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FOLIAR ZINC SUPPLEMENTATION ENHANCES YIELD AND MITIGATES OXIDATIVE DAMAGE IN SALT-STRESSED BINA DHAN-8

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Foliar Zinc Supplementation Enhances Yield and Mitigates Oxidative Damage in Salt-Stressed BINA dhan-8

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ABSTRACT

Soil salinity is one of the fastest-growing constraints on rice (*Oryza sativa* L.) productivity worldwide, currently affecting nearly 1.4 billion hectares of land, with a further one billion hectares at risk. Salinity-induced osmotic and ionic stress triggers excessive reactive oxygen species (ROS) accumulation, disrupting membrane integrity and nutrient homeostasis; zinc (Zn), a cofactor of key antioxidant enzymes, is itself often rendered less available in saline soils, compounding the damage. This study evaluated whether foliar Zn nutrition could mitigate salt-induced oxidative stress and sustain the yield of the salt-tolerant rice variety BINA dhan-8. A pot experiment was conducted in a Completely Randomized Design with two factors: four foliar Zn levels (0, 0.05, 0.10 and 0.15%) and four salinity levels (0, 3, 6 and 9 dS/m) - replicated three times (48 pots) during the 2025 Aman season at EXIM Bank Agricultural University, Bangladesh. Chapainawabganj. Both factors and their interaction significantly ($p < 0.05$) affected all growth and yield-contributing traits. The combination of 0.05% Zn with moderate salinity (6 dS/m) produced the tallest plants (85.30 cm), the highest number of effective tillers (14.33 hill⁻¹), the longest panicles (24.10 cm), the most filled grains (97.33 panicle⁻¹) and the highest grain yield (22.57 g pot⁻¹); 68% above the unsprayed, most severely salinized control (Zn₀S₃, 13.47 g pot⁻¹). Excess Zn (0.15%) did not add further benefit, pointing to an optimum foliar dose near 0.05%. Correlation analysis confirmed grain yield was most strongly associated with panicle length ($r = 0.98$) and negatively associated with unfilled grain number ($r = -0.98$). These results demonstrate that a single, low-cost foliar Zn spray schedule can substantially buffer rice yield losses on moderately saline soils, offering a practical, scalable option for salinity-prone deltas such as coastal Bangladesh.

1.0 INTRODUCTION

Rice (*Oryza sativa* L.) is the staple cereal for more than half of the world's population and underpins food security across South and Southeast Asia, yet its productivity is increasingly constrained by the degradation of arable land through soil salinization. According to the Food and Agriculture Organization's first global assessment of salt-affected soils in fifty years, nearly 1.4 billion hectares of land - over 10% of the world's total land area are already affected by salinity with a further one billion hectares at risk from climate change and human mismanagement (FAO, 2024). Coastal and deltaic regions such as Southern Bangladesh are disproportionately exposed, where tidal intrusion, cyclonic surges and reduced dry-season freshwater flow are progressively raising soil and irrigation-water salinity (Shokri et al., 2024). Salinity impairs rice growth through a well-characterized two-phase mechanism: an initial osmotic phase that restricts water uptake and cell expansion, followed by a slower ionic phase in which excessive Na^+ and Cl^- accumulation disrupts K^+ homeostasis, enzyme function and photosynthetic apparatus (Ahmed et al., 2024). Both phases converge on the overproduction of reactive oxygen species (ROS) superoxide, hydrogen peroxide and hydroxyl radicals - which, once they exceed the plant's antioxidant scavenging capacity, cause lipid peroxidation, protein oxidation and membrane damage, ultimately curtailing tillering, panicle development and grain filling (Fathi et al., 2025). Because zinc (Zn) is an essential structural and catalytic cofactor of superoxide dismutase and other antioxidant enzymes, and stabilizes cell membranes against oxidative injury, adequate Zn nutrition is central to a plant's capacity to withstand salt-induced oxidative stress (Ashraf et al., 2014; Dang et al., 2024). Paradoxically, Zn deficiency and salinity frequently co-occur in the field; the elevated pH, high Na^+ and HCO_3^- activity and reduced root Zn-transporter expression typical of saline and saline-sodic soils all suppress Zn uptake and translocation, even where total soil Zn is not limiting (Dang et al., 2024). Soil-applied Zn fertilizer is therefore often only partially effective under saline conditions, whereas foliar Zn application bypasses root-zone constraints, delivers the nutrient directly to metabolically active tissue, and has repeatedly been shown to alleviate salt stress symptoms and improve yield in rice and other cereals (Ashraf et al., 2014; Moradi and Jahanban, 2018; Ahmed et al., 2023). Recent work extending this approach to zinc oxide nanoparticle (ZnO-NP) foliar sprays has reported improved chlorophyll retention, K^+/Na^+ balance, grain yield and grain quality of rice under salt and heat stress (Guo et al., 2024; Lin et al., 2025), reinforcing Zn's role as a practical, scalable amendment for salinity-prone rice systems. Other exogenous protectants such as glycine betaine and proline have shown comparable yield-protective effects under salinity (Lamlom et al., 2025), underscoring that agronomic, foliar-delivered interventions are an active and promising research frontier. In Bangladesh, breeding programs led by the Bangladesh Rice Research Institute (BRRI) and the Bangladesh Institute of Nuclear Agriculture (BINA) have released several salt-tolerant varieties, including BINA dhan-8, which combines moderate salinity tolerance (up to approximately 6-8 dS/m) with acceptable yield potential and has become a preferred choice for the Rabi and Aman seasons in coastal and southwestern districts (BRJ productivity studies; site-suitability assessments for salt-tolerant rice varieties). However, even tolerant varieties experience measurable yield penalties as salinity intensifies, and the extent to which micronutrient management specifically graded foliar Zn dosing - can further buffer these losses in BINA dhan-8 has not been systematically quantified across a full salinity gradient. Despite the growing body of evidence linking Zn nutrition to salt-stress mitigation, few studies have examined the full factorial interaction between graded foliar Zn levels and a wide range of salinity levels within a single controlled experiment, and fewer still have identified an

optimum Zn dose beyond which additional Zn confers no further, or even a negative benefit. Understanding this dose-response relationship is essential for translating Zn nutrition research into a practical, cost-effective recommendation for farmers in salinity-affected deltas. The present study therefore aimed to: (i) quantify the individual and interactive effects of four foliar Zn levels and four salinity levels on the growth, yield components and grain yield of BINA dhan-8 rice; (ii) identify the Zn $\times$ salinity combination that maximizes yield; and (iii) characterize the inter-relationships among growth and yield-contributing traits through correlation analysis, to clarify the physiological pathway through which Zn nutrition translates into yield protection under salt stress.

2.0 MATERIALS AND METHODS

2.1 Experimental site and duration

A pot experiment was conducted at EXIM Bank Agricultural University Bangladesh, Chapainawabganj Sadar (Agro-Ecological Zone 26), during the 2025 Aman season (July-November) to evaluate the performance of the salt-tolerant rice variety BINA dhan-8 under a factorial combination of foliar zinc and soil salinity treatments.

2.2 Soil and plant material

Soil for the experiment was collected from the fields of Chapainawabganj Sadar Upazila. It was a silty clay loam with moderate organic matter content, brown mottles, and a pH ranging from acidic to slightly alkaline (5.1-7.5) (Mahedi et al., 2025). The collected soil was sun-dried, crushed, pulverized and thoroughly homogenized, with inert material, insect pests and propagules removed before pot filling. Seeds of BINA dhan-8, a salt-tolerant rice variety, were obtained from the Chapainawabganj sub-station of the Bangladesh Institute of Nuclear Agriculture (BINA), surface-sterilized with 1% sodium hypochlorite, and rinsed in autoclaved (121°C, 15 psi, 20 minutes) distilled water prior to germination.

2.3 Experimental design and treatments

The experiment followed a two-factor Completely Randomized Design (CRD) with three replications. Factor 1 comprised four foliar zinc levels: $Zn_0 = 0\%$ (control), $Zn_1 = 0.05\%$, $Zn_2 = 0.10\%$ and $Zn_3 = 0.15\%$ Zn, prepared from a commercial chelated zinc fertilizer (10% Zn) dissolved in water (0, 0.5, 1.0 and 1.5 g chelated Zn per 100 mL, respectively). Factor 2 comprised four salinity levels: $S_0 = 0$ dS/m (control), $S_1 = 3$ dS/m, $S_2 = 6$ dS/m and $S_3 = 9$ dS/m, imposed by dissolving 0, 5.76, 11.52 and 17.28 g NaCl, respectively, in 3 L of water per pot. The resulting sixteen Zn $\times$ salinity treatment combinations were assigned at random to plastic pots (25 cm top diameter, 18 cm bottom diameter, 20 cm depth; 8 kg soil pot⁻¹) across the three replications, giving 48 experimental units in total.

2.4 Crop establishment and treatment application

Pre-soaked, sprouted seeds were sown in a wet seedbed on 15 July 2025, one day after basal fertilizer application. Recommended doses of N, P, K and S were applied, with full doses of TSP, MOP and gypsum plus one-third of the urea incorporated during final pot preparation. Four-week-old seedlings were transplanted into the treatment pots on 12 August 2025. Salinity was imposed from two weeks after transplanting, with the calculated NaCl solution (equivalent to 640 mg NaCl L⁻¹ per 1 dS/m) applied in three equal weekly installments to avoid osmotic shock; electrical conductivity was monitored with a meter and adjusted as required throughout the growing period. The remaining urea was top-dressed in two equal splits at 25 and 50 days after transplanting (DAT).

2.5 Foliar zinc application

Foliar Zn solutions (0, 0.05, 0.10 and 0.15% w/v) were prepared from the chelated Zn fertilizer with 0.05% (v/v) Tween-20 surfactant to improve leaf wetting and absorption, and applied at three vegetative growth stages - 15 DAT (early tillering), 30 DAT (maximum tillering) and 45 DAT (panicle initiation) using a hand-held atomizer sprayer in the late afternoon, until foliage was wetted to the point of run-off. Physical shields were placed between pots to prevent spray drift between treatments. Control (Zn₀) plants received an equivalent volume of distilled water containing 0.05% Tween-20 under identical conditions.

2.6 Intercultural operations and harvesting

Weeds and visible insect pests were removed by hand as required, and soil was periodically loosened to maintain aeration. Water was added to each pot as needed to maintain constant soil moisture and the target salinity level. The crop was harvested at physiological maturity on 15 November 2025, and grain and straw yields were recorded on a per-pot (g/pot) basis.

2.7 Data collection

Data were recorded on plant height (cm) and tiller number per hill at 30, 60 and 90 DAT and at harvest; number of effective and non-effective tillers per hill; panicle length (cm); number of filled and unfilled grains per panicle; 1000-grain weight (g); grain yield (g/pot); and straw yield (g/pot).

2.8 Statistical analysis

Collected data were subjected to two-factor Analysis of Variance (ANOVA) appropriate for a Completely Randomized Design using Statistix 10 software. Treatment means were separated using the Least Significant Difference (LSD) test and Duncan's Multiple Range Test (DMRT) at the 5% level of significance, and regression analysis was carried out where appropriate. Pearson correlation coefficients among all measured growth and yield-contributing traits were computed and visualized as a correlation heatmap; all graphical outputs were generated in R (v4.3) using the ggplot2 and reshape2 packages.

3.0 RESULTS

3.1 Plant height and tiller production

Foliar Zn application, salinity level, and their interaction significantly ($p < 0.05$) influenced plant height and tiller number per hill at all growth stages (Table 1). Among Zn levels, Zn₁ (0.05%) produced the tallest plants at every stage, reaching 78.28 cm at harvest; 16% taller than the untreated Zn₀ control (67.38 cm), followed by Zn₂ (75.73 cm) and Zn₃ (72.15 cm), indicating a clear dose optimum at the lowest Zn rate rather than a linear dose response. A similar pattern held for tiller number, where Zn₁ produced the most tillers per hill (11.08) versus 7.58 in Zn₀. For the salinity main effect, moderate salinity (S₂, 6 dS/m) unexpectedly produced the greatest plant height (74.83 cm at harvest) and tiller number (10.25), slightly exceeding the non-saline control (S₀), while severe salinity (S₃, 9 dS/m) caused the sharpest reduction in both traits (70.75 cm; 7.67 tillers). The Zn $\times$ salinity interaction showed that the single best-performing combination for both plant height and tiller number was Zn₁S₂ (0.05% Zn with 6 dS/m salinity), which produced the tallest plants (85.30 cm at harvest) and the highest tiller number (14.33 hill⁻¹) of all sixteen treatment combinations - even surpassing the fully non-saline, Zn-treated combinations. In contrast, the untreated, most severely salinized

combination Zn₀S₃ recorded the shortest plants (66.47 cm) and fewest tillers (6.00 hill⁻¹). Figure 1 illustrates the growth trajectory of plant height from 30 DAT to harvest across all Zn × salinity combinations, highlighting how the yield-protective effect of Zn widens with crop age and becomes most pronounced under moderate salinity.

Table 1. Effect of foliar zinc and salinity levels on plant height and tiller number per hill of BINA dhan-8 rice at different days after transplanting (DAT)

Zinc level	Plant height (cm)				Tiller number per hill			
	30 DAT	60 DAT	90 DAT	Harvest	30 DAT	60 DAT	90 DAT	Harvest
Zn ₀ (0%)	28.43 d	45.61 d	64.12 d	67.38 d	7.58 d	7.58 d	7.58 d	7.58 d
Zn ₁ (0.05%)	33.10 a	53.20 a	74.43 a	78.28 a	11.08 a	11.08 a	11.08 a	11.08 a
Zn ₂ (0.10%)	31.76 b	51.43 b	72.19 b	75.73 b	10.00 b	10.00 b	10.00 b	10.00 b
Zn ₃ (0.15%)	30.50 c	48.91 c	68.41 c	72.15 c	8.92 c	8.92 c	8.92 c	8.92 c
LS	*	*	*	*	*	*	*	*
Salinity level								
S ₀ (0 dS/m)	30.28 c	49.54 c	70.08 c	73.76 c	10.08 a	10.08 a	10.08 a	10.08 a
S ₁ (3 dS/m)	31.58 b	50.41 b	70.69 b	74.19 b	9.58 b	9.58 b	9.58 b	9.58 b
S ₂ (6 dS/m)	32.00 a	51.03 a	71.19 a	74.83 a	10.25 a	10.25 a	10.25 a	10.25 a
S ₃ (9 dS/m)	29.93 d	48.17 d	67.19 d	70.75 d	7.67 c	7.67 c	7.67 c	7.67 c
LS	*	*	*	*	*	*	*	*
Zn × Salinity interaction								
Zn ₀ S ₀	27.30 i	44.87 k	64.23 k	67.37 k	8.33 ef	8.33 ef	8.33 ef	8.33 ef
Zn ₀ S ₁	29.37 g	46.53 i	65.43 j	68.43 j	8.33 ef	8.33 ef	8.33 ef	8.33 ef
Zn ₀ S ₂	28.57 h	45.60 j	63.60 l	67.23 k	7.67 f	7.67 f	7.67 f	7.67 f
Zn ₀ S ₃	28.47 h	45.43 j	63.20 l	66.47 l	6.00 g	6.00 g	6.00 g	6.00 g
Zn ₁ S ₀	31.47 d	52.70 c	73.40 d	77.43 d	11.33 b	11.33 b	11.33 b	11.33 b
Zn ₁ S ₁	34.13 b	54.27 b	76.30 b	80.40 b	10.33 bc	10.33 bc	10.33 bc	10.33 bc
Zn ₁ S ₂	37.30 a	58.43 a	81.47 a	85.30 a	14.33 a	14.33 a	14.33 a	14.33 a
Zn ₁ S ₃	29.50 g	51.40 d	65.57 i	75.23 e	8.33 ef	8.33 ef	8.33 ef	8.33 ef
Zn ₂ S ₀	31.83 cd	52.20 c	74.27 c	77.90 c	11.33 b	11.33 b	11.33 b	11.33 b
Zn ₂ S ₁	32.13 c	51.47 d	71.67 e	75.33 e	10.33 bc	10.33 bc	10.33 bc	10.33 bc
Zn ₂ S ₂	31.50 d	50.67 e	71.27 e	74.47 f	9.67 cd	9.67 cd	9.67 cd	9.67 cd
Zn ₂ S ₃	31.63 d	51.40 d	71.57 e	75.23 e	8.67 def	8.67 def	8.67 def	8.67 def
Zn ₃ S ₀	30.53 ef	48.40 g	68.40 g	72.33 g	9.33 cde	9.33 cde	9.33 cde	9.33 cde

Zn ₃ S ₁	30.70 e	49.37 f	69.37 f	72.60 g	9.33 cde	9.33 cde	9.33 cde	9.33 cde
Zn ₃ S ₂	30.63 e	49.43 f	68.43 g	72.33 g	9.33 cde	9.33 cde	9.33 cde	9.33 cde
Zn ₃ S ₃	30.13 f	48.43 g	67.43 h	71.33 h	7.67 f	7.67 f	7.67 f	7.67 f
LS	*	*	*	*	*	*	*	*
CV (%)	0.88	0.65	0.35	0.28	6.52	6.52	6.52	6.52

Means followed by the same letter within a column are not significantly different by DMRT ($p = 0.05$). * = significant at 5% level; CV = coefficient of variation. Zn₀-Zn₃ = 0, 0.05, 0.10, 0.15% Zn; S₀-S₃ = 0, 3, 6, 9 dS/m, LS=Level of Significance

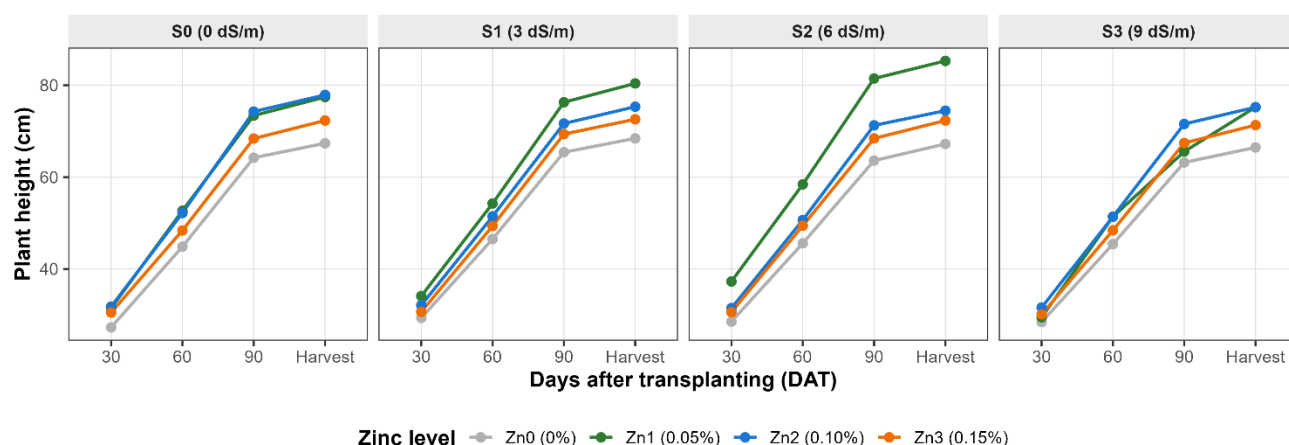


Figure 1. Plant height growth trajectory of BINA dhan-8 rice from 30 DAT to harvest under four foliar zinc levels, faceted by salinity levels

3.2 Effective tillers, non-effective tillers and panicle length

The number of effective (grain-bearing) tillers per hill, non-effective tillers, and panicle length were all significantly affected by Zn level, salinity level and their interaction (Table 2). Zn₁ (0.05%) produced the most effective tillers (9.50 hill⁻¹), the fewest non-effective tillers (2.33 hill⁻¹) and the longest panicles (21.39 cm), reflecting more efficient conversion of vegetative tillers into productive, grain-bearing tillers under adequate Zn nutrition. Salinity alone reduced effective tiller number and panicle length only at the highest level (S₃), while low-to-moderate salinity (S₁-S₂) did not significantly depress these traits relative to the non-saline control, and S₂ in fact produced the highest number of effective tillers among salinity main effects (8.92). The Zn × salinity interaction again identified Zn₁S₂ as the outstanding combination, producing 14.33 effective tillers per hill, only 1.00 non-effective tiller, and the longest panicle recorded in the trial (24.10 cm); 81% longer than the shortest panicle recorded under Zn₀S₃ (13.33 cm). This pattern indicates that moderate salinity, when paired with adequate foliar Zn, does not compromise and may even enhance reproductive tiller development, whereas the same salinity level under Zn-deficient conditions sharply increases the proportion of non-productive tillers.

Table 2. Effect of foliar zinc and salinity levels on effective tillers, non-effective tillers and panicle length of BINA dhan-8 rice

Zinc level	Effective tillers hill ⁻¹	Non-effective tillers hill ⁻¹	Panicle length (cm)
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Zn ₀ (0%)	5.83 d	4.08 a	15.13 d
Zn ₁ (0.05%)	9.50 a	2.33 c	21.39 a
Zn ₂ (0.10%)	8.42 b	3.08 b	20.33 b
Zn ₃ (0.15%)	7.00 c	3.33 b	18.97 c
LS	*	*	*
Salinity level			
S ₀ (0 dS/m)	8.33 ab	3.33 ab	19.71 a
S ₁ (3 dS/m)	7.75 b	2.67 c	19.36 b
S ₂ (6 dS/m)	8.92 a	2.92 bc	19.40 b
S ₃ (9 dS/m)	5.75 c	3.92 a	17.36 c
LS	*	*	*
Zn × Salinity interaction			
Zn ₀ S ₀	6.33 ef	4.00 bc	16.60 i
Zn ₀ S ₁	6.67 ef	2.67 de	15.83 j
Zn ₀ S ₂	6.00 fg	4.33 ab	14.77 k
Zn ₀ S ₃	4.33 g	5.33 a	13.33 l
Zn ₁ S ₀	10.00 b	2.33 e	21.53 bc
Zn ₁ S ₁	8.00 cde	2.33 e	21.83 b
Zn ₁ S ₂	14.33 a	1.00 f	24.10 a
Zn ₁ S ₃	5.67 fg	3.67 bcd	18.10 h
Zn ₂ S ₀	9.67 bc	4.00 bc	21.23 c
Zn ₂ S ₁	9.00 bcd	2.67 de	20.40 d
Zn ₂ S ₂	8.00 cde	2.67 de	20.03 de
Zn ₂ S ₃	7.00 ef	3.00 cde	19.67 ef
Zn ₃ S ₀	7.33 def	3.00 cde	19.47 f
Zn ₃ S ₁	7.33 def	3.00 cde	19.37 f
Zn ₃ S ₂	7.33 def	3.67 bcd	18.70 g
Zn ₃ S ₃	6.00 fg	3.67 bcd	18.33 gh
LS	*	*	*
CV (%)	13.41	22.49	1.18

Means followed by the same letter within a column are not significantly different by DMRT ($p = 0.05$). * = significant at 5% level; CV = coefficient of variation. Zn₀-Zn₃ = 0, 0.05, 0.10, 0.15% Zn; S₀-S₃ = 0, 3, 6, 9 dS/m, LS=Level of Significance

3.3 Yield components and grain/straw yield

Grain filling and final yield traits followed the same overall pattern (Table 3). Zn₁ (0.05%) produced the most filled grains per panicle (87.00), fewest unfilled grains (10.75), heaviest 1000-grain weight (18.61 g), and the highest grain (20.13 g/pot) and straw (31.92 g/pot) yields among Zn main effects 40% and 40% higher than the Zn₀ control for grain and straw yield, respectively. Salinity depressed yield significantly only at the severe level (S₃), while

S₀-S₂ produced statistically similar or even marginally higher grain yields, again pointing to a stimulatory or at least non-injurious effect of low-to-moderate salinity when Zn nutrition is adequate. The Zn × salinity interaction (Table 3, Figures 2-4) shows that Zn₁S₂ produced the highest grain yield of the entire trial (22.57 g/pot) and the highest straw yield (35.70 g/pot), corresponding to a 68% and 66% increase, respectively, over the poorest-performing combination, Zn₀S₃ (grain yield 13.47 g/pot; straw yield 21.53 g/pot). Notably, even under the most severe salinity (S₃), Zn₁ application (Zn₁S₃: 16.37 g/pot) maintained substantially higher grain yield than the Zn-untreated, non-saline control would suggest is possible under stress (Zn₀S₀: 15.13 g/pot), demonstrating that adequate foliar Zn buffers a large share of the yield penalty otherwise imposed by severe salinity. Excess Zn (0.15%, Zn₃) did not match the performance of Zn₁ at any salinity level, indicating that Zn benefits plateau, and can even reverse, beyond an optimal foliar concentration of approximately 0.05%.

Table 3. Effect of foliar zinc and salinity levels on yield components, grain yield and straw yield of BINA *dhan-8* rice

Zinc level	Filled grains panicle ⁻¹	Unfilled grains panicle ⁻¹	1000-grain wt. (g)	Grain yield (g/pot)	Straw yield (g/pot)
Zn ₀ (0%)	65.92 d	29.08 a	14.96 d	14.35 d	22.78 d
Zn ₁ (0.05%)	87.00 a	10.75 c	18.61 a	20.13 a	31.92 a
Zn ₂ (0.10%)	80.08 b	10.67 c	17.53 b	19.55 b	31.45 b
Zn ₃ (0.15%)	76.58 c	17.17 b	16.25 c	17.95 c	28.70 c
LS	*	*	*	*	*
Salinity level					
S ₀ (0 dS/m)	74.58 c	15.17 b	16.92 c	18.49 a	29.43 a
S ₁ (3 dS/m)	77.83 b	14.92 b	17.24 b	18.36 a	29.34 a
S ₂ (6 dS/m)	83.42 a	15.42 b	17.56 a	18.52 a	29.51 a
S ₃ (9 dS/m)	73.75 c	22.17 a	15.63 d	16.61 b	26.57 b
LS	*	*	*	*	*
Zn × Salinity interaction					
Zn ₀ S ₀	65.33 h	28.67 b	14.93 h	15.13 i	23.60 h
Zn ₀ S ₁	64.33 hi	26.00 bc	15.37 g	14.30 j	22.83 i
Zn ₀ S ₂	72.67 g	27.33 b	15.27 gh	14.50 j	23.17 hi
Zn ₀ S ₃	61.33 i	34.33 a	14.27 i	13.47 k	21.53 j
Zn ₁ S ₀	80.67 cd	8.33 g	18.23 c	20.43 c	32.03 c
Zn ₁ S ₁	87.33 b	8.33 g	19.37 b	21.13 b	33.77 b
Zn ₁ S ₂	97.33 a	3.33 h	21.30 a	22.57 a	35.70 a
Zn ₁ S ₃	82.67 c	23.00 c	15.53 g	16.37 h	26.17 g
Zn ₂ S ₀	77.33 def	8.67 g	17.83 d	19.87 d	32.47 c
Zn ₂ S ₁	82.00 c	8.67 g	17.70 d	19.47 de	31.13 d
Zn ₂ S ₂	83.67 bc	12.33 fg	17.33 e	19.53 de	31.27 d
Zn ₂ S ₃	77.33 def	13.00 ef	17.23 e	19.33 e	30.93 d
Zn ₃ S ₀	75.00 efg	15.00 def	16.67 f	18.53 f	29.63 e

Zn ₃ S ₁	77.67 de	16.67 de	16.53 f	18.53 f	29.63 e
Zn ₃ S ₂	80.00 cd	18.67 d	16.33 f	17.47 g	27.90 f
Zn ₃ S ₃	73.67 fg	18.33 d	15.47 g	17.27 g	27.63 f
LS	*	*	*	*	*
CV (%)	2.95	14.25	1.27	1.44	1.45

Means followed by the same letter within a column are not significantly different by DMRT ($p = 0.05$). * = significant at 5% level; CV = coefficient of variation. Zn₀-Zn₃ = 0, 0.05, 0.10, 0.15% Zn; S₀-S₃ = 0, 3, 6, 9 dS/m, LS=Level of Significance

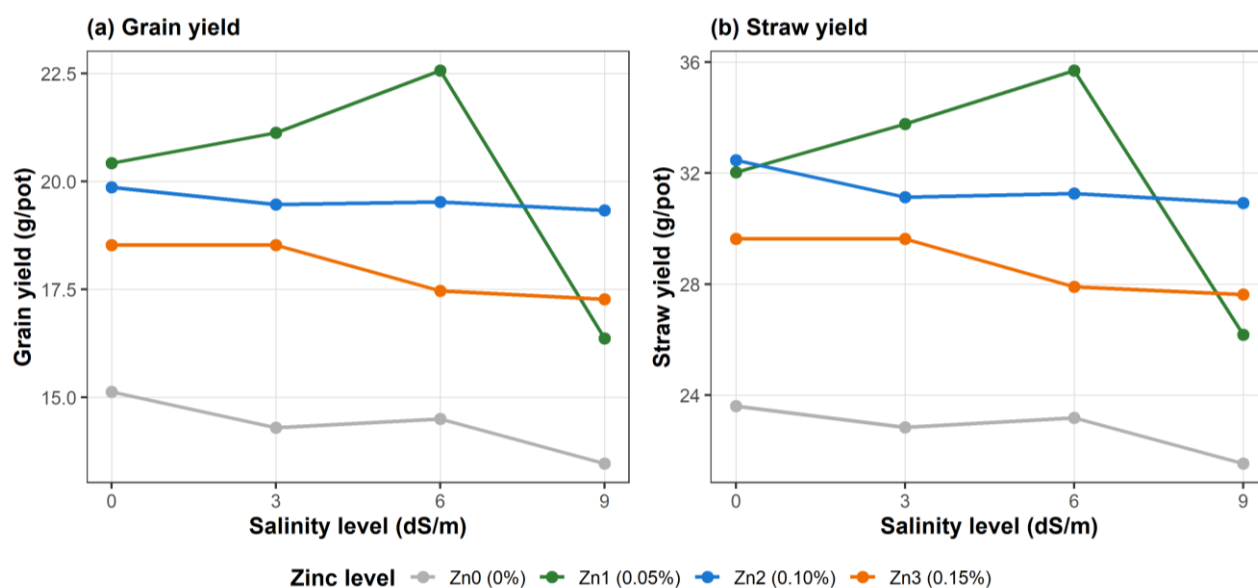


Figure 2. Grain yield (a) and straw yield (b) response of BINA dhan-8 rice across the salinity gradient at four foliar zinc levels

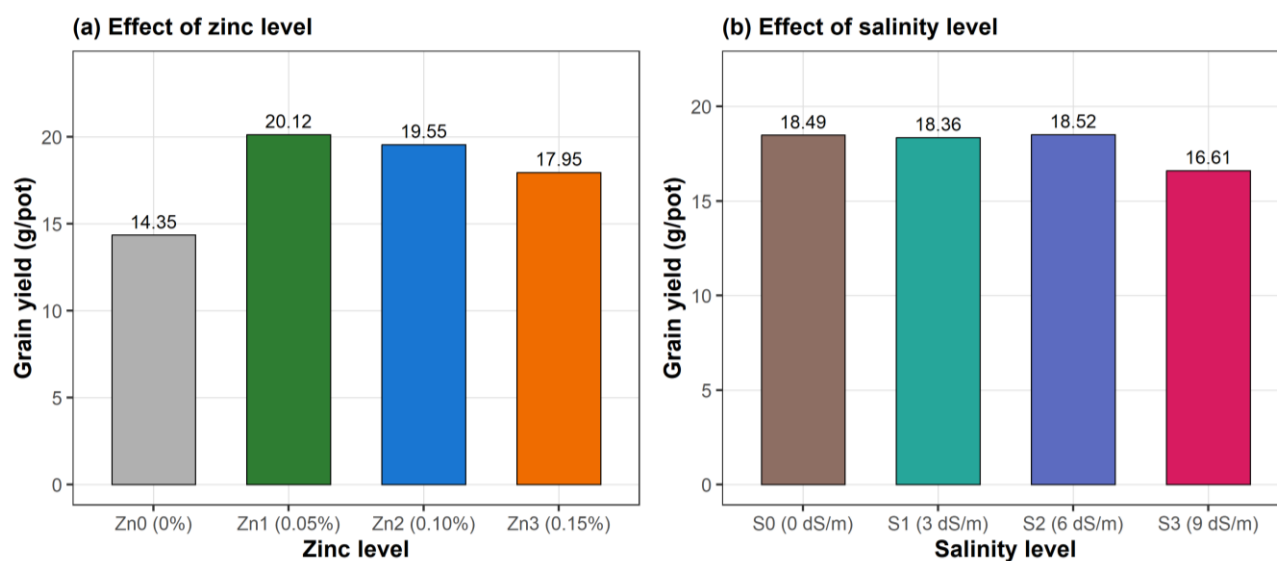


Figure 3. Main effects of (a) zinc level and (b) salinity level on grain yield of BINA dhan-8

rice, averaged across the other factor

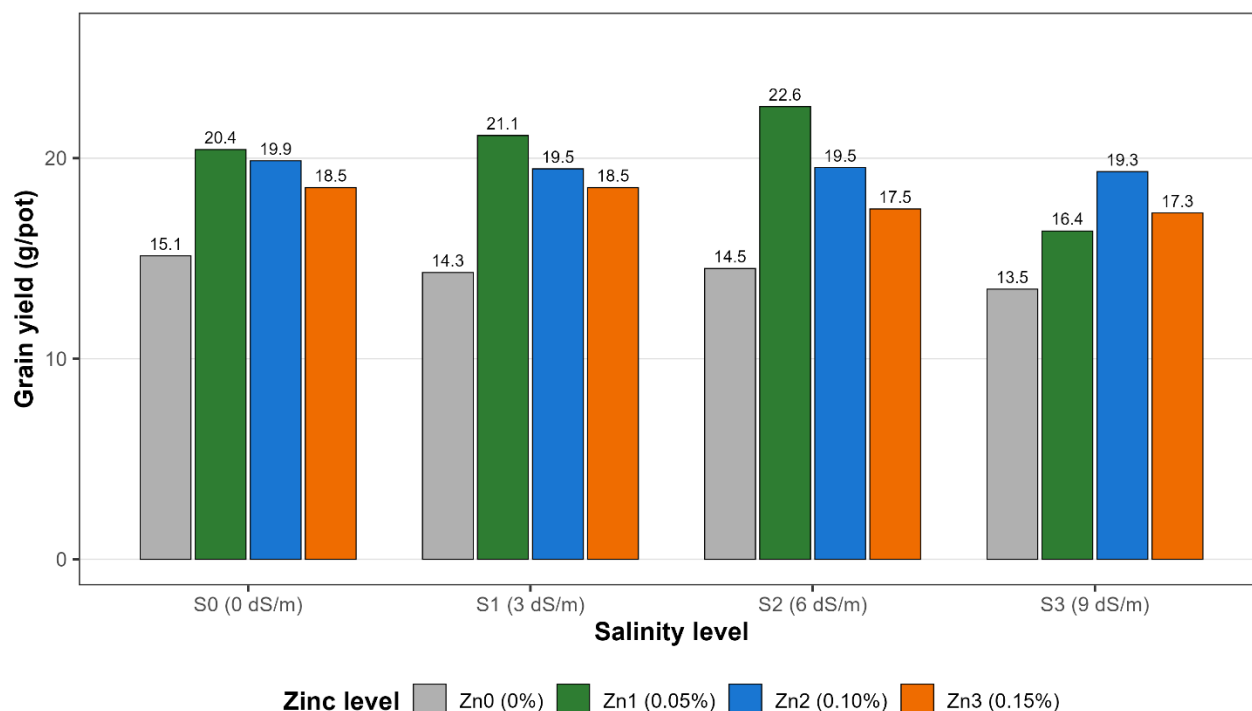


Figure 4. Zinc $\times$ salinity interaction heat map for grain yield (g/pot) of BINA dhan-8 rice, showing the yield optimum at 0.05% Zn combined with 6 dS/m salinity

3.4 Correlation among agronomic and yield-contributing traits

Pearson correlation analysis across the sixteen Zn $\times$ salinity treatment means (Figure 5) revealed strong positive associations among vegetative and reproductive growth traits: plant height was strongly correlated with tiller number ($r = 0.89$), 1000-grain weight ($r = 0.95$), and grain yield ($r = 0.93$). Grain yield showed its strongest positive association with panicle length ($r = 0.98$) and straw yield ($r > 0.99$), and its strongest negative association with the number of unfilled grains per panicle ($r = -0.98$), confirming that spikelet fertility - rather than tiller number or plant height alone is the principal determinant of final yield in this system. Foliar zinc concentration was positively but moderately correlated with grain yield ($r = 0.45$) and straw yield ($r = 0.47$), and negatively correlated with unfilled grain number ($r = -0.47$), consistent with Zn's role in reducing spikelet sterility under stress. Salinity level showed comparatively weak direct correlations with final yield ($r = -0.24$), reflecting the non-monotonic, dose-dependent response observed in Tables 1-3, where moderate salinity did not depress yield when adequate Zn was supplied.

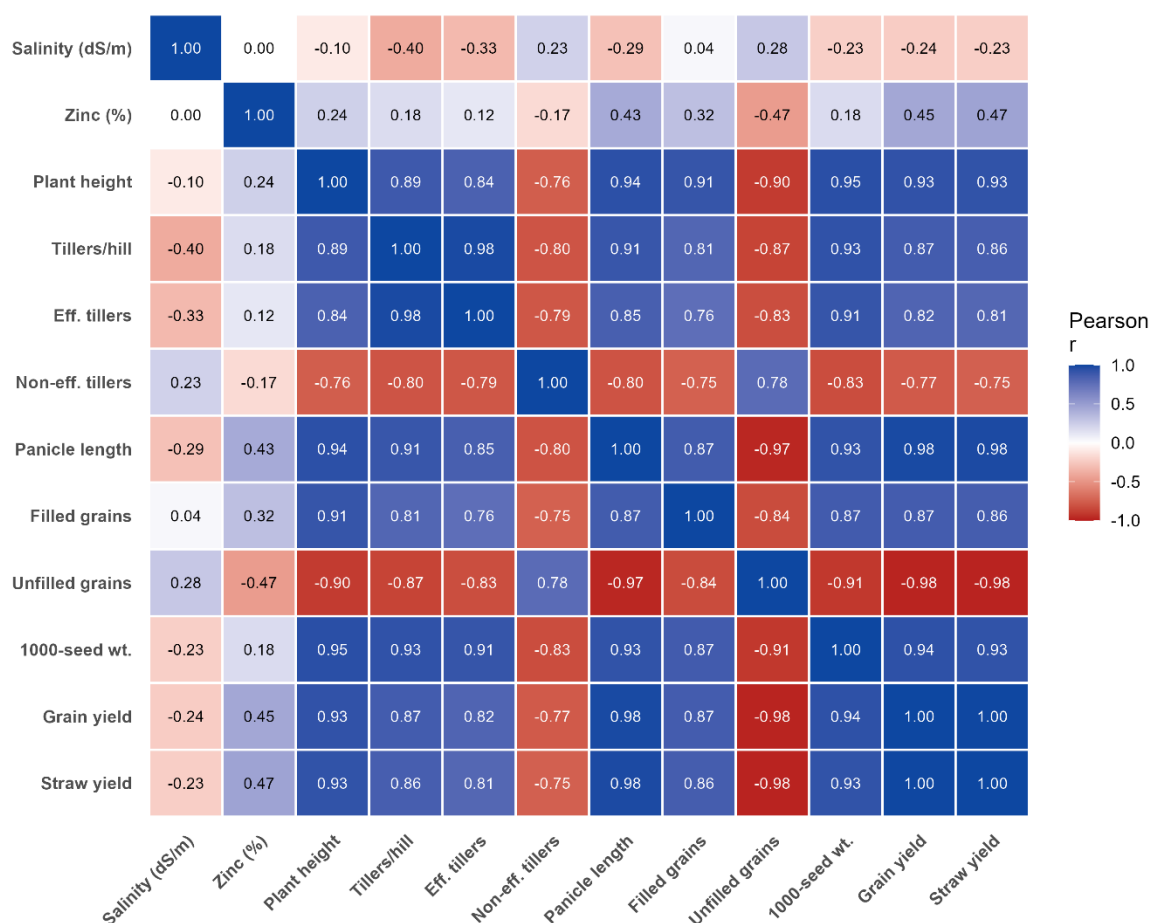


Figure 5. Pearson correlation heatmap among salinity level, zinc level and agronomic/yield-contributing traits of BINA dhan-8 rice ($n = 16$ treatment means). Blue shades indicate positive correlation, red shades indicate negative correlation.

4.0 DISCUSSION

The results demonstrate that foliar Zn nutrition markedly modifies the response of BINA dhan-8 rice to salinity, and that this response is dose-dependent and non-linear with respect to both Zn concentration and salinity level. The clearest finding is that a moderate foliar Zn dose (0.05%) combined with moderate salinity (6 dS/m) produced the best overall performance across virtually every growth and yield trait measured, exceeding even the non-saline, Zn-treated control. This pattern is consistent with a growing body of evidence that Zn strengthens the antioxidant defense system under salt stress: as a structural cofactor of Cu/Zn-superoxide dismutase and related enzymes, Zn helps convert superoxide radicals generated by osmotic and ionic stress into less damaging species, limiting lipid peroxidation and preserving membrane integrity in leaf and root tissue (Dang et al., 2024; Ashraf et al., 2014). The observation that Zn-supplied plants tolerated, and even benefited from, moderate salinity while unsupplied plants deteriorated progressively with increasing salinity mirrors findings from rice cultivar screening trials in which foliar Zn ameliorated declines in photosynthetic rate, stomatal conductance and water relations under salt stress (Ashraf et al.,

2014), and from nano-Zn studies in which Zn application on saline-sodic soils improved nutrient uptake and restored a favorable cationic balance in rice (Ahmed et al., 2023).

The apparent stimulation of growth and yield at moderate salinity when Zn was adequate (S_2 outperforming S_0 in several traits) is consistent with the concept of stress hormesis, whereby a low-to-moderate stress dose can trigger an adaptive up-regulation of antioxidant and osmotic-adjustment machinery that leaves the plant better buffered against subsequent, cumulative stress than an entirely unstressed plant with no such priming (Fathi et al., 2025). Because ROS act as both damaging agents and signaling molecules depending on their concentration and duration (Fathi et al., 2025), a controlled, moderate increase in ROS under 6 dS/m salinity may have triggered protective transcriptional and enzymatic responses that, combined with the antioxidant cofactor supplied by Zn, ultimately enhanced sink capacity as reflected in the exceptionally low unfilled-grain count and long panicles recorded for Zn_1S_2 . This interpretation is consistent with reports that Zn oxide nanoparticles interact with stress-responsive transcription factor families (AP2/EREBP, WRKY, NAC, bZIP) to coordinate antioxidant and osmotic-adjustment gene expression under salinity (Ahmed et al., 2024).

At the highest salinity level (9 dS/m), however, this buffering capacity was insufficient to fully offset stress, and yield declined substantially even in Zn-treated pots, though the decline was consistently smaller than in the Zn-untreated equivalent. This is in line with dose-dependent Zn-salinity interaction studies in rice and maize, where Zn's protective effect narrows as salinity intensity approaches or exceeds the tolerance threshold of the genotype (Moradi and Jahanban, 2018), and reflects the two-phase osmotic-ionic injury model of salinity stress, in which severe, sustained ionic toxicity eventually overwhelms even a well-supported antioxidant system (Ahmed et al., 2024).

A second key finding is the clear plateau, and partial reversal, of Zn benefit at the highest tested concentration (0.15%). Zn_3 consistently underperformed Zn_1 and, in several traits, Zn_2 as well. This non-monotonic dose response is a common feature of micronutrient physiology: while Zn is essential in trace amounts for antioxidant enzyme function and hormone (auxin) synthesis, excess foliar Zn can itself generate localized oxidative stress at the leaf surface, interfere with the uptake or translocation of other divalent cations (e.g., Fe, Mn, Cu) through shared transporter systems, or induce phytotoxic symptoms that offset its protective benefits (Ashraf et al., 2014). The present results therefore identify a practical optimum foliar Zn concentration close to 0.05% for BINA dhan-8 under the tested conditions, above which further Zn investment yields diminishing or even negative agronomic returns.

The correlation analysis reinforces a coherent physiological narrative; grain yield was overwhelmingly explained by panicle length and (negatively) by unfilled-grain number, rather than by plant height or tiller number in isolation. This is consistent with the well-established yield physiology of rice, in which final yield is a function of sink size (spikelet number, largely set by panicle length and effective tiller number) and sink-filling efficiency (spikelet fertility), both of which are highly sensitive to the ionic and oxidative status of the plant during the reproductive and grain-filling phases (Nguyen et al., 2023; Wu et al., 2025). The moderate but consistent positive correlation between Zn concentration and both grain yield and spikelet fertility, together with its negative correlation with unfilled grains, supports the interpretation that Zn's principal agronomic benefit under salinity in this

system operates through protection of reproductive-stage fertility rather than through vegetative growth promotion alone.

From a practical standpoint, these findings support foliar Zn application - a low-cost, easily adopted, three-spray schedule timed to tillering and panicle initiation as a viable agronomic strategy to sustain BINA dhan-8 yield on moderately saline soils (up to approximately 6 dS/m) typical of much of coastal and southwestern Bangladesh, complementing ongoing varietal breeding efforts for salt tolerance (progress reports on BRRI/BINA salt-tolerant rice development). Because the experiment was conducted in pots under controlled salinity dosing over a single season, field validation across multiple seasons, soil types and irrigation-water salinity regimes - together with biochemical confirmation of antioxidant enzyme activity (SOD, catalase, ascorbate peroxidase) and Na^+/K^+ ratios - is recommended before the 0.05% Zn recommendation is generalized to farmer fields. Comparative trials against nano-formulated Zn sources (Ahmed et al., 2023; Lin et al., 2025) and against complementary osmoprotectant sprays such as glycine betaine or proline (Lamlom et al., 2025) would further clarify whether combined interventions can extend the demonstrated benefit to the most severely salinized (≥ 9 dS/m) soils, where foliar Zn alone was insufficient to prevent substantial yield loss.

5.0 CONCLUSION

Foliar zinc nutrition significantly and interactively modified the response of BINA dhan-8 rice to a graded salinity gradient. A foliar Zn concentration of 0.05%, applied at early tillering, maximum tillering and panicle initiation, consistently produced the best growth and yield outcomes, and combined with moderate salinity (6 dS/m) produced the highest grain yield recorded in the trial (22.57 g/pot), 68% above the untreated, most severely salinized control. Zinc's benefit operated principally through protection of reproductive-stage fertility (panicle length and filled-grain number) rather than through vegetative growth alone, consistent with a role in mitigating salt-induced oxidative stress at the grain-filling stage. Excess Zn (0.15%) conferred no additional benefit, indicating an optimal foliar dose near 0.05% under the tested conditions. These findings support low-cost foliar Zn application as a practical, readily adoptable agronomic tool to buffer yield losses in salt-tolerant rice varieties grown on moderately saline soils, while highlighting the need for multi-season field validation and complementary biochemical studies to fully establish the underlying antioxidant mechanism and to extend protection to more severely salinized soils.

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Author Contributions

M.A.E.S. conceived and supervised the study, and drafted and revised the manuscript; M.A.R., M.E.H.R., M.M.H., M.A.G.M., M.A.M., M.E.H., M.S.A.M. and M.F.A. conducted the pot experiment,

collected data, and contributed to data curation and statistical analysis. All authors read and approved the final manuscript.

Conflict of Interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

Data Availability Statement

The datasets generated and analyzed during the current study, including the treatment-mean dataset used for correlation analysis, are available from the corresponding author upon reasonable request.

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