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Changes of Oxidative Status of *Triticum aestivum* L. during Germination and Initial Phase of Growth Subjected to Abiotic Stress

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Abstract: ***Triticum aestivum is one of the most commonly cultivated species in the world and represents a key commodity for many areas worldwide, as its grain is used in the production of various foods and has a relevant role in the human diet, providing carbohydrates, proteins, vitamins, etc. as well as highly valued bioactive compounds. It is largely cultivated in the Mediterranean basin, where it is mainly grown under rain - fed conditions, thus currently undergoing abiotic stress, which can hamper yield potential and influence the qualitative characteristics of grain. When plants suffer abiotic stress, damage is established at the cellular level. This leads to the accumulation of ROS thus generating in turn an oxidative stress condition, which can ultimately result in the impairment of cellular integrity and functionality. This study investigates oxidative stress responses in Triticum aestivum L. during germination and early growth when subjected to abiotic stresses, including drought and salinity. Key biochemical markers such as malondialdehyde (MDA), proline, peroxide, soluble protein, and catalase activity were measured. Results indicated significant increases in oxidative stress indicators under both water and salt stress, suggesting severe impairment of cellular function. The findings highlight the plant's adaptive mechanisms and call for further exploration into enzyme behaviour under stress to improve wheat resilience.***

Keywords: abiotic stress, wheat, salinity, drought, oxidative stress

1. Introduction

One of the most important cereal crops for the world's food supply is wheat (*Triticum aestivum* L.). Abiotic stresses, such as drought, salt, very high or low temperatures, and heavy metal pollution, frequently reduce its production. Photosynthesis, biomass, germination, seedling growth, and yield are all adversely affected by these stresses [1]. In light of growing worries regarding climate change and environmental degradation, it is critical to comprehend how abiotic pressures affect wheat in order to create robust cultivars and guarantee sustainable farming methods [2]. A complicated physiological process, germination is extremely susceptible to environmental factors. Abiotic stresses interfere with germination - related metabolic processes, enzyme activation, and water absorption [3]. After germination, seedling establishment is also vulnerable to stresses. Water supply is restricted by drought stress, which prevents seeds from imbibing and delays germination. It decreases radicle emergence and modifies the hydrolysis of food reserves that have been accumulated [4]. It decreases total biomass, root elongation, and leaf growth in seedlings. Reactive oxygen species (ROS) build up as a result of drought - induced oxidative stress, harming cellular components and preventing photosynthesis. Osmotic and ionic stress caused by salinity interferes with water balance and nutrient intake. Root - shoot growth and seed germination are hampered by elevated Na^+ and Cl^- concentrations [5]. It disrupts cell division and elongation, lowers enzymatic activity, and induces membrane instability. Additionally, salinity changes hormonal signalling pathways including ethylene and abscisic acid (ABA) and causes oxidative damage [6]. During germination, enzyme activity and membrane stability are impacted by both high and low temperatures. This study aims to evaluate the oxidative stress markers in wheat during germination under abiotic stress.

2. Materials and Methods

2.1 Collection of seed

To ensure both experimental validity and seed quality, it is crucial to perform *Triticum* (wheat) seeds for a germination experiment. Healthy and fully maturity seeds of wheat were collected from Krushi Vigyan Kendra, located near to the University. To ensure viable seeds, they are chosen from plants that are free of disease and injury. The collected seeds are washed to get rid of chaff and debris after being collected, and they are then allowed to air dry in a shady location to avoid fungal infestation. Before the germination test, seeds are kept at a low temperature (4°C) in moisture - proof containers after they have dried. Seeds are surface sterilized with 1% sodium hypochlorite before being experiment in order to prevent microbial influence [7]. To ensure the traceability and dependability of the germination test findings, proper documentation and handling were carried out at every stage.

2.2 Method of germination

The purpose of the wheat seed germination experiment was to investigate, under controlled conditions, the germination rate, viability, and early seedling vigour of *Triticum* species. Chosen consistent, fully grown wheat seeds, then use a 1% sodium hypochlorite solution to surface sterilize them for two to three minutes. Seeds were rinsed thoroughly with distilled

water. Damped filter paper to Petri dish. In each plate, equally distribute 20 seeds. Paper was wet but not soggy by moistening it with distilled water. To keep the Petri dish moist, covered them and incubated them at 25°C for ten days. As necessary, added water to keep the area wet. Noted how many seedlings sprouted each day. The radicle must emerge by at least 2 mm for a seed to be deemed germinated. Determined the germination percentage by applying the following formula [8]:

Germination (%) = (Number of seeds germinated / Total seeds) × 100

2.3 Study of salt stress on Triticum aestivum

The test for salt stress tolerance assesses how well *Triticum aestivum* seeds or seedlings can sprout and develop in salty environments. This is essential for finding salt - tolerant cultivars to use in regions that are prone to salinity. Dissolved calculated quantities of NaCl in distilled water to create various concentrations e. g., 0 mM, 50 mM, 100 mM, 150 mM, and 200 mM. After two to three minutes of surface sterilization with 1% sodium hypochlorite, rinsed the seeds with distilled water. Used filter paper to line Petri plates and put 20 seeds on per plate. Added 10 ml of the specified NaCl solution to moisten each dish. As a control, used pure water (0 mM). Petri dishes were covered and incubated in indirect light at 25°C. Added the same salt solution as necessary to maintain moisture levels. Observed for ten days and on the last day, measured the lengths of the roots and shoots and noted how many seeds germinated each day.

Germination % = (Germinated seeds / Total seeds) × 100

2.4 Extraction and evaluation of Malondialdehyde (MDA) in stress

One important marker of lipid peroxidation and oxidative stress in plants is malondialdehyde (MDA). When *Triticum aestivum* (wheat) was subjected to stress (such as salt), measuring its MDA levels offers information on membrane damage and stress tolerance. By measuring malondialdehyde, a breakdown product that arises from the peroxidation of polyunsaturated fatty acids, the levels of lipid peroxidation were evaluated [9]. New leaves from plants were collected from the plants grown at different concentration of salt stress and control. In 5 ml of 0.1% TCA, grinded 0.5 gm of fresh leaf tissue with a cold mortar and pestle. Filled centrifuge tubes with the homogenate and centrifuged at 4°C for 10 minutes at 10, 000 rpm, collected the supernatant for analysis using MDA. Mixed 4 ml of 20% TCA with 0.5% TBA with 1 ml of supernatant. The mixture was heated for 30 minutes at 95°C in a water bath. Measure the clear supernatant's absorbance with a spectrophotometer at 532 and 600 nm.

MDA (nmol/g fw) = $(A_{532}-A_{600})$ ε× volume (ml) ÷s ample fresh weight (g)

3. Results and Discussion

The wheat seed germination experiment showed clear differences in germination rates and seedling vigour across treatments. Under control conditions (distilled water), germination began within 2 days, reaching a final germination percentage of 95% by day 7 (Table 1). Seeds germinated consistently, with good root and shoot growth.

Table 1: Percentage of seed germination of wheat (*Triticum aestivum)* under controlled condition. The values are average of 3 replicates ± SEM

No of days	Percentage of seed germination
0	-
2	10 ± 0.04
4	53 ± 0.07
6	95 ± 0.06
8	95 ± 0.03

However, under salt stress (e. g., 100 mM NaCl), germination was delayed, with only 50% of seeds germinating at the end of the observation period. Higher concentration still reduced the germination and at 200 mM only 10 percent seed are germinated (Table 2). The shorter roots and shoots indicated stunted growth in the seedlings under salt stress. The decreased rate of germination and development of seedlings in saline environments implies that salt stress impairs the absorption of water and the activity of enzymes required for germination. Ion toxicity, especially from Na^+ and Cl^+ ions, and osmotic stress probably played a factor in the afflicted seeds delayed or unsuccessful germination [10]. These results are in line with earlier research that found salt lowers *Triticum aestivum* seed vigour and germination.

Table 2: Effect of different concentration of NaCl on seed germinated of *Triticum aestivum* after 7 days. The values are average of 3 replicates ± SEM

Conc of NaCl	% of germination
0 mM (Control)	95 ± 0.01
50 mM	70 ± 0.04
100 mM	50 ± 0.09
150 mM	25 ± 0.02
200 mM	10 ± 0.05

An important effect of rising salinity on seed germination and seedling growth was found in the salt stress experiment conducted on *Triticum aestivum*. Under control circumstances (0 mM NaCl), wheat seeds exhibited robust root and shoot growth and a high germination rate (about 95%). On the other hand, germination percentage and seedling vigour significantly decreased as salt concentration rose. Germination decreased to around 50% at 100 mM NaCl, and root/shoot lengths were considerably shortened. Germination was less than 20% at 200 mM NaCl, and bleached leaves and stunted development were obvious symptoms of stress. These findings suggest that the physiological mechanisms involved in germination are adversely impacted by salt stress. While ionic toxicity from Na^+ and Cl^+ impaired enzyme function and cell division, the osmotic imbalance brought on by excessive salt concentrations probably limited water absorption. Reduced root length under salt stress is also indicative of early growth suppression and poor nutrient absorption [11]. Overall, the information supports the idea that *Triticum aestivum* is sensitive to salt, particularly in its early phases of growth. For sustainable agriculture in saline - prone regions, this experiment emphasizes the significance of testing wheat cultivars for salt tolerance. The assessment of *Triticum aestivum*'s malondialdehyde (MDA) content under stress circumstances showed a definite rise in MDA levels in

comparison to the control. MDA levels in non - stressed plants were comparatively low, suggesting less lipid peroxidation. But when exposed to salt stress, MDA levels rose dramatically, reaching their maximum at 200 mM NaCl treatment. This increase in MDA level is a result of increased oxidative stress and lipid peroxidation - induced cell membrane damage [12].

Table 3: Effect of different concentration of NaCl on % of activity of malondialdehyde (MDA) in *Triticum aestivum* at 7 days. The values are average of 3 replicates ± SEM

Conc of NaCl	malondialdehyde (MDA) % of activity
0 mM (Control)	50 ± 0.04
50 mM	50 ± 0.02
100 mM	70 ± 0.03
150 mM	80 ± 0.08
200 mM	20 ± 0.02

According to the findings, MDA production is increased up to 150 mM salt stress and then decreases (table 3). When salt stress triggers the development of reactive oxygen species (ROS), which target membrane lipids. Thus, the rise in MDA concentration serves as a trustworthy gauge of the degree of oxidative damage and stress experienced by wheat plants. These results shed light on the physiological reactions of wheat under salt stress and support the use of MDA as a biochemical marker for stress tolerance research.

4. Conclusion

Understanding how oxidative stress impacts wheat germination helps in developing stress - resistant varieties for sustainable agriculture. The rate of germination was significantly lowered under salt stress, and delayed germination and stunted development were caused by greater NaCl concentrations. The elevated MDA levels under stress circumstances suggested that stressed plants had more severe oxidative cell membrane damage, which was indicative of compromised physiological and metabolic functions. Important information about the plant's stress response mechanisms may be gained from the alterations in *Triticum aestivum* L. oxidative state during germination and the early stages of growth under abiotic stress. Malondialdehyde (MDA), a biomarker of lipid peroxidation, was shown to be raised under both salt and drought stress, indicating that both conditions greatly boosted the formation of reactive oxygen species (ROS) and consequent oxidative damage. According to this, oxidative stress, which impairs germination rates, root and shoot development, and overall seedling vigour, is most likely to harm wheat during its early stages of growth. Under drought stress, comparable outcomes were noted, where a lack of water enhanced oxidative damage and ROS generation. Some wheat cultivars, however, demonstrated more resilience to these pressures, growing better in salty or drought - prone environments and accumulating less MDA. This study reveals that salt and drought stresses significantly impair wheat germination and early growth by increasing oxidative stress, as indicated by elevated MDA levels. These findings highlight the importance of enhancing antioxidant defences to improve stress resilience in wheat. Continued research should focus on identifying genetic traits contributing to stress tolerance for sustainable wheat production. To generate stress - resistant cultivars for cultivation in difficult settings, further study is required to pinpoint the precise genes or processes that improve wheat's resistance to oxidative stress.

References

[1] Wu, Y., Liu, J., Zhao, L., Wu, H., Zhu, Y., Ahmad, I., & Zhou, G. (2024). Abiotic stress responses in crop plants: A multi - scale approach. *Journal of Integrative Agriculture*. https: //doi. org/10.1016/j. jia.2024.09.003

[2] Hossain, A., Skalicky, M., Brestic, M., Maitra, S., Alam, M. A., Syed, M. A., Hossain, J., Sarkar, S., Saha, S., Bhadra, P., Shankar, T., Bhatt, R., Chaki, A. K., Sabagh, A. E., & Islam, T. (2021). Consequences and Mitigation Strategies of Abiotic Stresses in Wheat (*Triticum aestivum* L.) under the Changing Climate. Agronomy, 11 (2), 241. https: //doi. org/10.3390/agronomy11020241

[3] Shiade, S. R. G., Rahimi, R., Zand - Silakhoor, A., Fathi, A., Fazeli, A., Radicetti, E., & Mancinelli, R. (2024). Enhancing seed germination under abiotic stress: Exploring the Potential of Nano - Fertilization. Journal of Soil Science and Plant Nutrition, 24 (3), 5319–5341. https: //doi. org/10.1007/s42729 - 024 - 01910 - x

[4] Zia, R., Nawaz, M. S., Siddique, M. J., Hakim, S., & Imran, A. (2020). Plant survival under drought stress: Implications, adaptive responses, and integrated rhizosphere management strategy for stress mitigation. Microbiological Research, 242, 126626. https: //doi. org/10.1016/j. micres.2020.126626

[5] Arif, Y., Singh, P., Siddiqui, H., Bajguz, A., & Hayat, S. (2020). Salinity induced physiological and biochemical changes in plants: An omic approach towards salt stress tolerance. Plant Physiology and Biochemistry, 156, 64–77. https: //doi. org/10.1016/j. plaphy.2020.08.042

[6] Wei, Y., Javed, T., Liu, T., Ali, A., & Gao, S. (2024). Mechanisms of abscisic Acid - mediated Plant Defense Responses: An Updated review. *Plant Stress*, 100724. https: //doi. org/10.1016/j. stress.2024.100724

[7] Varma, A., & Jain, A. (2021). Protocol for seed surface sterilization and in vitro cultivation. In Springer eBooks (pp.265–282). https: //doi. org/10.1007/978 - 981 - 16 - 3832 - 9_13

[8] Zhang, T., Fan, S., Xiang, Y., Zhang, S., Wang, J., & Sun, Q. (2020). Non - destructive analysis of germination percentage, germination energy and simple vigour index on wheat seeds during storage by Vis/NIR and SWIR hyperspectral imaging. Spectrochimica Acta Part a Molecular and Biomolecular Spectroscopy, 239, 118488. https: //doi. org/10.1016/j. saa.2020.118488

[9] Tsikas, D. (2016). Assessment of lipid peroxidation by measuring malondialdehyde (MDA) and relatives in biological samples: Analytical and biological challenges. *Analytical Biochemistry*, *524*, 13–30. https: //doi. org/10.1016/j. ab.2016.10.021

[10] Malakar, P., & Chattopadhyay, D. (2021). Adaptation of plants to salt stress: the role of the ion transporters. Journal of Plant Biochemistry and Biotechnology, 30 (4), 668–683. https: //doi. org/10.1007/s13562 - 021 - 00741 - 6

[11] Zou, Y., Zhang, Y., & Testerink, C. (2021). Root dynamic growth strategies in response to salinity. *Plant*

Cell & Environment, *45* (3), 695–704. https: //doi. org/10.1111/pce.14205

[12] Tsaturyan, V., Poghosyan, A., Toczyłowski, M., & Pepoyan, A. (2022). Evaluation of Malondialdehyde Levels, Oxidative Stress and Host–Bacteria Interactions: Escherichia coli and Salmonella Derby. *Cells*, *11* (19), 2989. https: //doi. org/10.3390/cells11192989