Investigating the relationship between the activities of antioxidant enzymes and genetic Parameters under drought stress conditions of durum wheat genotypes (*Triticum durum* Desf).

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Abstract

Drought stress and limited water at the reproductive stage (terminal phase of wheat growth) significantly impact wheat productivity. This study examines terminal drought effects on water relations in wheat. Under drought stress, plants experience hyperosmotic stress at the cellular level, leading to ROS accumulation and oxidative stress, which can impair cell integrity. Plants counter this oxidative damage with a range of enzymatic and non-enzymatic antioxidants to maintain ROS balance. An experiment was conducted using a randomized complete block design with three replicates in both stressed and non-stressed conditions across six durum wheat (*Triticum durum*) cultivars. Combining abilities for yield, drought tolerance, and quality traits were estimated using a 6 x 6 half-diallel mating design. F1 seeds and parental varieties were evaluated under terminal drought stress, measuring antioxidant enzymes (catalase, ascorbate peroxidase, superoxide dismutase) and grain yield. Results showed that the additive-dominance model was significant for all enzyme traits at the 1% level, with both additive and dominance effects observed. Broad-sense heritability (GCA) was significant at 1% for antioxidant traits and at 5% for catalase; narrow-sense heritability (SCA) was significant at 1% for grain yield. Correlation analysis revealed that antioxidant factors and grain yield were significantly correlated at $p < 1\%$ under drought. In conclusion, drought stress enhances the activity of antioxidant enzymes, which plays a crucial role in maintaining yield in *durum wheat* under terminal drought conditions. Antioxidant enzyme activity is a useful screening tool for drought-resistant genotypes.

Keywords: Diallel cross, GCA, SCA, antioxidants enzymes, correlation analysis, drought stress.

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Introduction

Durum wheat (*Triticum turgidum* L. subsp. *durum* Desf.) is one of the most commonly cultivated wheat species worldwide, accounting for nearly 5% of global wheat production (Colasuonno et al.,

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2021). The EU is the primary producer, with Italy leading in European *durum wheat* production, followed by France and Turkey. *Durum wheat* is also cultivated in the northern United States, Canada, Mexico, and sub-Saharan Africa, with most of its production occurring in countries around the Mediterranean basin. Other smaller cultivation areas include Russia, Kazakhstan, Australia, India, and Argentina. This species is a key commodity globally, as its grain is used to produce many foods, with significant variation across producing countries (Laus et al., 2021). Pasta is the most popular *durum*-based product worldwide, while couscous and bulgur are common in North Africa and the Middle East, respectively. *Durum wheat* bread is also traditionally important in Southern Italy, Spain, Turkey, and other Mediterranean regions.

Durum wheat grain is a valuable food source with a significant role in the human diet, providing carbohydrates (70.2%), proteins (12.2%), lipids (1.9%), fiber (1.6%), and minerals (1.6%). Whole grains also contain bioactive compounds beneficial for health, including high levels of antioxidants such as carotenoids and polyphenols, along with vitamins, sodium, potassium, calcium, and magnesium. In Mediterranean regions, *durum wheat* is mainly grown under rain-fed conditions, often encountering drought stress that hampers yield (Marcotuli et al., 2020). Recent studies show complex interactions between membrane transport proteins and ROS in plants, where ROS signaling can activate membrane transporters for substance transport (Li et al., 2021). This transport promotes metabolic responses that enhance antioxidant enzyme activity, scavenge excess ROS, and improve stress tolerance (Yuan et al., 2024).

Under abiotic stress, ROS accumulate in various forms ($^1O_2$, $O_2^{\bullet-}$, H_2O_2, and $\bullet OH$) in the cytosol and plant organelles. Excess ROS can disrupt cell homeostasis, damage lipids and DNA, and lead to cell apoptosis. Reducing excess ROS is crucial for stress resistance, achieved by inhibiting ROS production and promoting ROS catabolism. ROS signaling activates membrane transporters, creating a complex system network (Mittler et al., 2022). Abiotic stresses disrupt cellular biochemistry, producing excessive ROS (Apel and Hirt, 2004). ROS play dual roles in plant growth, acting as signaling molecules and potential oxidative agents that damage plant tissues (Mishra et al., 2023).

Changes in antioxidant enzyme expression or activity have been noted in response to oxidative stress across species (Rajput et al., 2021). Overexpression of antioxidant enzymes is a potential stress biomarker, with enhanced enzyme activity linked to improved stress tolerance (Dumanović et al., 2021). *Durum wheat* plants with varying drought tolerance exhibit different antioxidant enzyme activities and molecule levels under stress. A pattern of changes in antioxidant enzymes under hyperosmotic stress has been suggested for both sensitive and tolerant *durum wheat* genotypes (Ami et al., 2020; Huseynova et al., 2014; Huseynova et al., 2015; Sheng et al., 2020). The correlation between catalase (CAT), superoxide dismutase (SOD), ascorbate peroxidase (APX) activities, root and shoot dry weights, and stress tolerance index suggests that biomass and enzymatic antioxidant parameters could be reliable markers for screening wheat genotypes for water stress tolerance during early growth stages (Pour-Aboughadareh et al., 2020).

Alterations in SOD, APX, and CAT activity and ROS concentration have been observed in wheat plants in both field and laboratory (Huseynova et al., 2014; Mishra et al., 2013; Samineni et al., 2022).

The aim of this study was to determine antioxidant enzyme activity in *durum wheat* genotypes under drought conditions and examine their relationship with productivity.

Materials and Methods

Experiments were carried out using a randomized complete block design with three replications and two conditions: water stress and normal, conducted at Islamic Azad University, Ardabil Branch. The experiment was laid out with six varieties of *durum wheat*, including Fadda, Yavarus, Altar, Omrabi5, Zardak, and Korifera. Measurements were taken 12 days after applying drought stress, selecting about 10 flag leaves per

Table 1
Analysis of varians in normal irrigation for grain 6 durum cultivars and antioxidant enzymes attributes.

S.O.V	df	Ascorbate Peroxidase (mM/gr fw)	catalase (mM/gr fw)	Superoxide Dismutase (Unit/gr fw)	Grain Yield (Kg/hectar)
Replication	2	0.076 Ns	0.095 Ns	0.075 Ns	131.48 Ns
cultivars	5	1.5 Ns	0.083 Ns	0.226 Ns	12.3 Ns
Error	10	-	-	-	-

Ns: non Significant   mM/g fW= milimolar per resh wiegh of tissue   Unit/g FW=Enzyme activity per fresh wiegh

Table 2
Analysis of varians in dry Stress for grain 6 durum cultivars and antioxidant enzymes attributes.

S.O.V	df	Ascorbate Peroxidase (mM/gr fw)	Catalase (mM/gr fw)	Superoxide Dismutase (Unir/gr fw)	Grain Yield (Kg/hectar)
Replication	2	.0005 Ns	0.568 Ns	0.72*	29.85 Ns
cultivars	5	0.087**	7.743**	1.62**	537**
Error	10	-	-	-	-

NS: non-Significant   **: Significant at $p < 0.01$ *: Significant at $p < 0.05$
mM/g FW= milimolar per fresh wiegh of tissue ,   Unit/g FW=Enzyme activity per g fresh wiegh

Table 3
Combined analysis of variance for antioxidant enzymes attributes in 6 durum wheat.

S.O.V	df	Ascorbate Peroxidase (mM/gr fw)	Catalase (mM/gr fw)	Superoxide Dismutase (Unir/gr fw)	Grain Yield (Kg/hectar)
Environment (E)	1	0.017**	6.59**	0.983 Ns	1406**
Error1	4	0.0002	0.32	0.4	80
genotype (A)	5	0.56**	4.43**	1.25**	1719**
E×A	5	0.034**	3.39**	0.619*	67 Ns
Error2	20	0.001	0.356	0.207	178
Total	35	-	-	-	-

Ns: non-Significant   **: Significant at $p < 0.01$ *: Significant at $p < 0.05$
mM/g FW= milimolar per g fresh wiegh of tissue , Unit/g FW=Enzyme activity per g fresh wiegh

genotype and calculating their average. All agricultural operations were performed uniformly under both stress and normal conditions; for drought stress, the final stage of irrigation was omitted. Leaves collected from plants grown under stress were transferred to the laboratory, frozen with liquid nitrogen, ground in a mortar, and stored in a refrigerator.

Enzyme activity for ascorbate peroxidase was measured using the Sayram method (Naz et al., 2022; Sairam et al., 1998), while catalase and superoxide dismutase activity were assessed by the methods of Gyannopolities and Ries (1977), with catalase extracted using the Chance and Maehly method (1955). The units for ascorbate peroxidase and catalase activity were expressed as millimoles per gram of fresh weight of *durum wheat* tissue, while superoxide dismutase activity was calculated per gram of tissue. Statistical analysis using diallel analysis was performed with Griffing's method (Griffing, 1956), and genetic parameters for the half diallel cross were estimated using the Singh method ((Singh and Singh, 1984). Broad-sense heritability (H^2b), narrow-sense heritability (H^2n), and average degree of dominance (D) were calculated according to Mather and Jinks (1971).

The diallel cross provided valuable information. Antioxidant enzymes are regarded as indicators of drought tolerance in wheat, but few studies have examined their genetic basis (Chaudhari et al., 2023; Hussein and Zaater, 2024; Seied-Khamesi et al., 2022; Shahbazi et al., 2018). Common statistical software, including SPSS, MSTATC, SAS, EXCEL, and Dial98, was used for analysis of variance and mean comparisons in this project.

Table 4
Means comparison of studied traits in 6 durum wheat cultivars.

S.O.V	Omrabi5	Altar	Yavarus	Fadda	Korifla	Zardak
APO	0.497a	0.138b	0.104b	0.020c	0.095b	0.102b
CAT	5.76a	2.91bc	3.18b	0.87e	2.21d	2.66c
SOD	7.77a	3.9b	4.07b	0.26d	2.53c	4.54b
GY	2903a	2249b	1035d	435e	1801c	448e

Ascorbate peroxidase (APO), Catalase (CAT), Superoxide dismutase (SOD), grain yield kg/hectare(GY)

Table 5
Correlation Analysis by Pearson method in Normal Condition.

	Ascorbate peroxidase	Catalase	Superoxide dismutase	Grain Yeild
Ascorbate peroxidase activity	1			
Catalase activity	-0.208 Sig 0.408	1		
Superoxide dismutase activity	-0.009 Sig 0.973	0 .423 Sig 0.080	1	
Grain yeild	0.233 Sig 0.353	0.098 Sig 0.699	0.451 Sig 0.060	1

** Correlation is Significant at $p < 0.01$
* Correlation is Significant at $p < 0.05$

Table 6
Correlation Analysis by Pearson method in Stress Condition.

	Ascorbate peroxidase	Catalase	Superoxide dismutase	Grain Yeild
Ascorbate peroxidase activity	1			
Catalase activity	0.861** Sig .000	1		
Superoxide dismutase activity	0.692** Sig .001	0.793** Sig .000	1	
Grain yeild	0.674** Sig .002	0.588* Sig .010	0.414* Sig .057	1

** Correlation is Significant at $p < 0.01$
* Correlation is Significant at $p < 0.05$

Results

According to variance analysis, drought stress showed a significant difference (P<1%), while under normal conditions, genotype effects were not significant (Table 1). This finding indicates that water stress conditions had a significant impact on genotype performance. Block effects were not significant, showing that the blocks in the experiment had balanced yield outcomes (Table 2). Combined analysis revealed significant differences (P<1%) in yield across different cultivars (Table 3). Yield, superoxide dismutase, catalase, and ascorbate peroxidase showed significant effects at the 1% level for genotype effects, indicating that antioxidant activity exhibited varied responses in *durum wheat* varieties. All sources of variation, including superoxide dismutase, catalase, and ascorbate peroxidase, showed significant interaction effects between genotype and environment (Table 3).

Therefore, Duncan's test was used to achieve detailed results in examining cultivar yield. The *Omrabi-5 durum wheat* cultivar was in Group A under drought conditions, while other cultivars exhibited different yields under these conditions (Table 4). Therefore, Omrabi-5 was identified as the most resistant variety in drought conditions, while Fadda was recognized as the most sensitive. In the correlation analysis of grain yield characteristics and antioxidant enzymes using Pearson's method, under normal conditions, enzyme activity did not show a positive, significant

Table 7
Estimates of genetic parameters in half-diallel.

parameters	Ascorbic acid	Super oxidase	Catalase	Grain yeild
D	396±0.005**	4.101563±0.326**	66.25±0.045**	1.6E-06±1377ns
H1	250±0.005**	5.660377±0.535**	44.16667±0.062**	6.16E-07±1201ns
H2	293.75±0.004**	7.679827±0.431**	101.1111±0.037**	1.1E-06±9358ns
F	285.7143±0.007**	1.859504±0.447 ns	41.66667±0.065**	4.38E-06±1492**
Sqr(H1/D)	0.952	1.92	1.22	0.542
(kd+kr) kd	0.8708	0.61	0.788	0.648
D/(D+E)	0.793	0.709	0.85	0.271
h2b	0.33	0.728	0.64	0.841
h2n	0.023	0.18	0.22	0.683
Mp	0.141	2.66	2.11	0.549
Mf1	0.074	2.29	2.06	2831
Vp	0.0125	0.606	0.125	3599828
Vf1	0.0002	0.485	0.027	10503046
Ep	13800±0.001**	39.6875±0.081**	281.25±0.008**	6.04E-05±1054**
Ef1	1600±0.026**	151.1111±0.032**	18888.89±0.003**	5.07E-05±1054**

**: Significant at $p < 0.01$ probability levels, respectively; D: Additive variance, H1: Uncorrected dominance variances, H2: Corrected dominance variances, F: Additive-dominance covariance, Sqr(H1/D): Average degree of dominance, (kd+kr)kd: Proportion of dominant genes, D/(D+E): Heritability by parents, H2b: Heritability for diallel in a broad sense, H2n: Heritability for diallel in a narrow sense, Mp: Mean of Parents, Mf1: Mean of F1s, Vp: Var. of Parents, Vf1: Var. of F1s, Ep: Env. Var. from Parents, Ef1: Env. Var. from F1s

relationship with grain yield (Table 5). However, in drought conditions, enzyme activity showed a positive, significant correlation with grain yield at the 1% and 5% probability levels (Table 6). The results demonstrated that antioxidant enzyme activity increased in dry conditions, showing a positive, significant relationship with grain yield. In particular, catalase and ascorbate peroxidase activities showed a significant correlation with grain yield at a probability level below 1%, and superoxide dismutase showed significance at a 5% level in dry conditions (Table 6). The variation in antioxidant enzyme activity among different genotypes was linked to different yields in drought stress conditions.

Additive variance parameters (D) and corrected dominance variance (H1) as well as uncorrected dominance variance (H2) were significant at the 1% level in antioxidant enzyme activity, including superoxide dismutase and ascorbate peroxidase (Table 7). The additive-dominance covariance index showed a significant effect at the 1% level for superoxide dismutase, ascorbate peroxidase, and grain yield. Environmental variance parameters of the parents (Ep) were significant for all traits at the 1% probability level (Table 7). Heritability for diallel in a broad sense (GCA) and in a narrow sense (SCA) was calculated using Griffing’s method (Table 8). Broad-sense heritability (GCA) was significant at 1% for antioxidant enzymes, superoxide dismutase, and ascorbate peroxidase, and at the 5% level for catalase activity. Narrow-sense heritability (SCA) was significant at 1**%** for grain yield.

Discussion

Based on the results, it was inferred that the additive-dominance model can adequately fit most characters. In the study by Seied-Khamesi and coworkers (Seied-Khamesi et al., 2022), additive genetic effects played a significant role in phenotypic variations of the bread wheat genotypes. In wheat plants, both SCA and GCA effects were significant for stem reserve remobilization (Hussein and Zaater, 2024). Generally, the traits showed high antioxidant enzyme levels, indicating that most of the

Table 8
General combining ability (GCA) and specific combining ability (SCA) mean squares for evaluated traits from a 6 × 6 half-diallel cross in stress conditions durum wheat genotypes.

Sourse	Grain yeild	Catalase	Super oxidase	Ascorbic acid
GCA	4457558ns	8.93*	25**	0.10**
SCA	2261185**	1. 56ns	2.6ns	0.01ns
Error	683422ns	0.93ns	21.16ns	0.01ns

*, **: Significant at $p < 0.05$ and $p < 0.01$ percent probability levels, respectively; GCA: General combining ability, SCA: Specific combining ability

variation among genotypes is genetic rather than environmental. Shabazi and coworkers (2018) also found high drought resistance among barley cultivars in a half-diallel cross in wheat.

The magnitude of the average degree of dominance indicated that, for plant resistance to drought, additive effects are more significant. Grain yield was significant at the 1% probability level for narrow-sense heritability (SCA), which showed the greater role of dominance effect in controlling drought resistance parameters. In Griffing's model, based on diallel in a broad sense (GCA), grain yield was not significant, indicating a greater role of additive gene effects in controlling traits. Based on this study's findings, low heterosis was estimated across all traits, which is typical in self-pollinating crops. Finally, based on GCA effects, the parents ((Hussein and Zaater, 2024; Seied-Khamesi et al., 2022; Shahbazi et al., 2018) possessed favorable alleles for drought resistance in wheat plants. These parents may produce offspring with improved genotypes under terminal drought conditions, and the findings showed the dominance effects higher influence on trait control.

The results showed that increased CAT, SOD, and ascorbate peroxidase activities were observed under drought stress, with this enhancement attributed to the antioxidative defense mechanisms contributing to yield. Mehdy (1994) found that intracellular free radical generation and drought stress were main factors causing plant damage. Some researchers believed that antioxidant enzyme activity increased in durum wheat under drought stress (Ami, et al. 2020). In this experiment, CAT, SOD, and ascorbate peroxidase activities in the drought-sensitive variety started to decrease at the end of the season under drought stress. We hypothesized that these enzyme activities represent a response mechanism of *durum wheat* cultivars to reduce H_2O_2 levels under drought stress during the final irrigation stage.

The results showed that in drought stress conditions during the last growth stage, antioxidant enzyme activity significantly increases. In drought-resistant genotypes, this enzyme activity increases under drought conditions, resulting in less yield reduction compared to drought-sensitive cultivars, where yield decreases more sharply in the final stage. The relationship between enzyme activity and the antioxidant defense system in drought conditions showed a positive and significant correlation in this research. The grain yield reduction in drought-resistant varieties was less than that in sensitive varieties under drought conditions. For *durum wheat* varieties subjected to drought in the last stage, the antioxidant defense mechanism quickly mitigates oxygen free radicals. The peak H_2O_2 accumulation was significantly higher in drought-sensitive cultivars than in drought-tolerant ones. In this study, we observed that enzyme activity in resistant genotypes peaked 12 days after drought stress, then rapidly declined, indicating that enzyme activity is regulated by genotype and environment. Under normal conditions, enzyme content did not significantly decrease.

Therefore, we hypothesized that the stress response for eliminating H_2O_2 achieves a dynamic balance between consumption and synthesis at this time. The likely reason was that the higher H_2O_2 levels required higher CAT, SOD, and ascorbate peroxidase activity. Maintaining a high antioxidant enzyme content under severe drought likely contributes to drought resistance. Maintaining high antioxidant capacity to scavenge ROS is associated with increased plant tolerance to various environmental stresses, which our research supports. Antioxidant enzyme activity can be a screening tool for drought-resistant genotypes in *durum wheat* plants. Our research results align with most studies (El Haddad et al., 2021; Laus et al., 2021; Mishra et al., 2023; Qayyum et al., 2021; Quagliata et al., 2023; Yuan et al., 2024).

Conclusion

Our results indicate that distinct metabolic responses in *durum wheat* are related to its genetic potential for yield, leading to improved tolerance against drought. The distinct induction of defense system activity under drought conditions was accompanied by a significant decrease in damage indices. Therefore, the degree of tolerance is a specific trait under drought stress, showing a positive and significant correlation between antioxidant activity in the present research. Under drought stress, unlike the continuous increase in antioxidant activity in durum cultivars, its relative increase in the tolerant cultivar did not cause cell damage. Instead, it likely acted as a factor inhibiting the effects of drought stress. The increase in ROS content may affect the tolerance level of the genotypes, so we measured antioxidant activity as one of the important parameters to determine drought tolerance.

According to the results of performance reviews and related traits under both water stress and normal conditions, as well as consideration of testing and variance analysis in correlation design, and review of treatments under water stress and normal conditions with respect to genotype effects, significant effects were observed in both genotype and interaction effects. We conclude that the present findings are consistent with the results of most studies. It was concluded that drought stress plays an effective role in the activity of Superoxide Dismutase, Catalase, and Ascorbate Peroxidase antioxidant enzymes. The activity of these antioxidant enzymes plays a vital role in preventing yield reduction in *durum wheat* genotypes under end-of-season drought conditions. Antioxidant enzyme activity can therefore be used as a screening tool for drought-resistant genotypes of *durum wheat* in plant breeding programs.

References

Ami, K., S. Planchais, C. Cabassa, A. Guivarc'h, A.-A. Very, M. Khelifi, R. Djebbar, O. Abrous-Belbachir and P. Carol. 2020. Different proline responses of two Algerian durum wheat cultivars to in vitro salt stress. *Acta physiologiae plantarum,* 42, (2) 21.

Apel, K. and H. Hirt. 2004. Reactive oxygen species: metabolism, oxidative stress, and signal transduction. *Annu Rev Plant Biol,* 55, (1) 373-399.

Change, B. and A. Maehly. 1955. Assay of catalases and peroxidase. *Methods Enzymol,* 2, 764-775.

Chaudhari, G. R., D. Patel, A. Kalola and S. Kumar. 2023. Use of graphical and numerical approaches for diallel analysis of grain yield and its attributes in bread wheat (*Triticum aestivum* L.) under varying environmental conditions. *Agriculture,* 13, (1) 171.

Colasuonno, P., I. Marcotuli, A. Gadaleta and J. M. Soriano. 2021. From genetic maps to QTL cloning: an overview for durum wheat. *Plants,* 10, (2) 315.

Dumanović, J., E. Nepovimova, M. Natić, K. Kuča and V. Jaćević. 2021. The significance of reactive oxygen species and antioxidant defense system in plants: A concise overview. *Frontiers in plant science,* 11, 552969.

El Haddad, N., H. Choukri, M. E. Ghanem, A. Smouni, R. Mentag, K. Rajendran, K. Hejjaoui, F. Maalouf and S. Kumar. 2021. High-temperature and drought stress effects on growth, yield and nutritional quality with transpiration response to vapor pressure deficit in lentil. *Plants,* 11, (1) 95.

Giannopolitis, C. N. and S. K. Ries. 1977. Superoxide dismutases: I. Occurrence in higher plants. *Plant physiology,* 59, (2) 309-314.

Griffing, B. 1956. Concept of general and specific combining ability in relation to diallel crossing systems. *Australian journal of biological sciences,* 9, (4) 463-493.

Huseynova, I. M., D. R. Aliyeva and J. A. Aliyev. 2014. Subcellular localization and responses of superoxide dismutase isoforms in local wheat varieties subjected to continuous soil drought. *Plant Physiology and Biochemistry,* 81, 54-60.

Huseynova, I. M., D. R. Aliyeva, A. C. Mammadov and J. A. Aliyev. 2015. Hydrogen peroxide generation and antioxidant enzyme activities in the leaves and roots of wheat cultivars subjected to long-term soil drought stress. *Photosynthesis research,* 125, 279-289.

Hussein, M. and M. M. Zaater. 2024. Estimates of genetic parameters, combining abilities and heterosis for some genotypes of bread wheat. *Journal of Agricultural Chemistry and Biotechnology,* 15, (7) 87-92.

Laus, M. N., M. A. De Santis, Z. Flagella and M. Soccio. 2021. Changes in antioxidant defence system in durum wheat under hyperosmotic stress: A concise overview. *Plants,* 11, (1) 98.

Li, H., C. Testerink and Y. Zhang. 2021. How roots and shoots communicate through stressful times. *Trends in plant science,* 26, (9) 940-952.

Marcotuli, I., P. Colasuonno, Y. S. Hsieh, G. B. Fincher and A. Gadaleta. 2020. Non-starch polysaccharides in durum wheat: a review. *International journal of molecular sciences,* 21, (8) 2933.

Mather, K. and J. Jinks. 1971. Biometrical Genetics. Chapman and Hall Ltd. *New Fetter Lane,* 14-19.

Mehdy, M. C. 1994. Active oxygen species in plant defense against pathogens. *Plant physiology,* 105, (2) 467.

Mishra, A. K., R. Rai and S. Agrawal. 2013. Individual and interactive effects of elevated carbon dioxide and ozone on tropical wheat (*Triticum aestivum* L.) cultivars with special emphasis on ROS generation and activation of antioxidant defence system. *Indian J Biochem Biophys,* 50, (2) 139-149.

Mishra, N., C. Jiang, L. Chen, A. Paul, A. Chatterjee and G. Shen. 2023. Achieving abiotic stress tolerance in plants through antioxidative defense mechanisms. *Frontiers in Plant Science,* 14, 1110622.

Mittler, R., S. I. Zandalinas, Y. Fichman and F. Van Breusegem. 2022. Reactive oxygen species signalling in plant stress responses. *Nature reviews Molecular cell biology,* 23, (10) 663-679.

Naz, R., F. Gul, S. Zahoor, A. Nosheen, H. Yasmin, R. Keyani, M. Shahid, M. Hassan, M. Siddiqui and S. Batool. 2022. Interactive effects of hydrogen sulphide and silicon enhance drought and heat tolerance by modulating hormones, antioxidant defence enzymes and redox status in barley (*Hordeum vulgare* L.). *Plant Biology,* 24, (4) 684-696.

Pour-Aboughadareh, A., A. Etminan, M. Abdelrahman, K. H. Siddique and L.-S. P. Tran. 2020. Assessment of biochemical and physiological parameters of durum wheat genotypes at the seedling stage during polyethylene glycol-induced water stress. *Plant Growth Regulation,* 92, (1) 81-93.

Qayyum, A., S. Al Ayoubi, A. Sher, Y. Bibi, S. Ahmad, Z. Shen and M. A. Jenks. 2021. Improvement in drought tolerance in bread wheat is related to an improvement in osmolyte production, antioxidant enzyme activities, and gaseous exchange. *Saudi Journal of Biological Sciences,* 28, (9) 5238-5249.

Quagliata, G., S. Abdirad, S. Celletti, F. Sestili and S. Astolfi. 2023. Screening of *Triticum turgidum* genotypes for tolerance to drought stress. *Plant Physiology and Biochemistry,* 194, 271-280.

Rajput, V. D., Harish, R. K. Singh, K. K. Verma, L. Sharma, F. R. Quiroz-Figueroa, M. Meena, V. S. Gour, T. Minkina and S. Sushkova. 2021. Recent developments in enzymatic antioxidant defence mechanism in plants with special reference to abiotic stress. *Biology,* 10, (4) 267.

Sairam, R., P. Deshmukh and D. Saxena. 1998. Role of antioxidant systems in wheat genotypes tolerance to water stress. *Biologia plantarum,* 41, 387-394.

Samineni, S., M. D. Mahendrakar, A. Hotti, U. Chand, A. Rathore and P. M. Gaur. 2022. Impact of heat and drought stresses on grain nutrient content in chickpea: Genome-wide marker-trait associations for protein, Fe and Zn. *Environmental and Experimental Botany,* 194, 104688.

Seied-Khamesi, M., V. Rashidi, H. Shahbazi, E. Khalilvand Behrouzyar and B. Mirshekari. 2022. Genetics of grain filling rate and remobilization of stem reserves in bread wheat under terminal drought stress. *Journal of Plant Physiology and Breeding,* 12, (1) 153-163.

Shahbazi, H., H. Bigonah and M. Alaei. 2018. Genetics and heritability of some physiological and agronomic traits in barley under drought stress. *Journal of Plant Physiology and Breeding,* 8, (2) 13-23.

Sheng, H., J. Zeng, Y. Liu, X. Wang, Y. Wang, H. Kang, X. Fan, L. Sha, H. Zhang and Y. Zhou. 2020. Differential responses of two wheat varieties differing in salt tolerance to the combined stress of Mn and salinity. *Journal of plant growth regulation,* 39, 795-808.

Singh, M. and R. Singh. 1984. A comparison of different methods of half-diallel analysis. *Theoretical and Applied Genetics,* 67, 323-326.

Yuan, D., X. Wu, X. Jiang, B. Gong and H. Gao. 2024. Types of membrane transporters and the mechanisms of interaction between them and reactive oxygen species in plants. *Antioxidants,* 13, (2) 221.