42.25 e	54.96 e	74.70e	95.68 e	116.5 e
BT2	38.31 c	47.18 d	55.82 d	75.41d	114.2 b	118.5 d
B/8/93	44.15 a	56.18 a	78.89 a	96.11a	143.3 a	163.7 a
A/8/8	33.19 e	37.46 f	41.86 f	52.43f	66.94 f	101.0 f
LSD $_{(0.05)}$	0.13	0.08	0.24	0.12	0.40	0.12
CV (%)	2.43	3.22	3.50	3.20	3.46	3.12

T_0= 36% soil moisture content, T_1= 30% soil moisture content, T_2= 24% soil moisture content, T_3= 18% soil moisture content. DDI: Days of drought imposition. Mean values in the column having similar letter(s) do not differ significantly at the 5% level of probability.

Root Dry Weight. Significant differences in root dry weights were detected among the treatments. The root dry weight was found to be decreased with the increase of drought condition and duration. The rate of reduction was lower in B/8/93 (4.313 g), Mz/39 (3.980 g) and A/17/7 (3.959 g) than remaining cultivars BT2 (3.692 g), Sh/9/65 (3.576 g) and A/8/8 (2.743 g) at 180 DDI (Table 3). Several studies also mentioned reduced root dry weights after drought imposition. For example, the tolerant tea genotypes performed a lower reduction in root dry mass (1.92 g) compared to susceptible one (1.69 g) (Hossain et al., 2025). Under drought stress, plants produce more roots to explore a larger soil volume, but the overall root dry weight decreases due to reduced photosynthesis and limited photosynthates allocation. Instead of thick roots, plants form finer and lighter roots, which increases surface area for water uptake but lowers biomass accumulation. This adaptive response helps survival but compromises total root mass (Das et al., 2016).

Table 3. Treatment-wise and genotypic effect on root dry weight (g) under nursery conditions.

Treatments	30 DDI	60 DDI	90 DDI	120 DDI	150 DDI	180 DDI
T_0	1.273 ab	1.385 a	1.828 a	2.492 a	2.933 a	4.019 a
T_1	1.287 a	1.394 a	1.814 a	2.239 b	2.912 a	4.011 a
T_2	1.261 ab	1.362 ab	1.752 a	2.088 c	2.717 b	3.647 b
T_3	1.221 b	1.296 b	1.645 b	1.951 d	2.583 b	3.164 c
LSD (0.05)	0.056	0.070	0.089	0.112	0.14	0.188
CV (%)	6.50	7.86	7.53	7.66	7.60	7.59
Genotypes						
A/17/7	1.273 b	1.447 b	1.897 b	2.355 b	3.216 a	3.959 b
Mz/39	1.293 b	1.554 a	2.023 a	2.508 a	3.133 ab	3.980 b
Sh/9/65	0.9950 d	1.253 c	1.810 bc	2.342 bc	2.912 cd	3.576 c
BT2	1.133 c	1.303 c	1.766 c	2.158 d	2.744 d	3.692 c
B/8/93	1.850 a	1.627 a	1.888 b	2.208 cd	3.017 bc	4.313 a
A/8/8	1.020 d	0.9733 d	1.175 d	1.582 e	1.697 e	2.743 d
LSD (0.05)	0.068	0.086	0.11	0.13	0.17	0.23
CV (%)	6.50	7.86	7.53	7.66	7.60	7.59

T_0= 36% soil moisture content, T_1= 30% soil moisture content, T_2= 24% soil moisture content, T_3= 18% soil moisture content. DDI: Days of drought imposition. Mean values in the column having similar letter(s) do not differ significantly at the 5% level of probability.

Shoot Dry Weight. Under drought stress, shoot dry weight declines as plants face physiological and metabolic constraints. Shoot dry weight decreases due to reduced photosynthesis from stomatal closure, which limits carbohydrate supply for shoot growth. Low turgor pressure restricts leaf expansion, which is one of the prime causes of low yield during drought stress. Drought stress not only prevented the translocation and photosynthesis but also hindered the biochemical process of utilization of the dry matter in leaf and stem (Langaroudi et al., 2023). It was observed that shoot dry weight was affected by drought severely (Table 4). Under 36% soil moisture contents (T_0), tea plants produced the maximum shoot dry weights followed by 30% (T_1) and 24% (T_2) soil moisture content, while 18% soil moisture content (T_3) produced the lowest shoot dry weights. Interestingly, genotype Mz/39 (25.16 g) showed maximum shoot dry weight, followed by Sh/9/65 (23.27 g), while the lowest was detected in A/8/8 (20.34 g). The lesser decrease of shoot dry weight in the tolerant tea genotype (22.33 g) than in the weak variety (16.21 g) was also reported in another study (Hossain et al., 2025).

Table 4. Treatment-wise and genotypic effect on shoot dry weight (g) under nursery conditions.

Treatments	30 DDI	60 DDI	90 DDI	120 DDI	150 DDI	180 DDI
T_0	3.904 a	5.254 a	7.627 a	11.96 a	17.31 a	27.36 a
T_1	3.792 ab	5.028 ab	7.169 b	11.44 ab	15.62 b	24.94 b
T_2	3.706 b	4.843 bc	6.809 c	10.85 b	14.18 c	22.39 c
T_3	3.619 b	4.598 c	6.383 d	10.18 c	13.41 c	19.20 d
LSD (0.05)	0.18	0.27	0.34	0.640	0.83	1.36
CV (%)	7.22	8.29	7.44	8.60	8.21	8.67
Genotypes						
A/17/7	3.737 b	4.536 c	6.569 c	11.30 ab	15.04 ab	23.44 ab
Mz/39	3.833 b	5.154 a	7.077 ab	11.57 a	14.98 ab	25.16 a
Sh/9/65	3.288 c	4.614 bc	7.140 ab	11.15 ab	15.87 a	23.27 b
BT2	3.818 b	4.923 ab	7.384 a	10.86 ab	15.40 a	24.44 ab
B/8/93	4.093 a	5.090 a	6.912 bc	11.15 ab	15.43 a	24.20 ab
A/8/8	3.764 b	5.268 a	6.900 bc	10.63 b	14.05 b	20.34 c
LSD (0.05)	0.22	0.33	0.42	0.78	1.019	1.67
CV (%)	7.22	8.29	7.44	8.60	8.21	8.67

T_0= 36% soil moisture content, T_1= 30% soil moisture content, T_2= 24% soil moisture content, T_3= 18% soil moisture content. DDI: Days of drought imposition. Mean values in the column having similar letter(s) do not differ significantly at the 5% level of probability.

Vertical Depth of Root. Vertical depth of root significantly increased with the increase of drought stress compared to the control condition (Table 5). Genotype B/8/93 produced deepest (24.71 cm) roots, followed by Mz/39 (22.34 cm) and A/17/7 (22.06 cm), whereas A/8/8 showed minimum depth of root (16.61 cm) at 180 DDI. This result suggests that the vertical depth of root growth was found to be increased under drought in B/8/93, Mz/39, and A/17/7 than in the BT2, Sh/9/65 and A/8/8. Deeper roots (less than 30 cm) played a strong role in mitigating drought stress in sorghum plants (Chen et al., 2020). Hormonal regulation by abscisic acid and auxin, along with more carbohydrate allocation to roots and suppression of lateral growth, promote vertical root elongation for plant survival in drought stress (Chen et al., 2020).

Table 5. Treatment-wise and genotypic effect on vertical depth of root (cm) under nursery conditions.

Treatments	30 DDI	60 DDI	90 DDI	120 DDI	150 DDI	180 DDI
T_0	7.21 b	9.51 b	11.89 b	14.71 b	16.75 b	19.03 c
T_1	7.36 b	9.91 ab	12.49 ab	15.68 a	17.39 b	19.99 c
T_2	8.01 a	10.3 a	12.76 a	15.99 a	18.57 a	22.80 a
T_3	7.37 b	9.96 ab	12.25 ab	15.87 a	17.01 b	21.43 b
LSD (0.05)	0.36	0.45	0.61	0.94	0.80	1.26
CV (%)	7.33	6.88	7.45	9.10	6.92	9.05
Genotypes						
A/17/7	7.37 b	10.57 a	12.50 bc	17.32 a	19.50 a	22.06 b
Mz/39	7.35 b	10.56 a	13.05 b	17.26 a	16.62 c	22.34 b
Sh/9/65	7.92 a	10.18 ab	12.04 cd	14.03 b	16.05 c	19.19 c
BT2	7.22 b	9.925 b	11.63 de	14.07 b	19.76 a	19.96 c
B/8/93	7.95 a	10.43 ab	13.99 a	17.71 a	17.81 b	24.71 a
A/8/8	7.11 b	7.90 c	10.89 e	12.99 b	14.83 d	16.61 d
LSD (0.05)	0.45	0.56	0.75	1.16	0.99	1.54
CV (%)	7.33	6.88	7.45	9.10	6.92	9.05

T_0= 36% soil moisture content, T_1= 30% soil moisture content, T_2= 24% soil moisture content, T_3= 18% soil moisture content. DDI: Days of drought imposition. Mean values in the column having similar letter(s) do not differ significantly at the 5% level of probability.

Plant Height. Limited water during drought stress reduces cell turgor pressure, which restricts cell expansion and elongation, ultimately reducing plant height. Additionally, reduced photosynthesis lowers carbohydrate availability, so plants allocate more resources to root growth for efficient water absorption instead of shoot elongation, resulting in stunted height. In this experiment, significant variations were observed in the response of cultivars for plant height towards the degree of stress (Table 6). Plant height was found to be decreased with the increase of the intensity of drought compared to the control condition. B/8/93 showed the tallest plant (47.15 cm), followed by Mz/39 (43.76 cm), BT2 (43.39 cm), A/17/7 (42.64 cm), whereas A/8/8 showed the shortest plant (33.28 cm) at 180 DDI. The reduced plant height under drought stress conditions was also detected in several studies. For instance, the tolerant plant showed a stable and higher plant height (94.3 cm) than susceptible genotypes (77.86 cm) (Hossain et al., 2025).

Table 6. Treatment-wise and genotypic effect on plant height (cm) under nursery conditions.

Treatments	30 DDI	60 DDI	90 DDI	120 DDI	150 DDI	180 DDI
T_0	20.41 a	21.47 a	25.84 a	32.60 a	42.21 a	48.94 a
T_1	20.06 ab	20.70 b	24.56 b	30.87 b	35.39 b	44.59 b

T_2	19.70 b	19.97 c	23.22 c	28.76 c	35.79 b	39.69 c
T_3	19.00 c	19.35 d	21.55 d	26.73 d	33.66 b	34.27 d
LSD $_{(0.05)}$	0.43	0.39	0.28	0.55	2.78	0.39
CV (%)	3.27	2.88	2.82	2.79	11.28	2.42
Genotypes						
A/17/7	20.67 a	20.66bc	23.98 b	29.35 c	35.41 c	42.64 c
Mz/39	19.94 b	21.09ab	25.31 a	30.53 b	40.88 a	43.76 b
Sh/9/65	19.71 bc	19.71 d	23.55 c	29.54 c	37.55a-c	41.03 d
BT2	20.15 ab	20.55 c	23.94 b	30.51 b	36.37 bc	43.39 b
B/8/93	19.09 d	21.48 a	24.99 a	32.12 a	39.65 ab	47.15 a
A/8/8	19.20 cd	18.75 e	20.99 d	26.39 d	30.71 d	33.28 e
LSD $_{(0.05)}$	0.53	0.48	0.35	0.68	3.40	0.48
CV (%)	3.27	2.88	2.82	2.79	11.28	2.42

T_0= 36% soil moisture content, T_1= 30% soil moisture content, T_2= 24% soil moisture content, T_3= 18% soil moisture content. DDI: Days of drought imposition. Mean values in the column having similar letter(s) do not differ significantly at the 5% level of probability.

Leaf Area. During drought stress in tea, both leaf area and number decrease as cell expansion and initiation are inhibited by limited water availability. ABA accumulation accelerates leaf senescence, while resources are diverted to root growth instead of producing new leaves. This reduction in leaf growth also helps to minimize water loss through transpiration (Upadhyaya & Panda, 2013). Genotype B/8/93 showed maximum (676 cm^2) leaf area, followed by Mz/39 (668.6 cm^2), A/17/7 (663.4 cm^2), A/8/8 (660.9 cm^2) and BT2 (660.7 cm^2), whereas Sh/9/65 showed the minimum (650.4 cm^2) (Table 7). Another investigation reported lower leaf area indices in the condition of restricted water stress and found that the three years of drought in the area under study had the yield losses of 7.72%, 11.92%, and 12.52%, respectively (Das et al., 2021).

Table 7. Treatment-wise and genotypic effect on leaf area (cm^2) under nursery conditions.

Treatments	30 DDI	60 DDI	90 DDI	120 DDI	150 DDI	180 DDI
T_0	278.4 a	313.6 a	373.7 a	491.6 a	643.8 a	765.9 a
T_1	273.5 a	303.1 b	353.9 b	460.0 b	590.3 b	697.7 b
T_2	252.6 b	292.9 c	338.6 c	432.5 c	539.7 c	622.9 c
T_3	258.8 b	279.5 d	330.9 c	416.1 d	530.2 c	566.7 d
LSD $_{(0.05)}$	9.24	0.07	12.83	0.07	13.31	19.38
CV (%)	5.19	3.98	5.48	3.42	3.45	4.36
Genotypes						
A/17/7	274.4 ab	295.3 d	335.4 bc	441.7 e	584.5 b	663.4 c
Mz/39	268.4 bc	305.2 b	347.0 b	478.8 a	603.5 a	668.6 b

Sh/9/65	247.9 d	274.4 f	329.0 c	421.5 f	554.8 c	650.4 f
BT2	257.0 cd	286.7 e	340.6 bc	442.0 d	578.7 b	660.7 e
B/8/93	266.7 bc	301.8 c	366.6 a	470.6 b	582.4 b	676.0 a
A/8/8	280.7 a	320.1 a	377.1 a	445.9 c	552.1 c	660.9 d
LSD (0.05)	11.33	0.08	15.72	0.08	16.31	23.73
CV (%)	5.19	3.98	5.48	3.42	3.45	4.36

T_0= 36% soil moisture content, T_1= 30% soil moisture content, T_2= 24% soil moisture content, T_3= 18% soil moisture content. DDI: Days of drought imposition. Mean values in the column having similar letter(s) do not differ significantly at the 5% level of probability.

Total Dry Matter. Total dry matter production was observed to be highest with 36% of soil moisture content and showed significant differences from all other stress treatments (Table 8). Total dry matter production was reduced gradually with the increase of stress treatments. At 180 DDI, maximum total dry matter accumulation was observed in Mz/39 (29.19 g), which was followed by B/8/93 (28.67), while lower accumulation was detected in A/8/8 (23.36 g). Lower dry matter was also observed in limited water stress conditions, such as drought susceptible Kenyan tea clone S15/10, produced lower dry matter 14.7 t/ha, for a period of 34 months at mature stages (Ng'etich & Stephens, 2001).

Table 8. Treatment-wise and genotypic effect on total dry matter (g) under nursery conditions.

Treatments	30 DDI	60 DDI	90 DDI	120 DDI	150 DDI	180 DDI
T_0	5.288 a	6.724 a	9.610 a	14.41 a	20.51 a	31.29 a
T_1	5.205 a	6.492 b	9.065 b	13.67 b	18.81 b	29.32 b
T_2	4.941 b	6.230 c	8.592 c	12.91 c	17.15 c	26.28 c
T_3	4.846 b	5.933 d	8.109 d	12.15 d	15.83 d	22.66 d
LSD (0.05)	0.13	0.12	0.12	0.11	0.32	0.38
CV (%)	3.95	2.94	2.11	3.24	2.69	2.09
Genotypes						
A/17/7	5.089 c	5.947 d	8.745 b	13.64 b	18.25 c	27.69 d
Mz/39	5.289 b	6.862 a	9.088 a	14.07 a	18.04 c	29.19 a
Sh/9/65	4.333 e	5.804 d	9.106 a	13.47 c	19.29 a	27.23 d
BT2	4.950 cd	6.225 c	9.158 a	13.02 d	18.43 bc	28.19 c
B/8/93	5.923 a	6.832 a	8.999 a	13.36 c	18.68 b	28.67 b
A/8/8	4.836 d	6.399 b	7.967 c	12.15 e	15.76 d	23.36 e
LSD (0.05)	0.16	0.15	0.15	0.13	0.39	0.47
CV (%)	3.95	2.94	2.11	3.24	2.69	2.09

T_0= 36% soil moisture content, T_1= 30% soil moisture content, T_2= 24% soil moisture content, T_3= 18% soil moisture content. DDI: Days of drought imposition. Mean values in the column having similar letter(s) do not differ significantly at the 5% level of probability.

Chlorophyll Stability Index (CSI). The chlorophyll stability index (CSI) is a physiological parameter used to assess a plant's ability to maintain chlorophyll content under stress, with higher CSI values indicating efficient chlorophyll retention and thus better tolerance in plants. Many studies reported the reduction of CSI% in drought conditions. For example, a lower CSI% (37.4%) was noticed in drought condition than in the control (85.7%) in several wheat varieties (Saini et al., 2021). CSI values in all the varieties were found to be significantly decreased under different treatments except control (Table 9). Genotype B/8/93 significantly showed highest CSI % (82.9%), followed by A/17/7 (80.08%) and Mz/39 (79.72%), whereas A/8/8 showed lowest CSI% (64.53%). Current results are in line with previous studies.

Table 9. Treatment-wise and genotypic effect on chlorophyll stability index (CSI)%, relative water content (RWC)%, and proline contents (µ mol g^{-1} fresh weight) under nursery conditions.

Treatments	Chlorophyll stability index			Relative water content			Proline contents		
	120 DDI	150 DDI	180 DDI	120 DDI	150 DDI	180 DDI	120 DDI	150 DDI	180 DDI
T_0	85.18 a	88.12 a	90.24 a	83.35 a	82.07 a	81.23 a	0.6173 b	0.6173 b	0.6380 b
T_1	80.68 b	82.66 b	82.55 b	80.37 b	78.86 b	77.03 b	0.6428 a	0.6472 a	0.6756 a
T_2	76.75 c	77.04 c	72.71 c	77.91 c	75.62 c	71.87 c	0.6476 a	0.6511 a	0.6800 a
T_3	72.38 d	73.69 d	62.90 d	75.30 d	73.72 d	66.95 d	0.6517 a	0.6561 a	0.6833 a
LSD (0.05)	0.59	0.52	0.55	0.55	0.54	0.58	0.02	0.02	0.02
CV (%)	2.12	1.98	3.07	3.05	3.04	2.17	1.65	1.72	1.98
Genotypes									
A/17/7	80.98 b	82.37 b	80.08 b	79.80 a	78.11 b	75.13 b	0.6603 a	0.6628 a	0.6919 a
Mz/39	80.56 bc	82.89 b	79.72 b	80.26 a	78.99 a	75.21 b	0.6578 a	0.6622 a	0.6863 a
Sh/9/65	78.12 d	79.90 c	76.35 d	80.07 a	78.58 ab	74.84 b	0.6580 a	0.6621 a	0.6862 a
BT2	79.95 c	82.57 b	79.01 c	79.91 a	78.89 a	75.21 b	0.6583 a	0.6614 a	0.6864 a
B/8/93	81.97 a	83.73 a	82.90 a	80.17 a	78.38 ab	76.22 a	0.6679 a	0.6712 a	0.6996 a
A/8/8	70.90 e	70.80 d	64.53 e	75.19 b	72.44 c	69.01 c	0.5369 b	0.5380 b	0.5648 b
LSD (0.05)	0.72	0.64	0.68	0.68	0.66	0.71	0.02	0.02	0.02

CV (%)	2.12	1.98	3.07	3.05	3.04	2.17	1.65	1.72	1.98

T_0= 36% soil moisture content, T_1= 30% soil moisture content, T_2= 24% soil moisture content, T_3= 18% soil moisture content. DDI: Days of drought imposition. Mean values in the column having similar letter(s) do not differ significantly at the 5% level of probability.

Relative Water Content (RWC). Relative water content (RWC)% in plants decreases under drought stress because of the limited soil moisture, which reduces water uptake, leading to cellular dehydration. Lower RWC reflects a decline in leaf turgidity, which hampers physiological processes like photosynthesis, transpiration, growth, etc. Thus, RWC is widely used as a reliable indicator of tolerance status under drought (Upadhyaya et al., 2012). All the varieties showed significant differences among themselves in the case of RWC (Table 9). From this observation it was evident that the RWC in the genotype B/8/93 was higher (76.22%) compared to A/8/8 (69.01%), while other cultivars showed statistically similar results after 180 DDI. Another study observed that, drought stress of 20 days substantially decreased the RWC in tea plants, ranging from 80 ± 3% to 89 ± 6% (Upadhyaya et al., 2012). These observations also agreed with the results of the present study.

Proline Contents. Leaf proline is synthesized via the glutamate pathway and serves as a key indicator of stress tolerance by maintaining osmotic balance and protecting cells in tea plants (Arefin et al., 2025). Elevated proline accumulation was also noticed in several Indian tea varieties after 20 days of drought, ranging from 0.84 ± 0.05 to 4.4 ± 0.1 µ mol g^{-1} (Upadhyaya et al., 2012). The proline content of leaves was increased gradually with the increase of drought treatments (Table 9). All the genotypes except A/8/8 showed statistically similar proline accumulation after 180 days of drought, which is similar to the findings compared to the above studies.

3.2 Effect of Drought under Field Conditions

The effect of drought on morpho-physiological, biochemical, and water relations traits in six tea cultivars under field conditions is given below:

Root Volume. The root volume was found to be decreased with the increase of intensity of drought compared to control (Fig. 3). All the cultivars except A/8/8 showed statistically similar root volume (Table 10). Drought stress leads to a reduction in root volume because insufficient soil moisture limits cell elongation and root expansion. Water scarcity also slows down root metabolism, producing thinner and less developed roots. Consequently, overall root volume declines even though root length may increase as an adaptive strategy to explore deeper soil for moisture (Rokhmah et al., 2022).

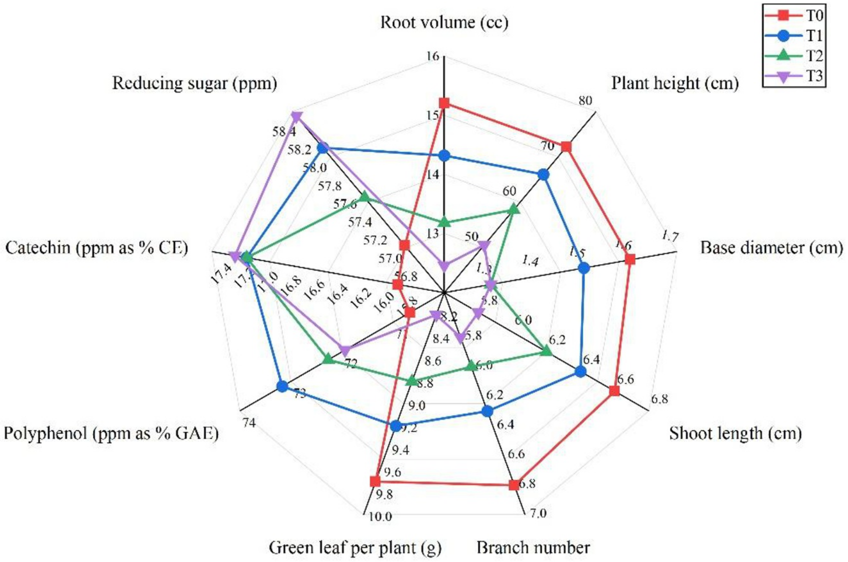

Fig. 3. Effect of different treatments on root volume (cc), plant height (cm), base diameter (cm), shoot length (cm), number of branches, green leaf per plant (g), polyphenol (ppm as % GAE), catechin (ppm as % CE), and reducing sugar (ppm) under field conditions. T_0= 36% soil moisture content, T_1= 30% soil moisture content, T_2= 24% soil moisture content, T_3= 18% soil moisture content.

Plant height. Plant height also varied significantly within various treatments, and stress treatments showed reduced height compared to the control (Fig. 3). Genotype B/8/93 exhibited maximum height (67.5 cm), followed by A/17/7 (64.3 cm), while A/8/8 had relatively lower height (48.8 cm) at 60 DDI (Table 10). Plant height in tea is generally influenced by pruning practices; however, in the present experiment, all the plants were subjected to a uniform pruning operation (decentering at 20 cm) and tipping height of 45 cm. Despite this standardized treatment, results revealed the significant genotypic variation in the reduction of plant height, indicating that water stress limited vegetative growth even under similar pruning regimes. The smaller reduction in morphological traits in tolerant tea genotypes under drought was also reported in various investigations (Cattivelli et al., 2008).

Base Diameter. The lateral growth, such as the growth of base diameter, was affected significantly due to drought stress. The mean values of all treatments showed that the 36% soil moisture content produced highest base diameter, followed by 30% soil moisture content while 18% soil moisture content produced lowest base diameter (Fig. 3). The base diameter of Mz/39 (1.57 cm) was less affected, followed by B/8/93 (1.55 cm) and A/17/7 (1.50 cm) under different treatments, possibly because of its higher tolerance to drought (Table 10). Drought stress in tea plants can significantly reduce stem diameter growth. For example, a tolerant genotype having a higher base diameter (1.84 cm) was identified than the susceptible plant (1.74 cm) during drought stress (Hossain et al., 2025). Studies have also shown that the impact on tea plant growth, including stem diameter, can be substantial, especially when water stress is prolonged (Yang et al., 2021).

Shoot Length. The shoot length of tea was an integrated result of various processes, including canopy photosynthesis, conversion of assimilates to biomass, and partitioning of assimilates to pluckable shoots (Damayanthi et al., 2010). In current experiment, shoot lengths were adversely affected by the drought stress over the control (Fig. 3). The genotypes B/8/93 and Mz/39 showed statistically similar and less relative reduction in shoot length compared to the other cultivars (Table 10). Two drought-tolerant cultivars (DG 39 and DN) were also identified that showed relatively more efficient plant growth than other cultivars during stress conditions (Langaroudi et al., 2023).

Number of Branches. In current study, after similar pruning at 20 cm (decentering) under field condition, the plants were subjected to drought stress, and branch number was recorded after 60 DDI. The branch number was gradually decreased with the increase of drought level (Fig. 3). B/8/93 and Mz/39 showed similar branch number (7.60), while A/8/8 exhibited lower number (4.75) (Table 10). The variation in branch number may be attributed to differences in genotype-specific tolerance capacity, where drought stress generally reduces carbohydrate availability, cell division, and bud sprouting. In tolerant genotypes, better water-use efficiency and stronger physiological adjustment (such as osmotic regulation and maintenance of turgor) supported higher bud break and branch initiation, while sensitive genotypes experienced suppressed branching due to limited photosynthate allocation and reduced hormonal activity (Yang et al., 2021). The highest branch number (3.75) in the best compatible root-scion tea saplings to mitigate drought stress after decentering was also recorded in another study (Ahmed et al., 2024).

Green Leaf Per Plant. The drought had a profound effect on the reduction of green leaf yield, and the reduction was severe with the increasing of drought (Fig. 3). The degree of relative reduction in green leaf yield was minimum in B/8/93, Mz/39, and Sh/9/65 cultivars (9.2 g) compared to the other genotypes at 60 DDI under field conditions (Table 10). The weight of green leaves depends on the size of pluckable shoots, which depends on the accumulation of photosynthates. Drought stress not only prevents the translocation and photosynthesis but also hinders the biochemical process of utilization of the food materials in new shoots as well as green leaf production (Hossain et al., 2017). A 30% reduction in green tea leaf production was reported in Iran in 2016, attributed to prolonged summer heat and reduced rainfall (Langaroudi et al., 2023).

Table 10. Genotypic differences in root volume (cc), plant height (cm), base diameter (cm), shoot length (cm), number of branches, green leaf per plant (g), polyphenol (ppm as % GAE), catechin (ppm as % CE), and reducing sugar (ppm) at combined treatments under field conditions after 60 DDI (days of drought imposition).

Genotypes	Root volume	Plant Height	Base diameter	Shoot length	Number of branch	Green leaf per plant	Polyphenol	Catechin	Reducing sugar
A/17/7	14.44 a	64.3 bc	1.50 ab	6.4 b	7.16 b	8.8 ab	74.55 c	17.92 b	62.72 c
Mz/39	14.77 a	63.4 c	1.57 a	7.1 a	7.60 a	9.2 a	77.17 b	17.86 b	63.97 b
Sh/9/65	14.31 a	65.0 b	1.48 b	6.07 b	5.28 d	9.2 a	71.93 d	15.79 c	54.99 d
BT2	14.11 a	61.6 d	1.47 b	6.4 b	6.45 c	9.1 ab	72.47 d	14.82 d	55.38 d
B/8/93	14.54 a	67.5 a	1.55 ab	7.2 a	7.60 a	9.2 a	89.81 a	23.07 a	72.91 a
A/8/8	10.58 b	48.8 e	1.11 c	6.4 b	4.75 e	8.4 b	55.13 e	13.73 e	42.71 e

Mean values in the column having similar letter(s) do not differ significantly at the 5% level of probability.

Total Polyphenol Content. In this study, total polyphenol content was found to gradually increase with the gradual increase of drought (Fig. 3). The maximum polyphenol content was accumulated in B/8/93 (89.81 ppm as % GAE), followed by Mz/39 (77.17 ppm as % GAE) and A/17/7 (74.55 ppm as % GAE), than the remaining genotypes (Table 10). A drought-tolerant tea cultivar (CNS 85) that accumulated more polyphenol (21.72%) under stress conditions was also reported in another study (Thiep et al., 2015). Polyphenol accumulation under stress also supplies energy for survival and growth, thereby helping the plants to tolerate stressful condition. Thus, the polyphenol content is a good indicator for screening drought-tolerant crop varieties (Zuo et al., 2024).

Catechin. The phenomenon of increased amounts of catechin in the leaves is another important factor during drought stress, which is a result of a series of metabolism interactions. In this experiment, the concentration of catechin in the new shoots was found to increase under drought stress (Fig. 3), and the accumulation was highest in B/8/93 (23.07 ppm as % CE) (Table 10). The accumulation of catechin in response to drought stress is also quite well documented (Zuo et al., 2024). The tea cultivar CNS 85 accumulated the highest amount of catechin (168.40 mg g^{-1}) under drought stress, which played a crucial role in enhancing its tolerance. Elevated catechin levels act as powerful antioxidants that protect cellular structures from oxidative damage caused by reactive oxygen species (ROS) during water deficit (Thiep et al., 2015).

Reducing Sugar. Reducing sugar content generally increases under drought stress as plants hydrolyze stored carbohydrates to maintain osmotic balance. This accumulation helps in osmotic adjustment, protects cellular structures, and provides an immediate energy source under limited photosynthesis (Zuo et al., 2024). In this study, unlike polyphenol and catechin content, reducing sugar was found to accumulate progressively with increasing levels of drought stress (Fig. 3). Among the six cultivars, B/8/93 showed the highest efficiency in reducing sugars (72.91 ppm), followed by Mz/39 (63.97 ppm) and A/17/7 (62.72 ppm), while A/8/8 was the lowest (42.71 ppm) (Table 10). A similar tendency was observed in the tea cultivar CNS 142, which showed the highest reducing sugar content (3.90%) under drought stress. This accumulation also provides an immediate source of energy to sustain essential metabolic activities when photosynthetic efficiency is reduced, thereby contributing to the drought tolerance capacity of CNS 142 (Thiep et al., 2015).

3.3 Identification of Tolerant Genotypes Through Clustering Analysis

Drought Tolerance under Nursery Conditions. Clustering analysis grouped the six tea genotypes into the following four distinct clusters (C-1, C-2, C-3, and C-4) based on their morphological, physiological, and water relations traits under nursery drought conditions (Fig. 4). Cluster 1 (C-1), comprising of A/8/8 cultivar, which was separated due to poor performance across most of the traits, reflecting sensitivity to drought stress. Cluster 2 (C-2), consisting of genotype B/8/93, showed consistently higher performance in maximum traits, indicating strong drought tolerance. On the other hand, Cluster 3 (C-3) and Cluster 4 (C-4), included BT2, Sh/9/65 and Mz/39, A/17/7 genotypes, respectively, showed moderate tolerance by their responses.

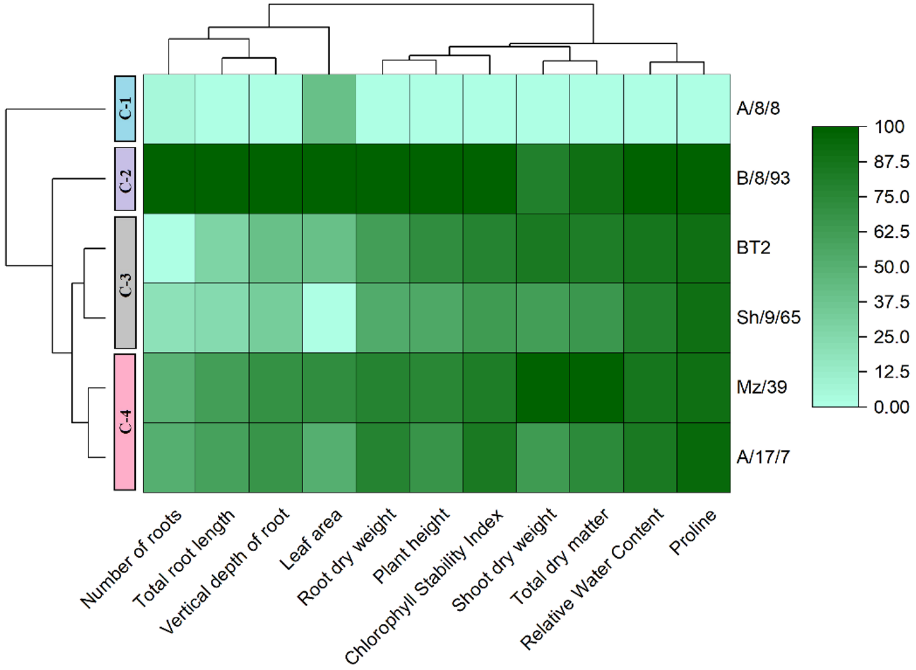

Fig. 4. Clustering heatmap represents the classification of genotypes for drought tolerance under nursery conditions. The mean values of traits are normalized and used to construct the heatmap (scaling from 0 to 100: light green to deep green, respectively).

Drought Tolerance under Field Conditions. Based on morpho-physiological and biochemical attributes, hierarchical clustering analysis catergorised the six tea genotypes into three distinct groups (C-1, C-2, and C-3) under field conditions (Fig. 5). Here also, Cluster 1 (C-1) was isolated from the others because of consistently weak performances and was comprised of A/8/8. On the contrary, B/8/93 and Mz/39 were placed into Cluster 2 (C-2), which demonstrated the strongest overall performance, reflecting their superior adaptability and resilience to drought stress in the field. Cluster 3 was grouped as intermediate performers and comprised of BT2, Sh/9/65, and A/17/7 cultivars.

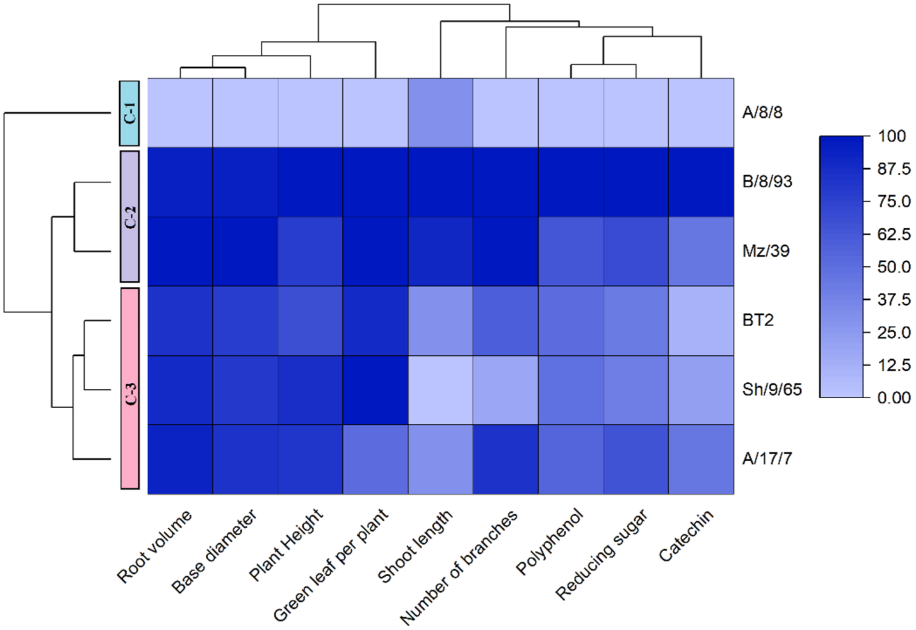

Fig. 5. Classification of genotypes for drought tolerance under field conditions by clustering heatmap. The mean values of traits are normalized and used to construct the heatmap (scaling from 0 to 100: light blue to deep blue, respectively).

Overall, B/8/93 emerged as the most tolerant genotype, while A/8/8 was identified as the most sensitive (Fig. 4 and Fig. 5). Mz/39 also performed well under field conditions, indicating its adaptability to natural drought stress, while the remaining genotypes (BT2, Sh/9/65, and A/17/7) showed intermediate tolerance. The clustering approach thus enabled efficient classification of tea genotypes into tolerant, moderately tolerant, and sensitive groups, facilitating selection for further breeding and management under water-limited conditions (Hossain et al., 2025).

4 Conclusion

The performances of six tea genotypes under drought stress revealed significant differences in morpho-physiological, biochemical, and water responses. Relative reduction in growth traits and yield, along with increases in proline, polyphenols, catechins, and sugars, proved to be useful indices for drought tolerance. Among the genotypes, B/8/93 was identified as the most tolerant, A/8/8 as the most sensitive, while Mz/39 showed wide adaptability under field conditions. The remaining cultivars (BT2, Sh/9/65, and A/17/7) exhibited intermediate tolerance. Further multi-environment and molecular studies are necessary to validate these findings and strengthen breeding strategies for drought-resilient tea.

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Disclosure of Interests. The authors declare that they have no conflicts of interest to report regarding the present study.

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