Exogenous Proline and Moringa Leaf Extracts Improved Drought Resistance in Wheat (*Triticum aestivum* L.) Cultivars

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Abstract

Moringa oleifera leaf extract (MLE) is rich in amino acids, ascorbate, zeatin, minerals, and many other compounds known for their growth-promoting potential, and proline has become a powerful osmolyte. Therefore, a pot experiment was aimed to study the effect of foliar spray with aqueous extracts of *Moringa oleifera* leaves (3%) and proline ($25gL^{-1}$) on some morphological, physiological, biochemical, and yield of five genotypes of wheat (*Triticum aestivum* L.) grown under water stress. The results showed that the exogenous proline and moringa leaf extracts led to increased plant height, total plant fresh weight, total plant dry weight, flag leaf area, total chlorophyll content, relative water content, membrane stability, total phenolic content, antioxidant capacity by DPPH, total soluble sugar, proline content, spike length, spike weight, spiklet per spike, number of grains per spike, grains weight per spike, number of spike per pot, grain yield per pot, biological yield per pot, harvest index with the exception of electrolyte leakage under drought stress. These results demonstrated the importance of foliar application of proline and moringa as tolerance inducers of drought stress leading to improvements in physiological and biochemical attributes in plants grown under the adverse conditions of environmental stresses. According to the results of principal component analysis (PCA), there were significant contribution and strong correlation between studied traits responded by wheat genotypes especially Hasad and Rezan.

Keywords: common wheat, abiotic stressors, osmoprotectants, growth-enhancer, tolerance markers.

Introduction

Wheat (*Triticum aestivum* L.) of the Gramineae family is a common grain crop worldwide. It helps many countries achieve food security. Wheat provides 20% of the protein and 21% of the dietary calories for more than 4.5 billion individuals on a daily basis (30). Wheat stood in third place behind corn and rice in terms of worldwide productivities, with yearly production estimated to reach about 600 million metric tons (31).

Climate change, a sharp rise in population, and various human activities have all led to a significant global decline in the amount of water available for irrigation, specifically for agricultural purposes (37). Drought stress during the growing season, which is a prevalent stress in most arid and semi-arid environments, substantially lowers wheat productivities (24).

Several ways to deal with water shortages have been suggested, but not much has been done to find drought-tolerant genotypes that can be grown in areas that are prone to drought. The greatest strategy to boost wheat yield for genotypes that like drought is to select genotypes that can survive in dry conditions (25). Genotypes should be tested to see how well they can handle drought based on their phenology, morphology, physiology, and biochemical response at different stages of growth.

In response to drought, plants can alter their physiological processes to increase their resistance to the effects of the stressor, including reducing losses in water content (10) ,chlorophyll content 22), stability of membrane (33), production of dry matter (37), accumulation of soluble sugars (23) , proline accumulation (19), and enzymatic and non-enzymatic activities (38) that serve as antioxidants. Therefore, drought-tolerant indices that can distinguish between drought tolerant and susceptible genotypes should be established.

Aside from the ability of plants to use these systems to withstand water deficit conditions, each plant has limit potential. As a result, there is a need to investigate ecologically acceptable ways for improving plant development under stress situations, such as plant-derived biostimulants (32). Furthermore, when drought stress available , the use of biostimulants aids plants in resisting the harmful effects of drought. Plant extracts contain a wide range of phytochemicals, including antioxidants, phytohormones, proteins, fatty acids, vitamins and minerals (32). These chemicals have the ability to boost plant growth and production while also mitigating the detrimental effects of abiotic stressors such a drought (38).

Moringa (*Moringa oleifera* L.) was employed as a powerful biostimulant to increase plant growth and productivities (29). Gibberellins, auxins, and cytokinins are all strong plant growth hormones that can be found in the extract of moringa leaves, along with other beneficial phytochemicals including antioxidants and other minerals (35). Plant stress tolerance is increased as a result of the presence of these chemicals in the plant's metabolism. When sprayed under either normal (36) or abiotic stress conditions (8), the extract from moringa leaves has been shown to boost crop growth and yields.

Proline has become a powerful osmolyte because it gives cells the energy they need to grow and lessens the damage to membranes caused by lipid peroxidation, which is typically increased in stressful conditions (4). Very recently, (20) stated the important roles of exogenous applications of proline in various stress conditions, including drought stress.

Huge research and knowledge gaps exist in wheat genotype evaluation of tolerance markers, phenology, physiology, and biochemistry under water deficit situations. The purpose of this study was to determine the regulating effect of proline and Moringa leaf extract foliar application on five wheat genotypes grown under water deficiency conditions.

Materials and Methods

Plant Materials and growth conditions

In this study, five wheat genotypes (Aras-G1, Ala-G2, Hawler2-G3, Hasad-G4, and Rezan-G5) were gathered from the Agriculture Research, Ministry of Agriculture, Sulaimani, Kurdistan region, Iraq. The experiment was conducted in a greenhouse of the Directorate of Agricultural Research Bakrajo – Sulaimani (latitude 35o 33" N, 45o 27" E, altitude 884.8 masl) during 2021-2022. The experiment was carried out in pots (diameter 30 cm, height 40 cm) filled with 10 kg of soil containing Silty Clay loam in texture, with an EC of 0.25 dSm^{-1}, a pH of 8.15, an organic carbon content of 1.55%, a total nitrogen content of 0.25%, Available Phosphate P of 8.836 $mgkg^{-1}$, a Soluble Potassium K^+ of 4.496 $mgkg^{-1}$, and CaCo3 of 26%. Ten seeds were sown per pot. Soil preparation was proposed according to the needs of soil and crop, with amounts of 5.652 $gpot^{-1}$ of each phosphorus as triple superphosphate and nitrogen as urea (46% N) fertilizer. Soil moisture content measured according to gravimetric method (15) and monitored daily until the imposition of the water restriction using soil moisture meter (Soil Moisture Monitor with Time Display, Model no: WH0291).

Experimental Design and Plant Treatment

The experiment consisted of two groups. The plants in the first group were well-watered (control) spray only with water, and the plants in the second group were stressed. To conduct the study, a factorial completely randomized design (CRD) with two ingredients was applied within three replicates. The first ingredient represents wheat genotypes and the second ingredient represent the treatment groups under stress condition, which consist of T1: untreated plants; T2: treated plants with proline; T3: treated plants with moringa leaf extract; T4: treated plants with both proline and moringa leaf extract. This extract was applied two times by foliar spray during the anthesis of the spikes (anthesis halfway) with one-week intervals.

Moringa Leaf Extract (MLE) and proline solution preparation

MLE was prepared following the method defined by (7). The solution of proline prepared by mixing 25 g of pure proline powder and macerating it in 1 L of distilled water, and then sprayed directly onto each plant by using a hand-sprayer pump for two weeks of foliage application.

Evaluation of Growth Characters

At the end of the stress duration, three plants per treatment used for measuring growth characteristics, such as plant height (cm), total plant fresh weight (g), total plant dry weight (g).

Measurement of Physiological Parameters

For each treatment at the end of stress period, physiological characteristics measured such as Total Chlorophyll Content (mgg^{-1}) using a SPAD-meter.

Flag Leaf Area (cm^2) was estimated following the equation proposed by (3).

$$\text{FLA } (cm^2) = Leaf\ Length * Leaf\ Width * 0.75 \ldots\ldots\ldots\ldots (1)$$

Relative Water Content (RWC) was determined according to (34) for each sample following the formula.

RWC (%) = $[(FW - DW)/(TW - DW)] * 100$ ……………. (2)

Where, FW: flag leaf fresh weight; DW: flag leaf dry weight; TW: flag leaf turgid weight

Relative Electrical Conductivity or Electrolyte Leakage (EL) was calculated by the formula described by (13).

EL (%) = $(EC1/EC2) * 100$ …………… (3)

Where, EC1: initial electrical conductivity of the solution at 32 °C

EC2: final electrical conductivity of the solution at 100 °C

Membrane stability index (MSI) was calculated following the equation proposed by (8).

MSI (%) = $[1 - (EC1/EC2)] * 100$ ………… (4)

Determination of Yield and Yield Component characters:

Yield and yield components were recorded at the harvest time such as Spike Length (cm), Spike Weight (g), No. of Spiklet per Spike, No. of Grain per Spike, Grain Weight per Spike (g), No. of Spike per Pot, Grain Yield per Pot (g), Biological Yield per Pot (g), and Harvest Index (%). The harvest index (HI) was accounted for following (5):

HI = $(Grain\ yield\ /Biological\ yield) * 100$ ……………. (5)

Estimation of Biochemical Attributes

The content of total phenolic compounds (TPC in $\mu g g^{-1}$) in each extract was measured according to (26) using the Folin–Ciocalteu method.

Antioxidant capacity by DPPH (μg $Trolox g^{-1}$ FM), Soluble sugar content (SSC in $\mu g g^{-1}$), and Proline Content in flag leaf samples was determined following the method defined by (26).

Statistical Analysis

XLSTAT version 2019 (Boston, USA) was used to conduct statistical analyses (two-way analysis of variance, Duncan's multiple range test, and principal component analysis (PCA)) for assessing the data obtained in this study at $p \leq 0.05$ was used to compare the means of the data.

Results and Discussion

Table 1 showed the analysis of variance for studied traits. Significant differences were discovered for all studied attributes across all stress treatments. When wheat materials were treated to stress, significant reactions among wheat gynotypes for the investigated parameters were observed. Similar responses were reported from wheat gynotypes in response to the present treatments. Regarding the interactions between gynotypes and characteristics, most traits exhibited no significant alterations, with the exception of the biochemical markers TPC, antioxidant, TSS, and PC, which exhibited significant responses.

Table 1. Variance analysis of all studied traits and it is interaction with five wheat genotypes under the presence of drought stress, foliar application of proline and moringa extracts.

Traits	Genotypes		Treatments		Genotypes*Treatments	
	F	Pr > F	F	Pr > F	F	Pr > F
PH	94.406	< 0.0001	14.568	< 0.0001	2.231	0.016
TPFW	9.287	< 0.0001	32.683	< 0.0001	1.171	0.323
TPDW	15.495	< 0.0001	19.149	< 0.0001	0.819	0.659

FLA	50.111	< 0.0001	30.419	< 0.0001	1.960	0.036
TCC	21.845	< 0.0001	59.308	< 0.0001	2.489	0.007
RWC	12.396	< 0.0001	136.352	< 0.0001	1.962	0.036
EL	24.746	< 0.0001	597.568	< 0.0001	1.848	0.050
MS	24.746	< 0.0001	597.568	< 0.0001	1.848	0.050
TPC	482.254	< 0.0001	3675.185	< 0.0001	102.230	< 0.0001
Antioxidant	68.404	< 0.0001	2391.906	< 0.0001	30.817	< 0.0001
TSS	424.031	< 0.0001	1635.786	< 0.0001	217.320	< 0.0001
PC	492.467	< 0.0001	4611.592	< 0.0001	199.927	< 0.0001
SL	7.087	0.000	13.454	< 0.0001	0.954	0.518
SW	8.646	< 0.0001	25.750	< 0.0001	1.274	0.251
S/S	93.239	< 0.0001	11.029	< 0.0001	1.079	0.399
No. of G/S	18.571	< 0.0001	147.836	< 0.0001	2.748	0.003
No. of S/P	44.929	< 0.0001	93.347	< 0.0001	1.207	0.296
GY/P	36.401	< 0.0001	249.996	< 0.0001	1.709	0.076
BY/P	3.712	0.010	28.508	< 0.0001	0.817	0.661
HI	21.062	< 0.0001	149.803	< 0.0001	1.821	0.054

PH: plant height, TPFW: total plant fresh weight; TPDW: total plant dry weight; FLA: flag leaf area; TCC: total chlorophyll content; RWC: relative water content; EL: electrolyte leakage; MS: membrane stability; TPC: total phenolic content; TSS: total soluble sugar; PC: proline content; SL: spike length; SW: spike weight; S/S: spiklet per spike; No. G/S: number of grain per spike; GW/S: grain weight per spike; No. S/Pot: number of spike per pot; GY/Pot: grain yield per pot; BY/Pot: biological yield per pot; HI: harvest index.

Principal component analysis among measured traits responded by wheat genotypes

Many methods were conducted by researchers to classify the genotypes under particular circumstances, such as stress conditions. Among those methods, multivariate analysis was the most suitable. The main advantage of this analysis is to develop an easier and new model with a smaller number of artificial analyses that account for most of the variance in the data set. In this regard, principal component analysis (PCA), has been used to obtain the quantitative association between morphological, physiological, and biochemical traits responded by wheat genotypes. The data gained from the response of wheat genotypes under presence of stress treatments to assess the involvement of studied traits and their relations to studied genotypes. Regarding the distribution of studied traits on the PCA plot, traits located far away from the scattering point of the plot in the positive patterns of distinct traits demonstrated the best performance under the specific conditions, whereas traits located close to the scattering point of the plot in the negative orientation of studied traits demonstrated poor results.

Based on differences and similarities in the correlation and variance for twenty-one studied traits, four well-defined clusters were detected. The Bi-plot divided studied traits into four separate groups, demonstrating that the outcomes of these traits for different wheat genotypes responses varied from each other. The analysis of PCA showed that the first two components (PC1 and PC2) captured 73.57 % of the total variance. The first major component (PC1) describes 53.46 % of the total variance, while the second component (PC2) describes 20.11%.

The majority of studied traits as shown in figure 1 were grouped in clade 2 and 4, indicating their significant contribution and strong correlation between these traits responded by wheat genotypes especially

G4 in clade 2 and G5 in clade 4. Interestingly, wheat genotypes were placed in different orientations, indicating different performances of these genotypes in terms of responses to physiological, morphological and biochemical traits. In clade 1, PC and FLA alone exhibited great linkage with G3, while G1 and G2 in clade 3 shared similar pattern of responses especially for their strong association with TCC and EL.

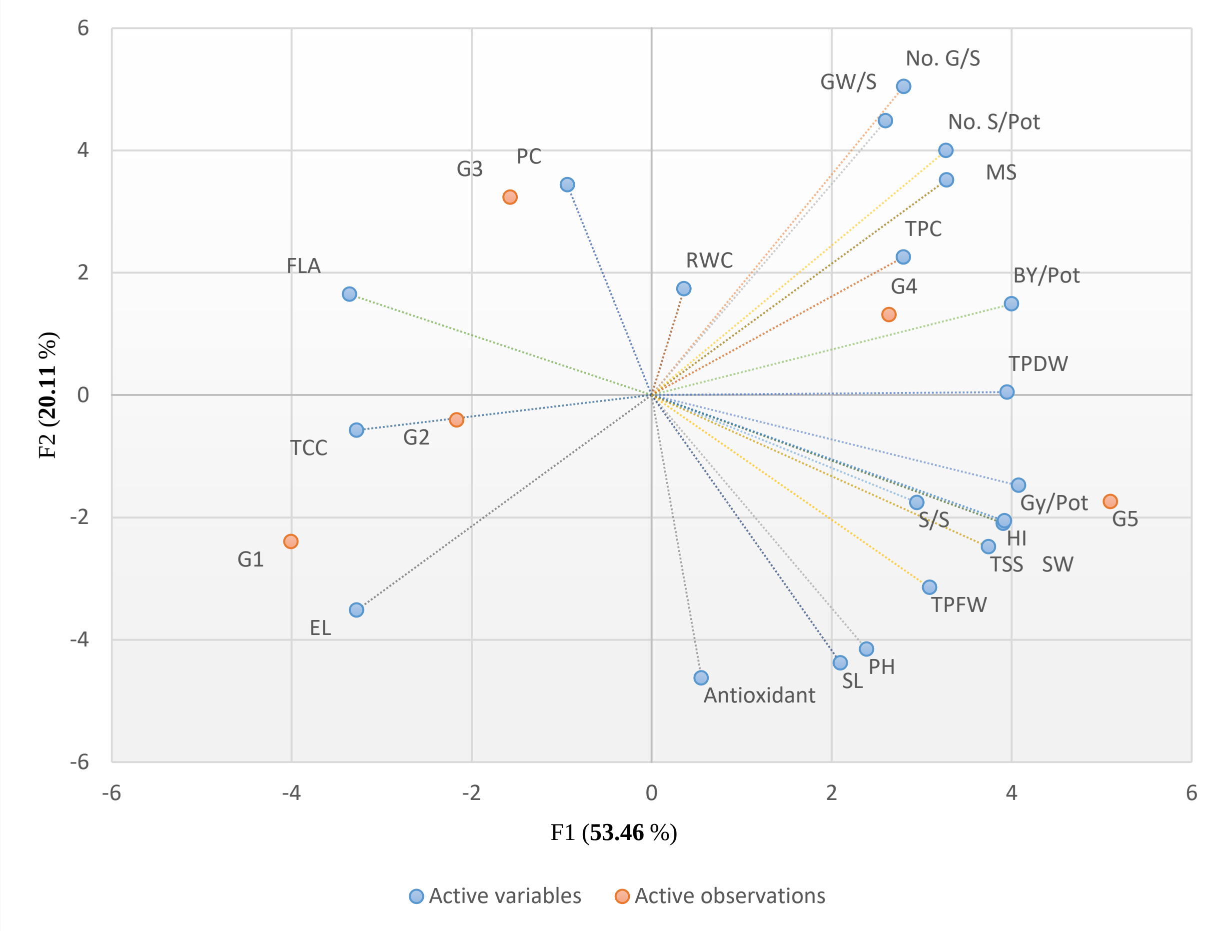

Figure 1. Principal component analysis (PCA) plot for tested traits under all stress treatments responded by five wheat genotypes. The x- and y-axes display the two main principal components of the total variance, 53.46 % and 20.11 %, respectively.

Growth and physiological parameters

Exposure to drought stress decreased PH: plant height, TPFW: total plant fresh weight, FLA: flag leaf area, TCC: total chlorophyll content, RWC: relative water content, MS: membrane stability with the exception of EL: electrolyte leakage, and TPDW: total plant dry weight in respect to control conditions by total wheat cultivars. Wheat genotypes reduced plant height under drought stress while maintaining plant height under the availability of both proline and moringa (T4) to have an almost identical length of 77.493 cm compared to the control condition (C). Noticeable reductions in fresh weight were detected under present drought stress (T1), in which more than 6 g compared to control conditions, while this reduction was decreased under all available applications alone or in combination. A similar response of dry weight accumulation was observed; even more accumulation with a value of 0.605 g compared to the control condition was detected in response to both applications (Table 2). The maximum value of flag leaves was documented under control conditions at 18.025 cm^2, followed by T4 (17.912 cm^2), proline (T2), and Moringa (T3) with values of 15.659 cm^2 and 16.152 cm^2, respectively, while the minimum value of this trait was stated under present drought stress alone with a value of 13.028 cm^2. The content of chlorophyll was sharply reduced by all wheat genotypes to reach about half its value at 12.707 mgg^{-1} compared to the control condition under water stress, while exogenous application of T2, T3, and T4 eliminated this reduction, resulting in values of 18.360, 19.313, and 21.133 mgg^{-1}, respectively. The relative water content ranged between 70.477% and 47.793% for well water (C) and stress conditions (T1), as stated in Table 2. Other important characteristics that were widely used in investigating the performance of studied materials and their response to any available stress conditions were electrolyte leakage and membrane stability. Under control conditions, the leakage from wheat genotypes was 39.001%, while these materials experienced huge leakage under water deficit conditions, reaching 87.141%. However, under the availability of exogenous applications, the electrolyte leakage by wheat genotypes was reduced by almost 27% compared to stress conditions alone. As indicated in Table 2, the maximum stability of the membrane was achieved under control conditions at 60.999%, while poor performance of this trait was noted under stress conditions without exogenous treatments to reach 12.859%. However, both exogenous and combined treatments improved stability, which only around 20% of the reduction compared to untreated circumstances was documented.

Table 2. Effect of exogenous application of growth promoting substances on different growth and physiological parameters of wheat genotypes under stress conditions.

Treatments	PH (cm)	TPFW (g)	TPDW (g)	FLA (cm^2)	TCC (mgg^{-1})	RWC (%)	EL (%)	MS (%)
C	76.800 ab	27.665 a	8.903 b	18.025 a	22.207 a	70.477 a	39.001 c	60.999 a
T1	70.553 d	20.976 c	7.033 c	13.028 c	12.707 c	47.793 e	87.141 a	12.859 c
T2	74.380 c	24.813 b	8.287 b	15.659 b	18.360 b	51.813 d	58.631 b	41.369 b
T3	74.993 bc	24.909 b	8.407 b	16.152 b	19.313 b	56.795 c	59.167 b	40.833 b
T4	77.493 a	27.534 a	9.508 a	17.912 a	21.133 a	59.756 b	59.243 b	40.757 b

C: control (untreated plants under normal condition); T1: untreated plants under stress condition; T2: treated plants with proline; T3: treated plants with moringa leaf extract; T4: treated plants with both proline and moringa leaf extract; PH: plant height, TPFW: total plant fresh weight, TPDW: total plant dry weight; FLA: flag leaf area; TCC: total chlorophyll content; RWC: relative water content; EL: electrolyte leakage; MS: membrane stability.

Biochemical traits

Four essential biochemical traits were investigated in this research, including TPC (µg/g), antioxidant (µg/g), TSS (µg/g), PC (µg/g). Drought stress and exogenous applications significantly ($p < 0.0001$) affected the activities of biochemical traits (Tables 1, 3). In addition, their interactions were also significantly ($p < 0.0001$) enhanced. The data shown in Table 3 demonstrate that the proline and moringa in combinations improved the accumulations in all studied traits: TPC (2.123 μgg^{-1}), antioxidant (1.056 μgg^{-1}), TSS (1.107 μgg^{-1}), PC (6.181μgg^{-1}) compared to the corresponding control with values of (0.968, 0.371, 0.330, 1.011) μgg^{-1} respectively for mentioned traits. Regarding the responses by wheat genotypes under stress conditions without exogenous applications, a similar pattern of accumulation was noted in all studied traits with respect to control conditions, with values of TPC (1.324 μgg^{-1}), antioxidant (0.696 μgg^{-1}), TSS (0.566 μgg^{-1}), PC (1.951μgg^{-1}) Table 3.

Table 3: Effect of exogenous application of growth promoting substances on different biochemical attributes of wheat genotypes under stress conditions.

Treatments	TPC (μgg^{-1})	Antioxidant (μgg^{-1})	TSS (μgg^{-1})	PC (μgg^{-1})
C	0.968 e	0.371 e	0.330 e	1.011 e
T1	1.324 d	0.696 d	0.566 d	1.951 d
T2	1.655 c	0.894 c	0.785 b	5.020 b
T3	1.846 b	1.016 b	0.727 c	3.361 c
T4	2.123 a	1.056 a	1.107 a	6.181 a

C: control (untreated plants under normal condition); T1: untreated plants under stress condition; T2: treated plants with proline; T3: treated plants with moringa leaf extract; T4: treated plants with both proline and moringa leaf extract; TPC: total phenolic content; TSS: total soluble sugar; PC: proline content.

Yield traits under availability of exogenous and stress treatments

Table 4 indicates the yield characteristics of plants grown in both water-stressed and fully irrigated conditions. The treatment of wheat plants with Moringa and proline, even under water stress conditions, successfully increased all studied yield and growth parameters in comparison to the respective control (Tables 1 and 4). This treatment effectively enhanced spike length (8.229 cm), spike weight (0.922 g), spikelet per spike (15.419), number of grains per spike (17.444), grain weight per spike (0.877 g), number of spikes per pot (26.333), grain yield per pot (26.049 g), biological yield (128.837 g), and harvest index (20.207) compared to corresponding stress conditions (T1). The availability of stress conditions without application sharply decreased all assessed growth and yield traits in contrast to fully irrigated conditions. However, applying moringa and proline separately and in combination effectively maintained or even enhanced all measured growth and yield parameters compared to their respective controls. For instance, the application of moringa and proline separately with almost similar values (19.503) significantly ($p \leq 0.001$) increased the harvest index (HI), which is the ratio of total grain to the accumulation of dry matter in shoots, and it is a good indicator for measuring the efficiency and performance of crop production. Similarly, for the rest of the studied parameters, both applications individually enhanced or kept their potentials compared to control conditions, as stated in Table 4.

Table 4. effect of exogenous application of growth promoting substances on yield and its components characters of wheat genotypes under stress conditions

Treatments	SL (cm)	SW (g)	S/S	No. G/S	GW/S (g)	No. S/Pot	GY/Pot (g)	BY/Pot (g)	HI
C	8.182 a	0.922 a	16.217 a	15.933 b	0.895 a	26.200 a	22.953 c	123.210 b	18.666 b
T1	6.938 c	0.395 c	13.820 c	8.800 d	0.119 d	16.733 c	10.910 d	110.350 c	9.830 c
T2	7.741 b	0.785 b	14.999 b	14.111 c	0.756 c	24.200 b	24.178 b	123.954 b	19.503 ab
T3	7.863 ab	0.760 b	15.063 b	16.333 b	0.824 b	25.467 a	24.548 b	125.810 ab	19.531 ab
T4	8.229 a	0.922 a	15.419 b	17.444 a	0.877 a	26.333 a	26.049 a	128.837 a	20.207 a

C: control (untreated plants under normal condition); T1: untreated plants under stress condition; T2: treated plants with proline; T3: treated plants with moringa leaf extract; T4: treated plants with both proline and moringa leaf extract; SL: spike length; SW: spike weight; S/S: spiklet per spike; No. G/S: number of grain per spike; GW/S: grain weight per spike; No. S/Pot: number of spike per pot; GY/Pot: grain yield per pot; BY/Pot: biological yield per pot; HI: harvest index.

Drought is one of the most important non-biological stresses threatening food security since it reduces agricultural output. Drought, which is frequently associated with other major abiotic pressures, has become a difficult issue as a result of climate change, increased demand for water, and declining water supplies (25). Drought stress has an impact on plant performance via physical damage, molecular alterations, biochemical and physiological disturbances. The significant (p < 0.001) decrease in physiological, and yield parameters (Tables 2 and 4) under drought stress conditions (T1) could be caused by the reduction of water absorption from the soil and subsequently to decreased the division of cell and elongation as well as growth of plant (38). The decline in growth and yields characteristics of wheat plants under drought stress circumstances is consistent with the findings of (16). When plants are under stress from drought or other stressors, they boost their production of antioxidant enzymes including; TPC, antioxidant,TSS, and PC to neutralize the reactive oxygen species (ROS) that would otherwise damage them (38).

(20) recenly in their review stated that proline foliar spray has a beneficial effect on plants subjected to drought stress in varies crop species including wheat. Under stress conditions, the amino acid proline has been shown to serve many roles. This biomolecule is crucial because it prevents damage to membranes, proteins, and other cellular components by neutralizing reactive oxygen species (ROS) (4). As a defense mechanism against the many non-living stressors, proline accumulates in high concentration. Proline's dual roles as an osmotic agent and a radical scavenger have helped preserve cells from damages (2).

Exogenous proline increased proline content, total phenolic compound concentration, and antioxidant enzyme activity, all of which lowered stress symptoms. In exchange, healthier plants were produced as shown in Tables (2,3 and 4). The same trend of response similar to our finding was recorded under drought stress in many plants under exogenous application of proline. (17) investigated the performance of two wheat cultivars under drought stress circumstances using three different proline dosage concentrations (50, 100, and 150 mgL^{-1}), finding that the highest dosage was most beneficial in maintaining physiological and yield traits. (2) applied a single foliar dose of 10 mM proline to *Hordeum vulgare* L. (Giza 126) after three weeks of seeding at Kafrelsheikh University in Egypt. They discovered that physiological, biochemical, and yield indices all improved dramatically after receiving this treatment. Possible explanations for proline's beneficial effects on plants include its ability to boost phytochelatins (PCS*)* activity and proline content, the latter of which can function as an antioxidant or osmolyte to mitigate the deleterious effects of abiotic stress (18).

Applying natural biostimulants, such as Moringa leaf extract (MLE), can mitigate the harmful impacts of drought stress on crops (27). For instance, after MLE treatment, the dry grain yield of wheat was considerably higher than under untreated conditions. (12). The phenotypic response and metabolic processes of drought-stressed plants are altered when moringa leaf extract is applied exogenously, enhancing the growth of plant productivity in rice (21), wheat (9), maize (28), and barley (36). Thus, MLE alleviates drought stress effectively, presumably through modifying the morphological and physiological properties of plants.

The present work agreed with (24) that exogenous MLE boosted antioxidant activities (CAT, APX, and SOD), which protect plants from oxidative damage. These enzymes degraded H_2O_2 during drought stress. Because folair contains antioxidants, osmoprotectants, and secondary metabolites, MLE treatment reduces drought's harmful effects by scavenging ROS (40). In addition, the free radical-scavenging capacity of phenolic compounds in MLE confers drought tolerance (14), and increase the level of antioxidant enzymes as stated in the present study (Table 3). As observed by (11), increased drought tolerance in wheat was associated with a higher level of ascorbic acid due to its presence in MLE. By mitigating the deleterious impacts of drought stress, seeds primed with MLE displayed considerably increased activity of both enzymatic and non-enzymatic antioxidants and decreased H_2O_2 concentrations in wheat leaves as stated by (1). In our study, moringa treatment at concentrations slightly lower than those used in different study was effective in reducing ROS-based oxidative damage in wheat (39). This was attributable to the sufficient presence of macro- and micronutrients in their organs (6). In addition, the leaf of moringa species contains flavionoid compounds (35). The system of boosting drought resistance was connected to ROS detoxification due to the action of flavinoid and phenolic as a singlet oxygen quencher and hydrogen donor (38).

Conclusion

In conclusion, the objective of this study was to examine the impact of proline and Moringa plant extracts on the drought tolerance of wheat plants. In comparison to well-hydrated control plants, wheat plants that were subjected to stress exhibited considerably inferior growth, biochemical, and yield characteristics. Despite this, foliar application of proline and moringa, either individually or in combination, improved all of the studied characteristics. Significantly, it was discovered that drought resistance in wheat cultivars was connected with rapid cell reinforcements. This was mostly related to the wheat cultivar's intrinsic potential to improve detoxifying activity against reactive oxygen species via the use of MLE and proline. In this respect, both applications can be utilized effectively for sustainable agriculture in regions where drought conditions exist.

Conflict of interest

The authors declare no conflict of interest.

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