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RESEARCH ARTICLE

Unraveling drought tolerance in spray chrysanthemum through multivariate analysis under hydroponic and pot culture conditions

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Abstract

Drought affects chrysanthemum growth and yield, highlighting the need for reliable indicators of tolerance. This study evaluated 68 genetically diverse genotypes under hydroponics and pot culture using a completely randomized design to identify key physiological, biochemical and morphological traits linked to drought adaptation. Stress was imposed with 10 % polyethylene glycol (PEG) in hydroponics at the seedling stage and 60 kPa soil moisture tension in pots for 10 days at the vegetative stage based on our preliminary studies. Traits measured included chlorophyll content, carotenoids, relative water content (RWC), membrane stability index (MSI), canopy temperature depression (CTD), chlorophyll fluorescence, biomass and reproductive parameters. Correlation analysis revealed strong positive associations among chlorophyll content, RWC, MSI, CTD and fluorescence, suggesting coordinated mechanisms that preserve photosynthetic efficiency and cellular stability. Principal component analysis (PCA) identified chlorophyll b, fluorescence, biomass and reproductive traits as major contributors to phenotypic variation. The first few PCs explained 71.0 % variation in hydroponic control, 75.5 % in hydroponic stress, 61.6 % in pot control and 77.9 % in pot stress, indicating context-dependent adaptive strategies. Under hydroponic stress, chlorophyll a, total chlorophyll, biomass, chlorophyll *b* and carotenoids contributed strongly to both PC1 and PC2, with Punjab Singar, Violet, Gulmohar, Garden Beauty and Shwet excelling in these traits, while Autumn Joy and Naughty White were distinguished by flowers per plant and chlorophyll a/b. In contrast, under pot stress, no trait contributed simultaneously to both PCs. Overall, chlorophyll content, RWC, MSI, CTD, chlorophyll fluorescence and reproductive traits emerged as robust indicators for breeding drought-resilient chrysanthemum cultivars.

Keywords: chrysanthemum; correlation analysis; membrane stability index; drought stress; polyethylene glycol; principal component analysis

Introduction

A variety of biotic and abiotic factors can seriously hinder growth and development in plants because of their sessile nature (1). Among these, moisture stress continues to be one of the most harmful abiotic constraints, limiting plant distribution, decreasing yields and threatening the sustainability of agriculture worldwide (2).

Widely grown as a cut flower, potted plant and landscape species, chrysanthemums (*Chrysanthemum morifolium* Ramat.) are among the most commercially valuable ornamental plants in the world (3). Its cultivation is, however, becoming more limited by abiotic challenges, especially water scarcity, which has a negative impact on quality, yield and geographic adaptibility (4). In the absence of cost-effective technologies for mitigating drought effects, breeding for stress moisture tolerance has become a vital strategy for sustaining and expanding production (5).

Plants modify their osmotic potential to withstand water scarcity by accumulating solutes. This lowers cellular osmotic potential and maintains turgor pressure, which is necessary for photosynthetic activity and stomatal function (6). Because of its high molecular weight and non-ionic features, PEG is frequently used in experimental systems to mimic drought by causing osmotic stress without causing cytotoxic effects (7). Early-stage drought screening of genotypes in controlled conditions frequently use PEG-induced stress models (8).

In both dryland and irrigated systems, drought stress reduces productivity by interfering with physiological, morphological and biochemical processes (9). Numerous physiological markers, including CTD, MSI, RWC and chlorophyll fluorescence, have been proven to be accurate indicators of drought tolerance in a wide range of crops (10 -12). Water stress also affects biomass, a crucial yield-related trait that is essential for choosing genotypes that can withstand stress (13).

For crop development to be effective, it is essential to determine how these traits interact under stress and non-stress situations. PCA helps find traits that significantly contribute to genetic diversity, whereas correlation analysis helps choose desirable traits. In ornamental breeding programs, these statistical tools are crucial for dissecting complicated traits like drought tolerance (14).

This study aimed to identify key morphological, physiological and biochemical traits associated with drought tolerance in chrysanthemum and to assess their interrelationships for use in trait-based breeding under contrasting moisture regimes. A total of 68 genetically diverse spray chrysanthemum (*Chrysanthemum* × *morifolium* Ramat.) genotypes were evaluated using hydroponic and pot culture systems, with correlation and PCA applied to highlight reliable indicators of tolerance. Spray chrysanthemums were selected owing to their rising commercial importance in cut flower and pot plant markets and their broad genetic diversity, which provides contrasting responses to stress. Hydroponics enabled rapid, uniform screening at the seedling stage using PEG-6000, while pot culture at the vegetative stage under soil moisture deficit captured field-like morphological and reproductive responses. To our knowledge, this is the first large-scale, dual-environment evaluation in chrysanthemum, providing a foundation for developing robust selection indices for drought-resilient cultivars.

Materials and Methods

Plant material and growing condition

The study was conducted under controlled greenhouse conditions at the Division of Floriculture and Landscaping, ICAR-Indian Agricultural Research Institute (ICAR-IARI), New Delhi. A total of 68 spray chrysanthemum (*Chrysanthemum morifoilium* Ramat.) genotypes were selected from the institute germplasm collection. Propagation was done during June -July using terminal cuttings of 5-7 cm length taken from healthy, disease-free mother plants. Prior to planting, cuttings were treated with 500 ppm indole-3-butyric acid (IBA) to enhance rooting. The cuttings were placed in pro-trays filled with a sterile rooting media consisting of cocopeat, vermiculite and perlite in a 2:1:1 ratio. To prevent soil-borne fungal infections, trays were drenched with carbendazim at 1 g L^{-1} concentration. The weather data were recorded daily and the average temperatures of the greenhouse were 29 ± 1 °C/18 ± 1 °C (day/night), 5.6 hours of bright sunshine hours, 71 % relative humidity and evapotranspiration of 3.68 mm.

Hydroponic culture and osmotic stress treatment

Thirty-day-old root cuttings were carefully removed from trays, gently washed and transferred to a hydroponic system containing Hoagland's nutrient solution for acclimatization. To simulate drought stress, PEG-6000 was incorporated into the medium. The PEG concentration had been standardized through a preliminary trial using 0, 5, 10 and 15 % levels, where plant responses were evaluated based on relative water content, wilting symptoms, chlorophyll content and survival rate. From these observations, 10 % PEG-6000 was identified as the optimum concentration that induced sufficient osmotic stress to discriminate between tolerant and susceptible genotypes without causing excessive mortality. Accordingly, seedlings were subjected to 10 % PEG-6000 for 7 days, after acclimatization in hydroponics (10 days) while control plants were maintained in regular Hoagland's solution (15). At the end of the treatment, several drought tolerance-associated traits were assessed.

Pot culture and moisture stress imposition

One month old seedlings of 68 genotypes were transplanted into white plastic pots containing 12 kg of a homogeneous field soil mixture for pot culture. Drought stress was imposed in two distinct cycles, at the vegetative and flower bud initiation stages, by maintaining soil matric potential at -60 kPa for 10 consecutive days (based on preliminary trail), monitored using tensiometers. Most chrysanthemum studies have applied mild soil moisture tensions and reported stress symptoms by around -32 kPa (16, 17). Therefore, -60 kPa was chosen to impose a stronger but non-lethal deficit, representing a realistic field-relevant stress level that differentiates tolerant and susceptible genotypes while avoiding irreversible damage. Soil water potential was standardized across all pots by daily weighing and irrigation to maintain the target potential, while pots were randomized and rotated periodically to minimize environmental variation. Control plants were maintained under ideal moisture conditions. Uniformity of stress was confirmed by measuring leaf RWC in representative plants. After the second stress cycle, morphological and physiological traits responsive to drought were recorded.

Different traits under study

MSI was estimated by a standard procedure (18) and RWC was measured using a widely adopted method (19). Chlorophyll and carotenoids were quantified by the DMSO extraction method with standard equations (20, 21). Canopy temperature was recorded on the 6th day of stress using an infrared thermometer (Fluke 59 Mini) and CTD was calculated from the difference between canopy and ambient temperature (22). Chlorophyll fluorescence was assessed with a FluorPen FP 100 (Photon systems instruments, Czech Republic) following standard protocols (23). Morphological traits including biomass (recorded as shoot dry weight after oven-drying at 70 °C to constant weight), plant height, plant spread, number of branches, stem girth, flower weight, flower diameter, number of flowers per plant and yield per plant were recorded at flowering according to PPV & FRA guidelines from all replicates (24).

Experimental design and statistical analysis

The experiment was conducted in a factorial CRD with three replications per genotype under control and drought stress conditions, with three biological replicates per trait. Traits were analyzed using analysis of variance (ANOVA) and treatment means were compared with Tukey's HSD test at $p \leq 0.05$. Correlation analysis and PCA were performed in R software (v2.15.1). PCA was validated using Scree plot analysis. For PCA, eigenvalues >1.0 were retained and trait loadings ≥0.30 were considered significant contributors, with cumulative variance used to identify the traits most responsible for genotype differentiation under stress and control conditions.

Results

Correlation analysis

Correlation analysis was conducted with data from 68 genotypes (n = 68) and all reported associations were significant at $p < 0.01$. Correlation analysis under control (Fig. 1A) conditions (Hoagland solution) revealed that chlorophyll a had a strong positive correlation with total chlorophyll and chlorophyll b, whereas the chlorophyll a/b ratio exhibited a significant negative correlation with chlorophyll b. Under osmotic stress (Fig. 1B) conditions (10 % PEG), chlorophyll a maintained a highly significant positive correlation with total chlorophyll and chlorophyll b. Additionally, chlorophyll b displayed strong positive correlations with carotenoids, RWC, CTD and chlorophyll fluorescence. The MSI also showed a significant positive correlation with chlorophyll b, RWC, CTD and chlorophyll fluorescence. Furthermore, a notable positive relationship was observed between RWC and MSI, chlorophyll b and chlorophyll fluorescence under drought conditions.

Under pot culture and non-stress conditions (Fig. 1C), the correlation of pigment traits closely mirrored the patterns observed under hydroponics control. Under stress condition (Fig. 1D) chlorophyll a showed a very high positive correlation with chlorophyll b, total chlorophyll and carotenoids. Similarly, total chlorophyll exhibited a strong positive correlation with carotenoids. Carotenoids were significantly and positively correlated with CTD and plant spread.

Under stress RWC displayed a highly significant positive correlation with MSI, chlorophyll fluorescence, CTD and plant spread. Additionally, CTD was strongly correlated with MSI, while chlorophyll fluorescence showed a significant positive correlation with both CTD and plant spread. Plant spread was also highly correlated with MSI and biomass. A strong positive correlation was found between the number of primary and secondary branches. Fresh flower weight was positively and significantly correlated with flower diameter.

Principle component analysis

The rotated component matrix (Table 1) presents the percentage of variation explained by selected principal components (PCs) with eigenvalues greater than 1 and their association with other traits under hydroponics. Under hydroponics stress treatment, two main components accounted for nearly 75 % of the overall variation, with PC1 and PC2 contributing cumulatively 75.47 % of the total variation (Fig. 2B). Chlorophyll b (0.38) and chlorophyll fluorescence (0.36) showed high positive loadings on PC1, while chlorophyll a (0.52) and total chlorophyll (0.46) had high positive loadings on PC2. Under hydroponics non-stress conditions, four PCs were significant, explaining 71.04 % of the total variation, with the first two PCs contributing 48.07 % (Fig. 2A). Chlorophyll b (0.53) and total chlorophyll (0.52) had high positive loadings on PC1, whereas canopy temperature depression (0.60) and biomass (0.46) loaded positively on PC2.

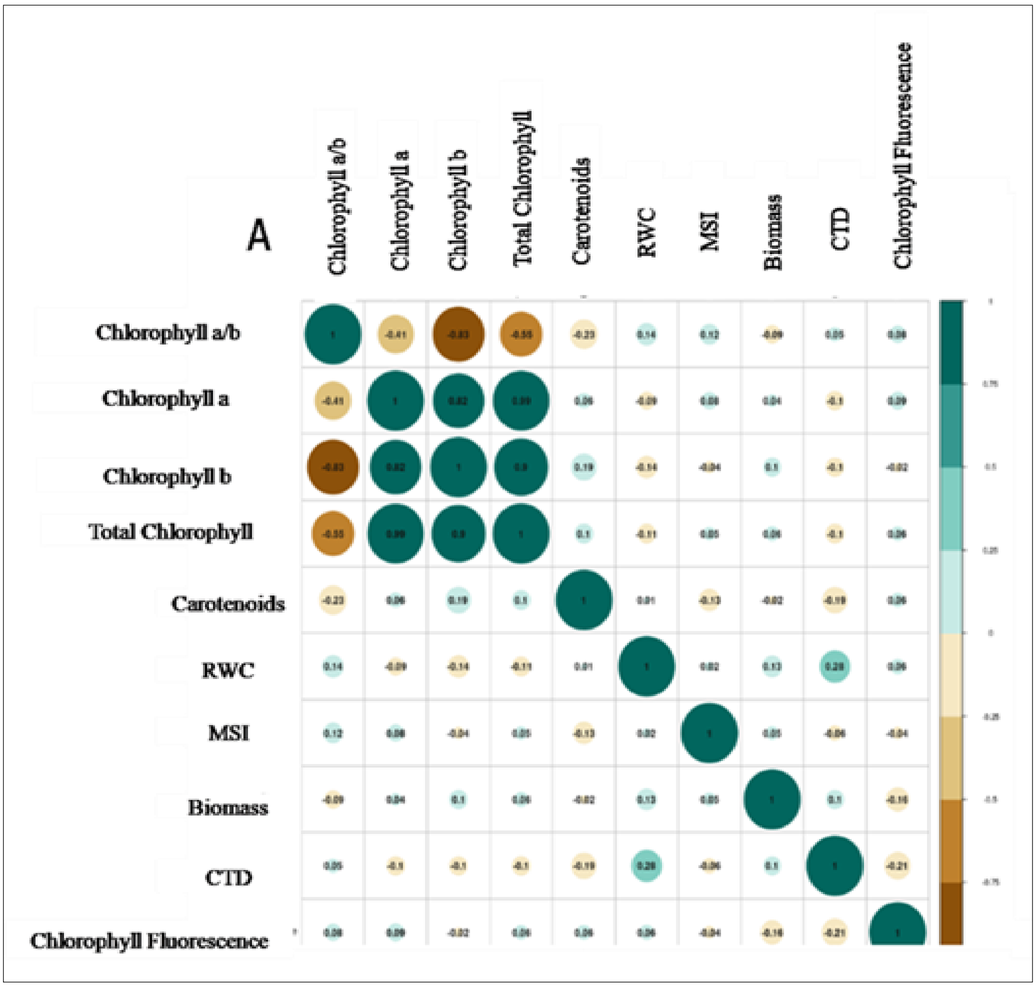
Table 1. Principle component matrix of 10 characteristics of 68 genotypes of chrysanthemums assessed in hydroponics under stress and non-stress conditions

Trait	Control				Stress	
	PC1	PC2	PC3	PC4	PC1	PC2
Eigenvalues	3.36	1.44	1.19	1.1	6.06	1.49
% variance explained	33.63	14.44	11.95	11.0	60.6	14.87
Cumulative % variance	33.63	48.07	60.03	71.04	60.6	75.47
Chlorophyll a/b	-0.41	-0.07	0.3	-0.14	-0.35	0.2
Chlorophyll a	0.49	0.05	0.25	-0.15	0.29	0.52
Chlorophyll b	0.53	0.07	-0.06	0.01	0.38	0.18
Total chlorophyll	0.52	0.06	0.17	-0.1	0.32	0.46
Carotenoid	0.13	-0.26	-0.55	-0.14	0.31	0.08
RWC	-0.1	0.39	-0.07	-0.69	0.33	-0.3
MSI	-0.01	0.09	0.68	0.1	0.28	-0.37
Biomass	0.05	0.46	-0.08	0.08	0.15	0.32
CTD	-0.1	0.6	-0.12	-0.16	0.34	-0.29
Chlorophyll fluorescence	0.02	-0.43	0.16	-0.65	0.36	-0.19

The rotated component matrix (Table 2) displays the percentage of variation explained by selected PCs with eigenvalues greater than 1 and their association with other traits under pot culture conditions. Under pot culture stress treatment, five main components accounted for nearly 78 % of the overall variation, with PC1 and PC2 contributing the largest cumulative influence of 52.37 % (Fig. 2D). Chlorophyll b (-0.32) and carotenoids (-0.31) had high negative loadings on PC1, while flower weight (-0.51) and flowers per plant (0.44) showed high negative and positive loadings, respectively, on PC2. In PCA, the sign of a loading (positive or negative) only shows the direction of association with the component. For the control (non-stress) condition, eight PCs were significant, explaining 79.22 % of the total variation, with the first two PCs accounting for 33.23 % (Fig. 2C). Under non-stress conditions, flower weight (0.39) and flowers per plant (-0.38) had high positive and negative loadings, respectively, on PC1, while biomass (-0.38) and yield per plant (-0.37) showed high negative loadings on PC2. The relationship between eigenvalues and percentage variation is depicted in the

Table 2. Principle component matrix of 19 characteristics of 68 genotypes of chrysanthemums assessed in pot culture under stress and non-stress conditions

Traits	Control								Stress				
	PC1	PC2	PC3	PC4	PC5	PC6	PC7	PC8	PC1	PC2	PC3	PC4	PC5
Eigen values	3.19	3.11	2.45	1.56	1.37	1.18	1.12	1.03	7.15	2.8	2.27	1.47	1.1
% variance explained	16.81	16.41	12.9	8.24	7.23	6.24	5.93	5.44	37.62	14.75	11.97	7.74	5.78
Cumulative % Variance	16.81	33.23	46.13	54.37	61.6	67.85	73.78	79.22	37.62	52.37	64.35	72.1	77.89
Chlorophyll a/b	0.1	-0.06	0.44	-0.44	0.07	-0.09	-0.29	-0.19	0.27	-0.05	-0.11	0.03	-0.03
Chlorophyll a	-0.32	-0.31	-0.17	-0.3	0.15	-0.08	-0.2	-0.18	-0.3	-0.02	0.3	-0.25	0.09
Chlorophyll b	-0.25	-0.1	-0.49	0.25	0.02	0.06	0.17	0.09	-0.32	0.02	0.26	-0.23	0.06
Total chlorophyll	-0.34	-0.3	-0.28	-0.2	0.13	-0.06	-0.13	-0.14	-0.3	-0.01	0.3	-0.25	0.08
Carotenoids	-0.11	-0.22	-0.09	0.11	0.42	0.04	0.19	-0.09	-0.31	0.05	0.17	-0.2	0.03
RWC	0.0	0.07	0.14	-0.35	0.39	0.05	0.32	0.33	-0.29	0.04	-0.07	0.4	-0.23
MSI	-0.09	-0.12	-0.1	-0.14	-0.36	0.51	-0.35	0.01	-0.28	0.04	-0.1	0.36	-0.14
CTD	-0.24	-0.03	0.03	-0.43	-0.11	0.24	0.3	0.06	-0.28	0.1	0.09	0.33	0.12
Chlorophyll fluorescence	-0.07	0.06	-0.1	-0.16	-0.51	-0.08	0.14	-0.31	-0.29	0.04	0.0	0.37	-0.05
Plant height	0.31	-0.33	0.07	0.14	0.01	0.13	0.11	-0.3	-0.11	-0.38	-0.24	-0.02	0.17
Plant sspread	-0.05	-0.29	0.18	0.07	0.08	-0.39	0.1	-0.36	-0.28	0.01	-0.17	-0.1	-0.28
Primary branches	-0.2	-0.17	0.33	0.2	0.18	0.43	-0.07	0.04	-0.08	0.25	-0.28	0.03	0.69
Secondary branches	-0.31	-0.13	0.35	0.31	0.02	0.14	-0.23	0.15	-0.15	0.37	-0.3	-0.07	0.29
Stem girth	-0.04	-0.28	0.17	-0.12	-0.27	0.08	0.52	0.21	-0.07	-0.02	-0.37	-0.4	-0.22
Flower weight	0.39	-0.27	-0.2	-0.07	0.02	-0.05	-0.15	0.21	-0.1	-0.51	-0.07	0.06	0.01
Flower diameter	0.3	-0.23	-0.13	-0.18	0.08	0.15	-0.06	0.11	-0.16	-0.39	0.0	-0.05	0.1
Flowers/plant	-0.38	0.05	0.15	0.01	-0.18	-0.41	-0.07	0.28	-0.07	0.44	-0.09	-0.16	-0.41
Yield/plant	0.1	-0.37	-0.03	-0.01	-0.15	-0.29	-0.21	0.52	-0.18	-0.17	-0.34	-0.05	-0.08
Biomass	0.05	-0.38	0.19	0.22	-0.25	-0.03	0.2	-0.07	-0.19	-0.05	-0.42	-0.17	-0.03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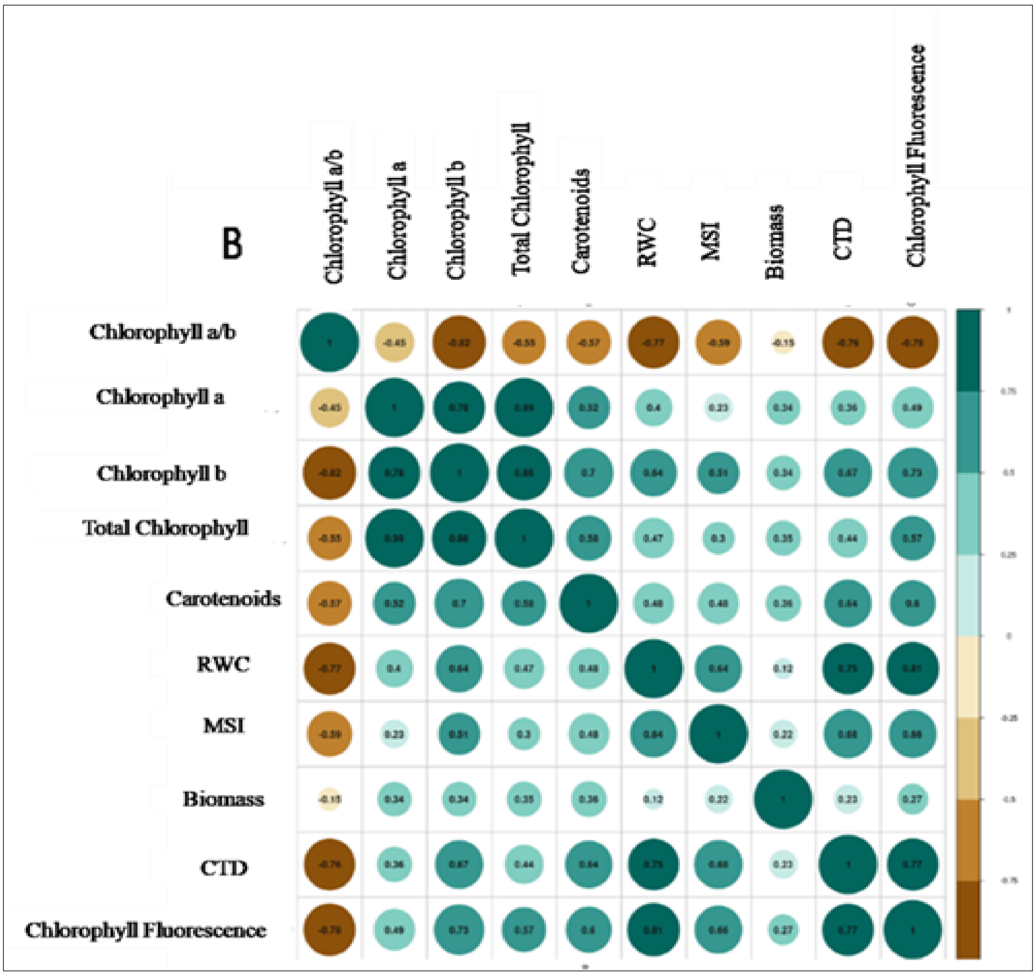
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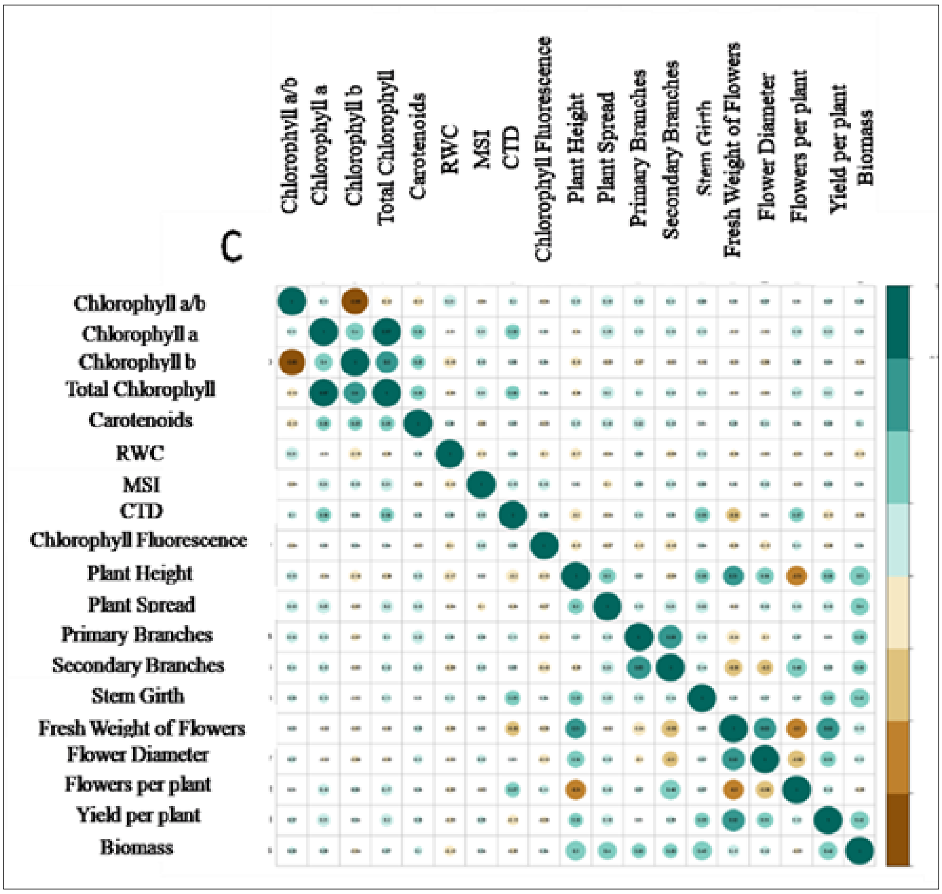

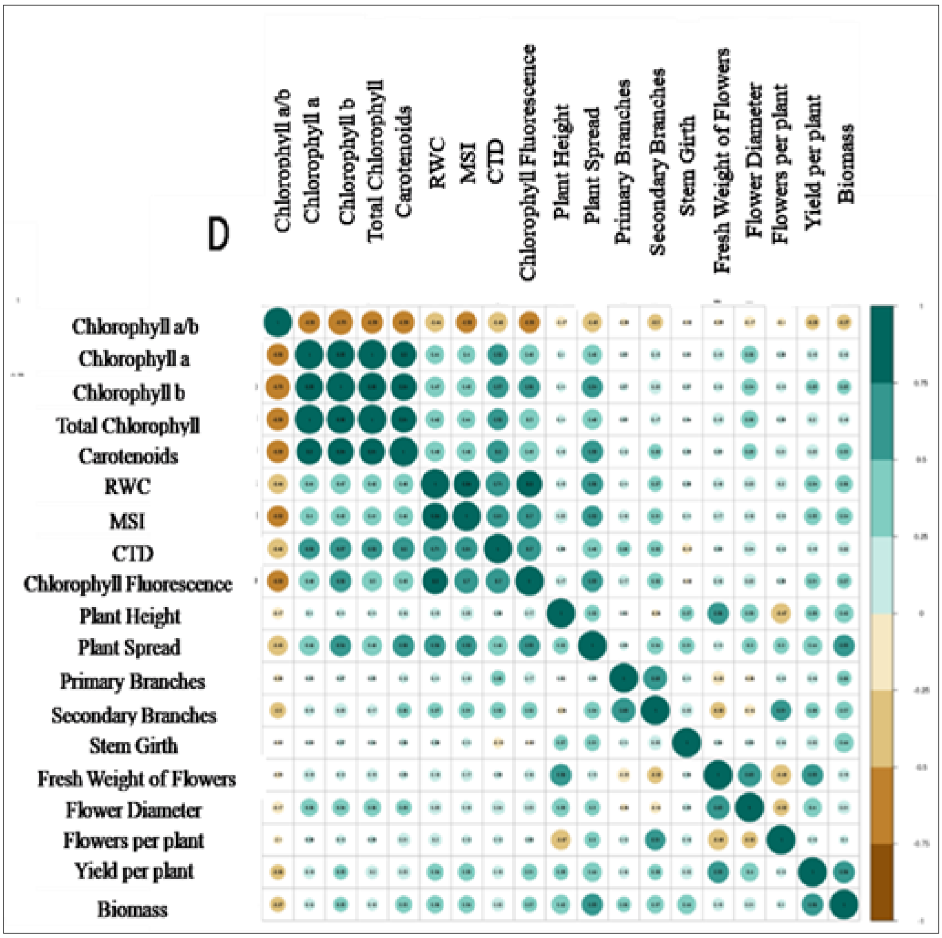

Fig.1. Correlation matrices of morphological, physiological and biochemical traits of chrysanthemum genotypes under contrasting moisture regimes: (A) control condition in hydroponics, (B) drought stress condition in hydroponics, (C) control condition in pot culture and (D) drought stress condition in pot culture. Correlation coefficients are shown, with significance levels at $p < 0.01$. Positive correlations are indicated in green and negative correlations in brown, with color intensity proportional to correlation strength.

scree plots (Fig. 2A- 2D). The principal component analysis under pot culture indicated that, under stress, chlorophyll b and carotenoids had a strong influence on selection and can be selected simultaneously, followed by chlorophyll a, total chlorophyll, RWC and chlorophyll fluorescence (Table 2).

Principal component biplot analysis

PCA biplots (Fig. 3A-3D) illustrate relationships among traits, genotypes and their associated PCs. The vectors representing variables showed that the closer a vector was aligned with a PC axis, the more it contributed to that PC. The length of the vector indicated the extent of variability explained by that trait. The angle between vectors reflected trait correlations.

Under hydroponic osmotic stress, chlorophyll a, total chlorophyll, biomass, chlorophyll b and carotenoids were the key traits positively contributing to both PC1 and PC2. Punjab Singar excelled in chlorophyll a, primarily influenced by biomass and total chlorophyll. Genotype Violet performed best in total chlorophyll, while genotype Gulmohar showed superior chlorophyll b levels and influenced by carotenoids. Genotypes Garden Beauty and Shwet were notable for carotenoids, influenced by chlorophyll b. Genotype Autumn Joy showed high performance for flowers per plant and genotype Naughty White for chlorophyll a/b (Fig. 3B). Under pot culture stress, no traits positively contributed to both PC1 and PC2, although flowers per plant and chlorophyll a/b were relatively important based on their proximity to the positive side (Fig. 3D)

Discussion

This study explored the physiological, biochemical and morphological traits underlying drought stress responses through correlation and PCA across hydroponic and pot culture systems. Beyond identifying trait associations, the findings provide insights into the mechanisms that govern drought tolerance and highlight key indicators relevant for breeding programs.

The strong correlations among chlorophyll pigments under both control and drought conditions reflect the tightly regulated biosynthetic pathways that sustain photosynthetic efficiency. The inverse relationship between the chlorophyll a/b ratio and chlorophyll b suggests adaptive remodeling of light-harvesting complexes, a strategy reported in earlier studies to optimize energy capture and reduce photodamage under stress (25). The persistence of pigment associations, especially between chlorophyll b, carotenoids and chlorophyll fluorescence, underscores the integrated role of pigments in photoprotection. Carotenoids not only safeguard photosystems through reactive oxygen species (ROS) scavenging but also interact with water-related traits, indicating their dual role in both structural stability and oxidative stress mitigation (26).

Water status emerged as a central determinant of drought resilience, as reflected in the positive linkage between RWC, MSI, CTD and chlorophyll fluorescence. This integrative relationship highlights how physiological water retention supports both cellular integrity and photosystem II stability. Previous research has emphasized CTD as a rapid, non-destructive proxy for stomatal conductance and transpirational cooling (27) and our findings reinforce its value for field-level screening. The alignment of these physiological markers with MSI suggests that drought tolerance involves coordinated water conservation and protection of membrane integrity rather than reliance on a single trait. The positive correlations between RWC, CTD and chlorophyll fluorescence highlight an integrated drought-response mechanism where efficient water retention sustains stomatal conductance and canopy cooling, which in turn protects photosystem II efficiency. Similar linkages between water status, thermal regulation and fluorescence performance have been reported in grasses (28).

Morphological parameters, particularly plant spread, branch number and reproductive traits, were closely linked with physiological traits, reflecting a whole-plant integration of stress

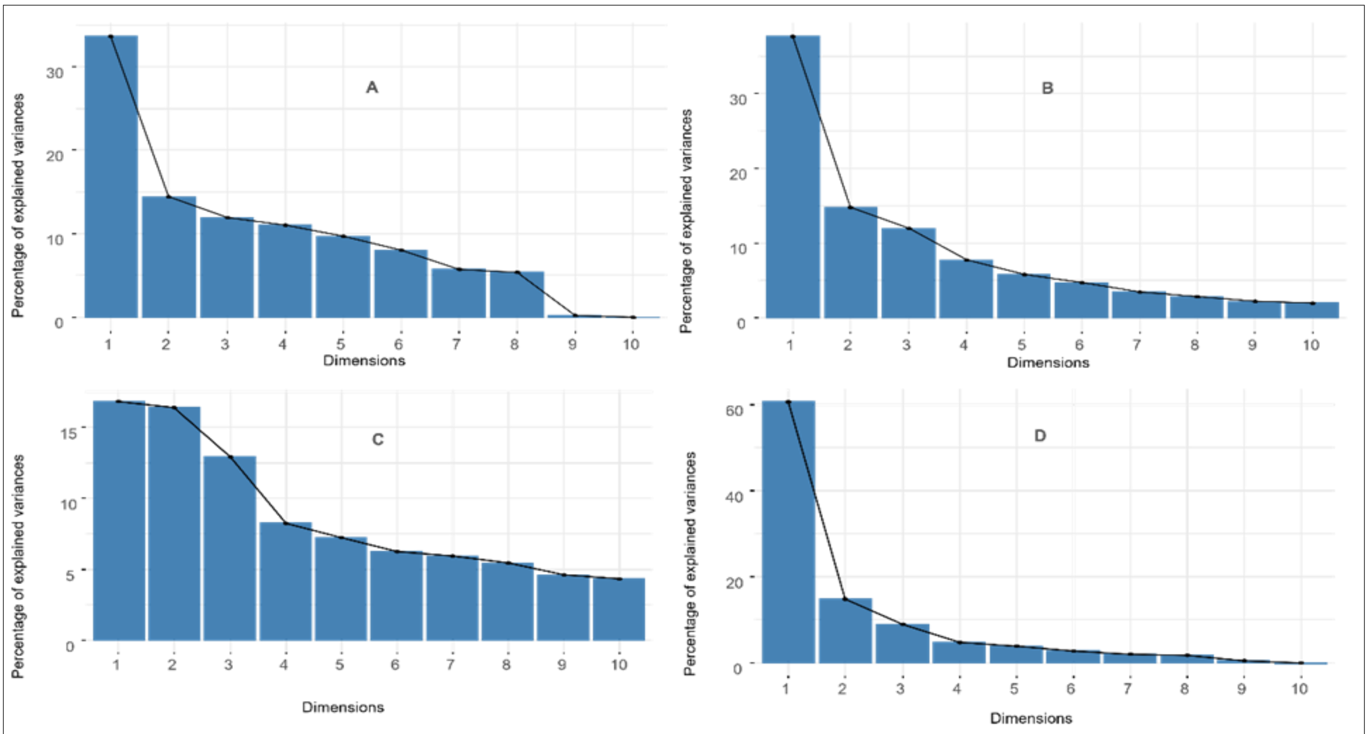

Fig. 2. Scree plots showing eigenvalue distribution of principal components derived from morphological, physiological and biochemical traits of chrysanthemum genotypes under contrasting moisture regimes. (A) Control condition in hydroponics, (B) Drought stress condition in hydroponics, (C) Control condition in pot culture and (D) Drought stress condition in pot culture.

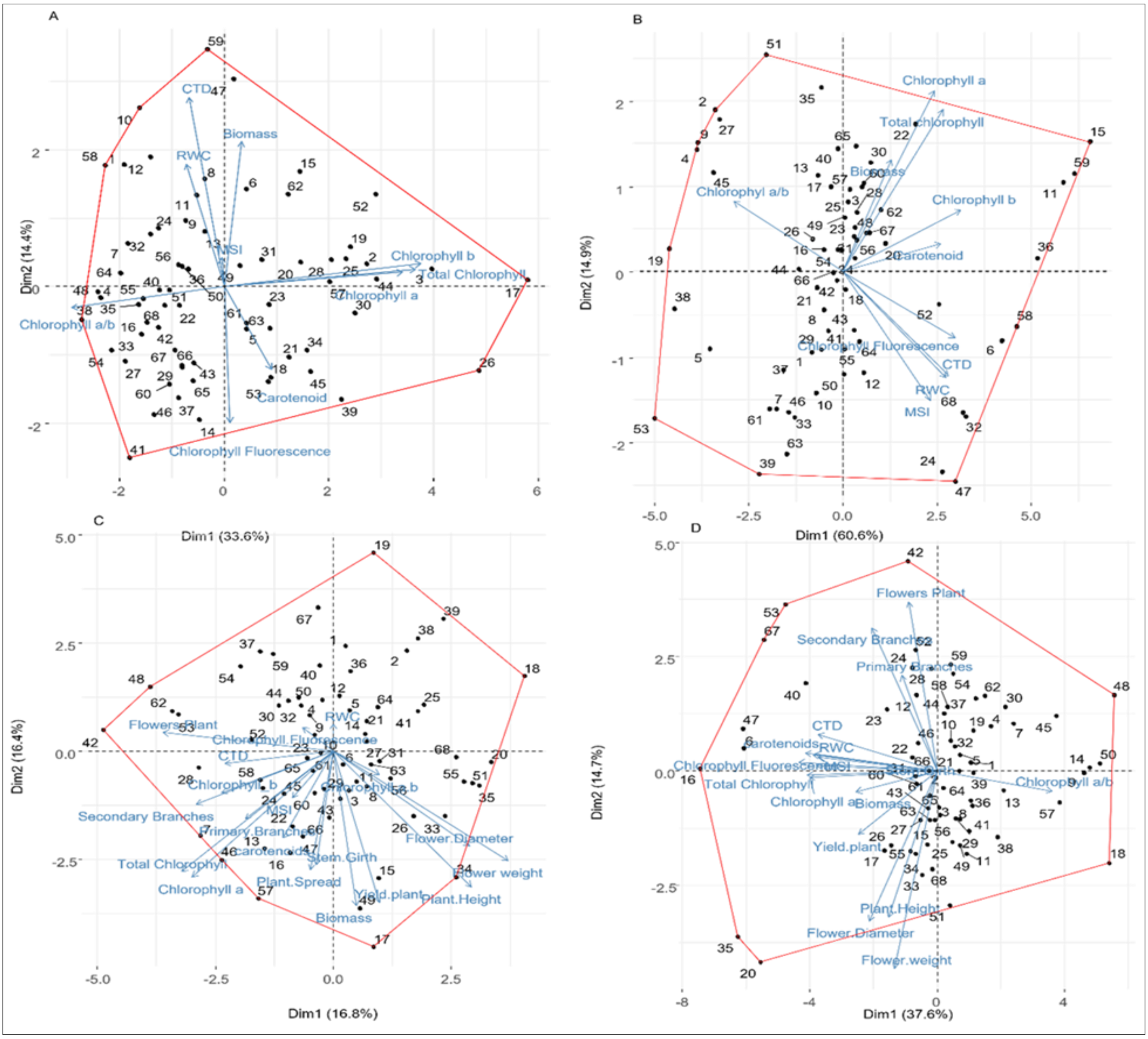

Fig. 3. PCA biplots of morphological, physiological and biochemical traits of chrysanthemum genotypes under contrasting moisture regimes. (A) Control condition in hydroponics, (B) Drought stress condition in hydroponics, (C) Control condition in pot culture and (D) Drought stress condition in pot culture.

responses. The maintenance of flower weight and diameter despite drought stress suggests an adaptive reallocation of resources to reproduction, a strategy consistent with reports of stress-induced reproductive plasticity (29). This capacity to safeguard reproductive fitness even under water-limited conditions has direct implications for yield stability.

Reproductive traits such as flowers per plant, flower diameter and flower weight maintained similar correlation patterns under both stress and non-stress conditions. This stability suggests that reproductive performance is buffered against drought, likely because allocation to reproductive structures is prioritized even when vegetative or pigment traits are affected. Such consistency aligns with reports in chrysanthemum and other ornamentals where reproductive traits show resilience across environments, supporting their use as reliable indicators in breeding programs (30).

PCA analysis further distinguished between hydroponic and pot culture conditions, revealing context-dependent trait hierarchies. Under hydroponic osmotic stress, chlorophyll pigments, carotenoids and biomass emerged as key contributors to variation, highlighting the importance of pigment stability and growth maintenance in early drought tolerance. Genotypes such as Punjab Singar, violet and gulmohar performed well for chlorophyll traits, while garden beauty and shwet stood out in carotenoids, indicating their role in antioxidant defense. Recent work in local chrysanthemum germplasm confirms that variation in pigment content strongly contributes to ornamental and physiological performance under stress, supporting these observations (30). In contrast, pot culture stress revealed no trait contributing simultaneously to both PC1 and PC2, suggesting a more complex adaptive response; here, flowers per plant and the chlorophyll *a/b* ratio were relatively more informative. Stability assessment studies of chrysanthemum hybrids across multiple environments also report that reproductive traits and pigment ratios often vary more than biomass under field conditions (31). Notably, positive and negative PCA loadings reflect trait direction along the component axis rather than trait importance, meaning that traits with negative loadings (e.g., carotenoids in certain PCs) represent contrasting but equally relevant adaptation strategies.

Taken together, the integration of correlation and PCA identified a suite of interlinked traits chlorophyll content, carotenoids, RWC, MSI, CTD, chlorophyll fluorescence and reproductive traits as robust indicators of drought tolerance. Their centrality across analytical approaches indicates that selection for one trait may confer indirect benefits on others due to pleiotropy or physiological linkage. However, the genotype-by-environment differences underscore that trait prioritization must remain context-specific; genotypes excelling under controlled hydroponic conditions may not necessarily translate to soil-based performance. This highlights the importance of multi-environment evaluation and breeding strategies tailored to specific agro-ecological contexts (32).

Conclusion

This study identifies chlorophyll content, carotenoids, RWC, MSI, chlorophyll fluorescence, plant spread and reproductive traits yield per plant as key indicators of drought tolerance. For breeding and field applications, chlorophyll fluorescence and CTD provide rapid, non-destructive screening tools, while RWC and MSI serve as robust physiological benchmarks. Reproductive traits under stress further ensure yield resilience. These findings emphasize that drought-tolerance breeding should prioritize these traits within environment-specific selection frameworks, enabling the development of cultivars that combine photosynthetic stability, efficient water use and yield assurance across diverse agro-ecological conditions.

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Authors' contributions

Conceptualization of research was done by N, BP, GK, SK, AMS and GK. N, BP, SP design the experiments. GK and KPS contributed the experimental materials. Execution of field/lab experiments and data collection was done by BP and N). Statistical analysis of data and interpretation performed by BP and N. BP, N and SP prepared the manuscript. All authors read and approved the final manuscript.

Compliance with ethical standards

Conflict of interest: The authors declare no conflict of interest.

Ethical issues: None

References

1. Umar OB, Ranti LA, Abdulbaki AS, Bola AUL, Abdulhamid AK, Biola MR, et al. Stresses in plants: Biotic and abiotic. In: Current trends in wheat research. London: IntechOpen; 2021. https://doi.org/10.5772/intechopen.100501
2. Seleiman MF, Al-Suhaibani N, Ali N, Akmal M, Alotaibi M, Refay Y, et al. Drought stress impacts on plants and different approaches to alleviate its adverse effects. Plants. 2021;10(2):259. https://doi.org/10.3390/plants10020259
3. Chen Q, Gao K, Xu Y, Sun Y, Pan B, Chen D, et al. Research advance on cold tolerance in chrysanthemum. Front Plant Sci. 2023;14:1259229. https://doi.org/10.3389/fpls.2023.1259229
4. Zhang Y, Gu J, Xia X, Zeng J, Sun H, Chen F, et al. Contrasting responses to drought stress between *Chrysanthemum japonense* and *C. nankingense*. Ornam Plant Res. 2022;2(1):1-11. https://doi.org/10.48130/OPR-2022-0016
5. Rasheed A, Jie Y, Nawaz M, Jie H, Ma Y, Shah AN, et al. Improving drought stress tolerance in ramie (*Boehmeria nivea* L.) using molecular techniques. Front Plant Sci. 2022;13:911610. https://doi.org/10.3389/fpls.2022.911610
6. Yang X, Lu M, Wang Y, Wang Y, Liu Z, Chen S. Response mechanism of plants to drought stress. Horticulturae. 2021;7(3):50. https://doi.org/10.3390/horticulturae7030050
7. Limbu AK, Dhami A, Rawal JS, RC P, Mandal A, Awasthi R, et al. Assessment of drought tolerance in different rice genotypes during germination and early growth stages by using polyethylene glycol-6000. Int J Appl Sci Biotechnol. 2024;12(4):194-200. https://doi.org/10.3126/ijasbt.v12i4.73010
8. Reyes JAO, Casas DE, Gandia JL, Parducho MJL, Renovalles EM, Quilloy EP, et al. Polyethylene glycol-induced drought stress screening of selected Philippine high-yielding sugarcane varieties. J Agric Food Res. 2023;14:100676. https://doi.org/10.1016/j.jafr.2023.100676
9. Haghpanah M, Hashemipetroudi S, Arzani A, Araniti F. Drought tolerance in plants: Physiological and molecular responses. Plants. 2024;13(21):2962. https://doi.org/10.3390/plants13212962
10. González-Espíndola M, García-Ruiz H, Acosta-Gallegos JA, Molina-Cano JL, García-Moya E, Terán H, et al. High-throughput phenotyping reveals multiple drought responses of common bean (*Phaseolus vulgaris* L.). Front Plant Sci. 2024;15:1385985. https://doi.org/10.3389/fpls.2024.1385985
11. Bian Z, Wang Y, Zhang Y, Wang Y, Li T. A transcriptome analysis revealing the new insight of green light-induced drought tolerance in tomato seedlings. Front Plant Sci. 2021;12:649283. https://doi.org/10.3389/fpls.2021.649283
12. Sánchez-Reinoso AD, Ligarreto-Moreno GA, Restrepo-Díaz H. Evaluation of drought indices to identify tolerant genotypes in bush bean (*Phaseolus vulgaris* L.). J Integr Agric. 2019;18(2):391-401. https://doi.org/10.1016/S2095-3119(19)62620-1
13. Jarin AS, Islam MM, Rahat A, Ahmed S, Ghosh P, Murata Y. Drought stress tolerance in rice: Physiological and biochemical insights. Int J Plant Biol. 2024;15(3):692-718. https://doi.org/10.3390/ijpb15030051
14. Sruthi SR, Kumar LN, Kishore K, Johnny SI, Anbuselvam Y. Principal component analysis in rice (*Oryza sativa* L.) varieties for three seasons in Annamalai Nagar, an east coast region of Tamil Nadu. Plant Sci Today. 2024;11(4). https://doi.org/10.14719/pst.4105
15. Taiz L, Zeiger E. Plant physiology. 3rd ed. Sunderland, Massachusetts: Sinauer Associates; 2002.
16. Lieth JH, Burger DW. Growth of chrysanthemum plants under various soil water potentials. J Am Soc Hortic Sci. 1989;114(3):387-92.
17. Katsoulas N, Baille A, Kittas C, Rigakis N. Infrared thermometry for early detection of drought stress in chrysanthemum. Acta Hortic. 2006;719:93-100.
18. Marcum KB, Anderson SJ, Engelke MC. Salt gland ion secretion: A salinity tolerance mechanism among five zoysia grass species. Crop Sci. 1998;38:1414. https://doi.org/10.2135/cropsci1998.0011183X003800030031x
19. Barrs HD, Weatherley PE. A re-examination of the relative turgidity technique for estimating water deficit in leaves. Aust J Biol Sci. 1962;15:413-28.
20. Arnon JD. Copper enzymes in isolated chloroplast. Polyphenoloxidase in *Beta vulgaris*. Plant Physiol. 1949;24:1-15.

21. Hiscox JD, Israelstam GF. A method for the extraction of chlorophyll from leaf tissue without maceration. Can J Bot. 1979;57:1332-4.

22. Lepekhov SB. Canopy temperature depression for drought- and heat stress tolerance in wheat breeding. Vavilov J Genet Breed. 2022;26(2):196-201. https://doi.org/10.18699/VJGB-22-24

23. Huang X, Wang J, Wang Q, Hu W, Wang S, Zhou Z. Exploring the potential of chlorophyll a fluorescence for detecting phosphorus status in cotton leaves (*Gossypium hirsutum* L.) to optimize phosphorus management in cotton production. Ind Crops Prod. 2025;225:120561. https://doi.org/10.1016/j.indcrop.2025.120561

24. Protection of Plant Varieties and Farmers' Rights Authority (PPV&FRA). Guidelines for the conduct of test for distinctiveness, uniformity and stability on chrysanthemum. New Delhi: Government of India; 2010. p.1-66.

25. Daler S, Kaya Ö, Cantürk S, Korkmaz N, Kılıç T, Karadağ A, et al. Silicon nanoparticles (SiO_2 NPs) boost drought tolerance in grapevines by enhancing some morphological, physiological and biochemical traits. Plant Mol Biol Rep. 2025;43:1057-75. https://doi.org/10.1007/s11105-024-01520-y

26. Rosmaina R, Ridho A, Zulfahmi Z. Response of morpho-physiological traits to drought stress and screening of curly pepper (*Capsicum annuum*) genotypes for drought tolerance. Biodiversitas. 2022;23:5469-80. https://doi.org/10.13057/biodiv/d231059

27. Toscano S, Romano D. Morphological, physiological and biochemical responses of zinnia to drought stress. Horticulturae. 2021;7(10):362. https://doi.org/10.3390/horticulturae7100362

28. Katuwal KB, Yang H, Huang B. Evaluation of phenotypic and photosynthetic indices to detect water stress in perennial grass species using hyperspectral, multispectral and chlorophyll fluorescence imaging. Grass Res. 2023. https://doi.org/10.48130/GR-2023-0016

29. Sun Z, Yin Y, Zhu W, Zhao Y. Morphological, physiological and biochemical composition of mulberry (*Morus* spp.) under drought stress. Forests. 2023;14(5):949. https://doi.org/10.3390/f14050949

30. Wang F, Chen X, Huang Z, Wei L, Wang J, Wen S, et al. Phenotypic characterization and marker-trait association analysis using SCoT markers in chrysanthemum (*Chrysanthemum morifolium* Ramat.) germplasm. Genes. 2025;16(6):664. https://doi.org/10.3390/genes16060664

31. Kamal S, Rana A, Devi R, Kumar R, Yadav N, Chaudhari AA, et al. Stability assessment of selected chrysanthemum (*Dendranthema grandiflora* Tzvelev) hybrids over the years through AMMI and GGE biplot in the mid hills of North-Western Himalayas. Sci Rep. 2024;14(1):14170. https://doi.org/10.1038/s41598-024-61994-4

32. Ranjan R, Yadav R, Gaikwad K, Kumar M, Kumar N, Babu P, et al. Genetic variability for root traits and its role in adaptation under conservation agriculture in spring wheat. Indian J Genet Plant Breed. 2021;81(1):24-33. https://doi.org/10.31742/IJGPB.81.1.2

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