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Integrated Use of Potassium, GA_3, and Cytokinin in Mitigating Salinity-Induced Mineral Imbalance in Wheat Seedlings

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Abstract: ***Salinity is a significant abiotic stress factor in wheat (Triticum aestivum L.). Salinity alters the mineral concentration and its distribution in various plant parts. Due to this ion homeostasis get disrupted and result in poor seedling growth in wheat. Present work is to find the possibility of exogenous application of potassium (K^+), gibberellic acid (GA_3) and cytokinin (CK) treatment alone or in combination to alleviate the salinity induced change in mineral ion concentration at various salinity levels (NaCl salinity) in wheat seedlings. It was observed that there is accumulation of chloride (Cl^-) in radicle and coleoptile, while the level of nitrate (NO_3^-), phosphate (PO_4^{3-}) and sulphate (SO_4^{2-}) reduced dramatically. All the treatments (alone or in combination) given to seeds improved the mineral ion concentration. Treatment $K^+ + GA_3$ + CK was found the best treatment or most successful treatment in restoring mineral level in seedlings of wheat. It indicates that ion uptake, re-establishment of nutrient transport and reduction in chloride toxicity is a complex function. Integrated treatment of nutrients and hormones enhanced wheat's ion homeostasis and salt tolerance for crop productivity in alkaline soils.***

Keywords: Salinity stress, Potassium (K^+), Gibberellic acid (GA_3), Cytokinin (CK), Ion homeostasis, Chloride toxicity

I. INTRODUCTION

Salinity stress is one of the major environmental challenges on global agriculture. According to FAO statistics (2021), 424 million hectares of upper soil layer and 833 million hectares subsoil is affected by salt across 118 countries. The situation is becoming adverse every year and by the end of 2050 this number will increase sharply (Mukhopadhyay *et al*., 2021).

Wheat, a significant staple crop, is moderately sensitive to salinity. Under salt stress, plant faces ionic and osmotic stress, which causes reduced water intake and disturbance in nutritional homeostasis (Nikolic *et al.*, 2023). In saline soils, conc. of sodium (Na^+) and chloride (Cl^-) is high that displace the vital nutrients such as potassium, nitrate, phosphate and sulphate (Lindberg and Premkumar, 2024). This ionic imbalance affects the metabolic activities and overall development of the plants (Irik and Bikmaz, 2024). Exogenous application of potassium stimulates the preference of potassium uptake over Na. Hence, potassium plays a significant role in ion balance in saline conditions (S. Gupta, 2025). Exogenous application of potassium improves salt tolerance in wheat as it helps in maintaining the K/Na ratio, which results in improved anion transport and chlorophyll production (Abbas *et al*., 2025). Phytohormones (Gibberellic acid and Cytokinin) play a significant role in plant physiology under abiotic stress (Attia, 2022; Oyebamiji *et al*., 2024). Cytokinin is responsible for cell division, new chloroplast synthesis, and it delays senescence, while Gibberellins promote cell elongation, seed germination and leaf growth in wheat (Srivastava and Pandey, 2023). Growth regulators can mitigate the salt induced effects in plants as they improve ion transport and enhance antioxidant defense system (Singh *et al*., 2022). Recent research has demonstrated that the combined treatment of minerals and plant growth regulators have better impact on mitigating salinity induced effects. For example, when GA3, CK and K are given together, it increased the early seedling growth, ion balance and membrane stability under saline conditions (Sarwar *et al*., 2023). Seed priming with Gibberellins and cytokinin also enhanced nitrate and phosphate accumulation. Present study was to find how the anion content of wheat seedlings were affected with salinity and to what extent it can be mitigated by seed priming with mineral and growth regulators, either individual or in combination. So, to improve the salt tolerance in plants, our aim is to identify how these treatments are helping in ion balance and recovery of plant physiology.

Seed germination is the first stage in plant life cycle and better germination plays a crucial role in seedling growth and overall plant development. This phase (Seed germination) is very sensitive to changes in environmental conditions. Abiotic stresses severely affect not only seed germination but also the seedling growth and development of the crop. (Sabagh EL.*et al*., 2021., Atta *et al*., 2023).

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The high sensitivity of seed germination to other growth stages is because the seedling completely depends on seed storage for its early growth (Irik and Bikmaz, 2024). To survive under abiotic stress, seedlings show various modifications in their morphological, physiological, biochemical and genetical responses (Wani, 2023, Rehman et al., 2025). Plant hormones significantly help in such adaptive processes.

Plant hormones influence the series of biochemical reactions to control cell functions and help not only in seed germination but also seedling development under both saline and non- saline conditions. Recent studies also clearly show that the seed priming or exogenous application of plant growth regulators enhance plant adaptation to salinity during seed germination and early seedling growth (Sabagh El, 2022, Zahid *et al*., 2023). So, for sustainable agriculture, it is important to understand that plant growth depends upon better seed germination and early seedlings growth and as this stage is vulnerable to abiotic stress, so work should be done to mitigate the salt stress at this stage.

II. MATERIAL AND METHODS

Mineral Estimations: 96h old seedlings were used for mineral analysis. Prior to digestion, the endosperms, coleoptile, and radicle were oven- dried.

Digestion: 70 mg of dried and powdered material was acid digested in 5ml of 9:1 Sulphuric acid and Perchloric acid mixture and was diluted with 25 ml distilled H_2O.

Estimation of Chloride: The acid digest of seedlings was used to determine chloride content. Ion analyser LI-126 (Elico) using standard KCl was used for estimation.

Phosphorus: To estimate P content in seedlings, the method of Jackson (1973) was used.

Nitrogen: To estimate N content in seedlings, the method of Linder (1944) was used with some modifications.

Sulphate estimation: Turbidimetric method Tabatabai and Bremner (1970) was adopted for sulphate estimation

III. RESULTS AND DISCUSSION

A. Nitrogen Content

Figure 1 demonstrates that the N concentration in radicle and coleoptile decreased quite sharply as salinity levels rose, although it was more pronounced in endosperm. The harmful buildup of salt ions may be the cause of the decreased mobilisation of dietary material. In radicle and coleoptile, the N content was likewise marginally increased by the seed soaking priming treatments in comparison to the water-treated controls. On the other hand, endosperm showed a reverse trend in N content, which is a decrease with seed priming treatments. These findings demonstrated unequivocally that nutrients and growth regulators partially eliminated the inhibition of food material mobilization from the endosperm to the embryo axis caused by salt. In this regard, it should be mentioned that K^++GA_3+CK was the most effective treatment. There was no significant interaction.

At the S_{12} level of salinity, the N content of the radicle and coleoptile decreased by 33% and 35%, respectively, in comparison to the control. Nevertheless, endosperm exhibited a rise in N content with salinity, with the depletion of N content decreasing as salinity increased and reached its maximum in control (Fig.1). Since total nitrogen and protein content fell while NO^{3-} and NH_4^+ content increased, salinity also disrupted the accumulation of the N fraction, indicating impairment of the basic synthetic processes. Different seed priming treatments increased the nitrogen content by changing the membrane's selectivity to favour nutritional ions over harmful salt ions.

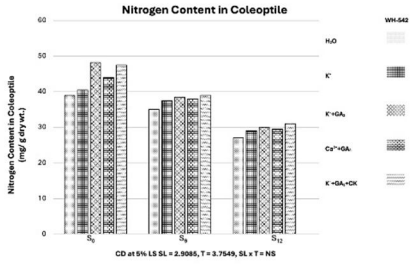

Figure 1a

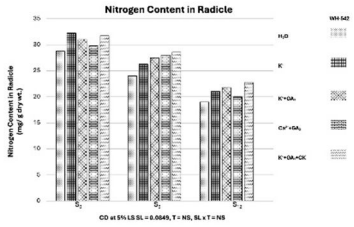

Figure 1b

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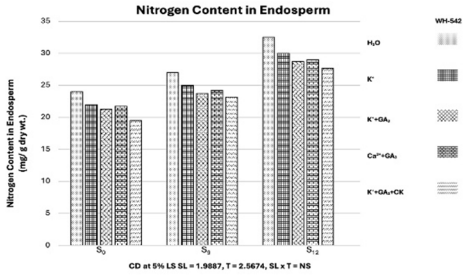


Figure 1c

B. Phosphorus Content

Figure 2 illustrates the synergism between P and Cl uptake by demonstrating how the phosphorus content of radicle, coleoptile, and endosperm increased with increasing salinity. However, the P level in every organ examined for this purpose was reduced by seed priming with nutrients and growth regulators, both separately and in combination. This could indicate that P was used more frequently in other organic or inorganic cell or tissue components, whereas priming therapies resulted in lower P concentration. The relatively high P level in radicle and endosperm compared to coleoptile was another intriguing finding. Nevertheless, it was determined that there was no significant interaction between the salt levels and the seed priming treatments. Salinity negatively impacts phosphorus uptake in seedlings of different crop species, according to recent studies. For instance, Aicha *et al.* (2022) found that in wheat under increasing salinity stress had a much lower phosphorus content, which they attributed to compromised root absorption processes. Similarly, a decrease in phosphate level with salinity was also observed by Dey et al., 2025. This suggests that phosphorus translocation is impeded during early growth stages by salinity-induced ionic imbalances.

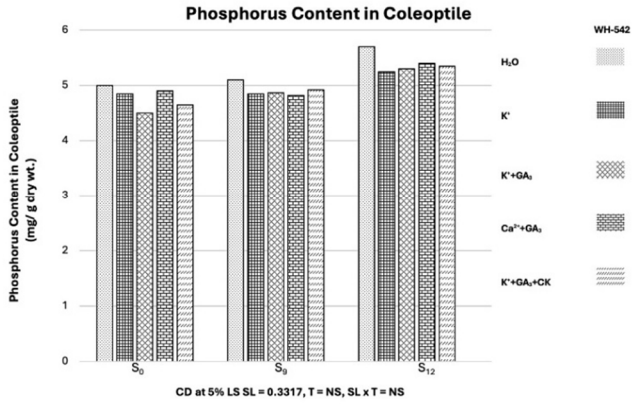
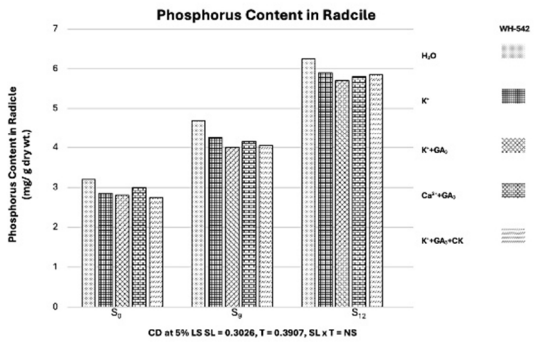


Figure 2a



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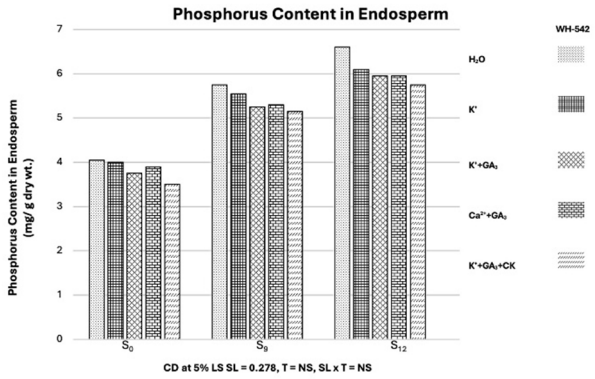
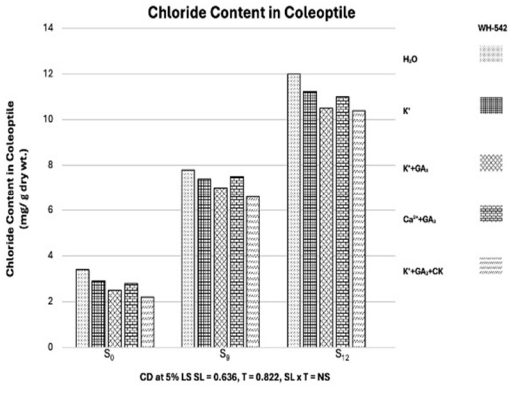


Figure 2c

C. Chloride Content

The chloride content increased three and six times at S_9 and S_{12} salinity levels in the radicle and coleoptile, respectively, according to the diagrammatic representation of the material in Fig. 3. The Cl content of endosperm, on the other hand, increased three and four times at the S_9 and S_{12} levels, respectively. On the other hand, the Cl concentration decreased with seed priming treatments, and the depletion was greater with K^++GA_3+CK and K^++GA_3, while the reduction was very slight. It is important to remember that at the same salinity levels, the radicle and coleoptile had a higher Cl content than the endosperm. Nevertheless, it was determined that there was no significant interaction between the salt levels and the seed priming treatments.

In wheat seedlings, it has been observed that the amount of chloride steadily rises as salinity levels do. This pattern is corroborated by recent research, which shows that excessive chloride buildup in plant tissues is caused by high external salt concentrations. Akhtar et al., (2021) documented an ionic imbalance caused by chloride in Bangladesh wheat cultivars under saline stress and role of Gibberellins and cytokinin in ion accumulation or Ion homeostasis.



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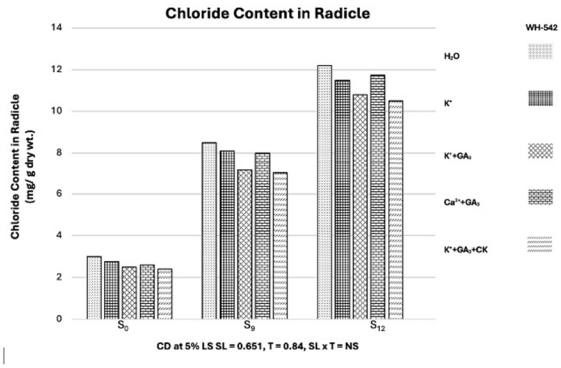

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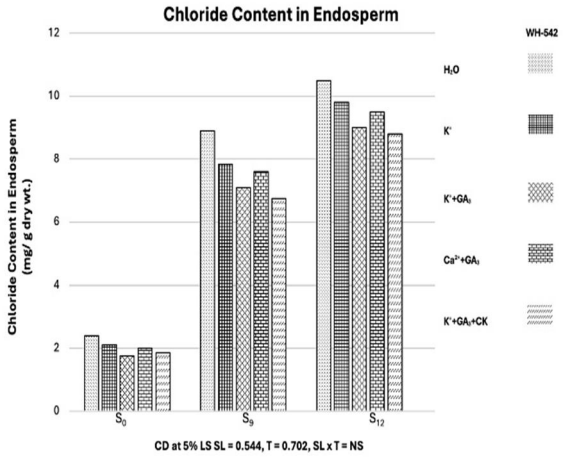

Figure 3c

D. Sulphate Content

The information shown in Fig. 4 shows that the sulphate content increased as salinity levels rose. With the highest salinity, there has been an approximate two-fold increase in comparison to controls. The sulphate concentration decreased with seed priming treatments, but K++GA3+CK showed a notable decrease. Nearly identical patterns have been noted for both endosperm and coleoptile. Nevertheless, it was determined that there was no significant interaction between the salt levels and the seed priming treatments.

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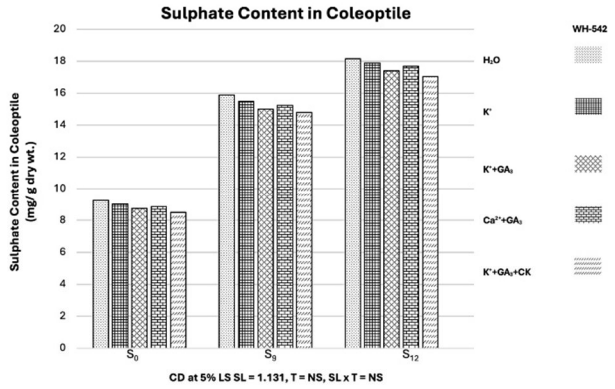

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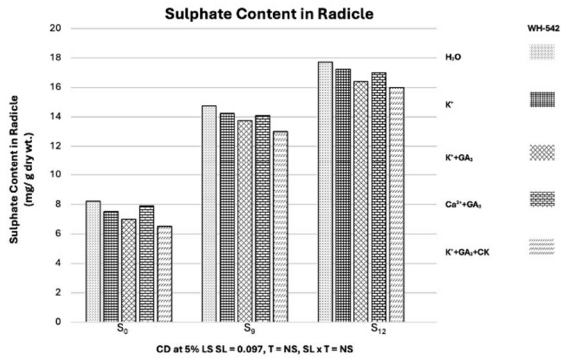

Figure 4b

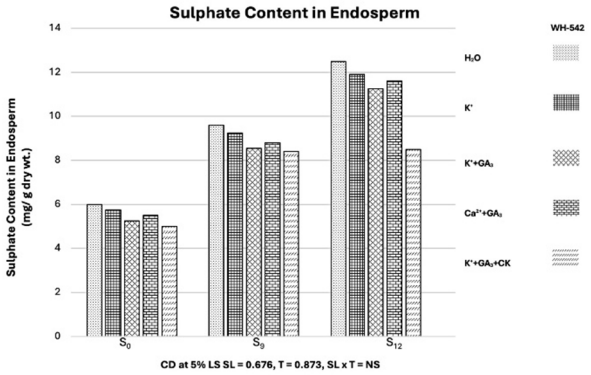

Figure 4c

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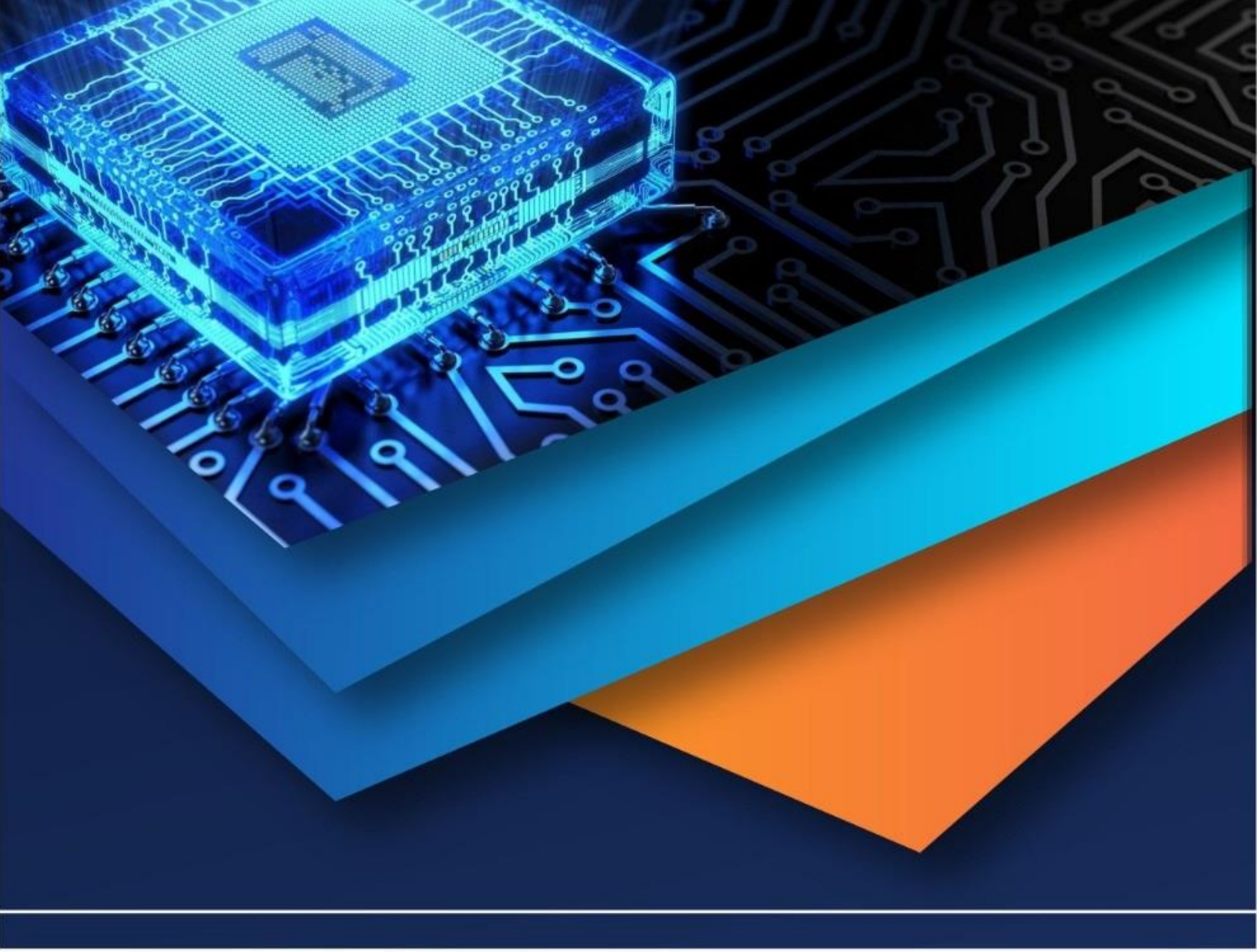

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