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ENHANCING SEED GERMINATION AND SALT STRESS TOLERANCE IN MUNG BEAN THROUGH SPIRULINA EXTRACT PRIMING

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Abstract

Salinity stress is a major abiotic constraint affecting crop establishment in coastal agricultural regions of Bangladesh. Seed priming using natural biostimulants offers a simple and environmentally friendly approach to improve germination under saline conditions. This study evaluated the potential of Spirulina (*Arthrospira platensis*) extract as a seed priming agent for enhancing germination and early seedling growth of mung bean (*Vigna radiata*) under salt stress. Seeds were primed with aqueous Spirulina extract at concentrations of 1–

5% (w/v) and germinated under NaCl-induced salinity levels of 0, 25, 50, 100, and 150 mM. Germination percentage, mean germination time, shoot length, root length, and seedling vigor index were evaluated. Salinity stress progressively reduced germination and seedling growth in untreated seeds. Spirulina priming improved germination performance and seedling development across salinity treatments, with concentrations of 2–3% generally showing the most consistent improvement. At 100 mM NaCl, Spirulina-primed seeds showed higher germination percentage, faster germination, and improved seedling growth compared with the untreated control. The results indicate that Spirulina extract may support early-stage seedling establishment under saline conditions. This study suggests that microalgal-based seed priming has potential as a sustainable and low-cost strategy for improving crop performance in salt-affected environments. Further research under soil and field conditions is required to confirm practical applicability.

Keywords:

Spirulina, Biopriming, Salinity Stress, Mung Bean, Bangladesh

1. Introduction

Salinity stress stands as a critical abiotic challenge crippling coastal agriculture across Bangladesh, where climate-induced sea-level rise, frequent cyclones, tidal surges, and inadequate drainage have salinized over 1.02 million hectares of prime arable land—approximately 13-20% of total cultivable area. This environmental crisis disproportionately impacts short-cycle legumes like mung bean (*Vigna radiata* L.), a nutritional powerhouse delivering 20-24% protein, nitrogen-fixing capabilities, and suitability for rice-fallow rotations, yet suffering 50-80% germination failure and yield reductions beyond 50 mM NaCl due to disrupted water relations and ionic disequilibrium.

Bangladesh ranks among the global hotspots for soil salinization, with coastal districts like Satkhira, Bagerhat, Khulna, and Barisal recording electrical conductivity (EC) values exceeding 4-8 dS/m during dry seasons. Mung bean seedlings face compounded threats: osmotic inhibition limits imbibition, Na^+/Cl^- toxicity disrupts $\text{K}^+/\text{Ca}^{2+}$ homeostasis, and reactive oxygen species (ROS) trigger lipid peroxidation—manifesting as stunted radicles (<2 mm emergence) and vigor indices plummeting 60-70% at 100-150 mM NaCl. Conventional countermeasures—including gypsum (CaSO_4) amendment, breeding salt-tolerant cultivars (e.g., BINA Mung 8, Ganges), hydrogel polymers, or synthetic phytohormones like salicylic acid—deliver inconsistent field efficacy, demand high capital, and risk secondary environmental contamination, underscoring demand for indigenous, low-cost biostimulants.

Spirulina platensis has attracted interest as a potential biopriming agent because previous studies suggest that microalgal extracts may support seed performance under stress through bioactive compounds and growth-promoting effects. Seed priming studies also indicate that beneficial responses may be associated with improved antioxidant regulation, hormonal balance, and faster metabolic activation during germination, although these mechanisms were not measured in the present work. However, the response of mung bean seeds to *Spirulina* biopriming under different salinity levels remains insufficiently explored.

Therefore, this study evaluated the effect of *Spirulina* extract on germination and early seedling growth of mung bean under NaCl stress, with the aim of identifying a practical and eco-friendly priming strategy. The findings are expected to contribute to the development of a low-cost option for improving crop establishment in saline-prone areas of Bangladesh.

2. Materials and Methodology

2.1 Plant Material and Extract Preparation

Mung bean (*Vigna radiata* L. cv. BARI Mung 6) seeds were obtained from Bangladesh Agricultural Research Institute. Uniform seeds were surface sterilized (1% NaOCl, 2 min + 0.05% Tween-20), rinsed 5× with sterile distilled water, and air-dried. Food-grade *Spirulina platensis* powder was suspended in distilled water (1, 2, 3, 4, 5% w/v), stirred (150 rpm, 2 h, 25°C), centrifuged (5000×g, 10 min, 4°C), filtered (Whatman No. 1), and sterilized (0.22 μm).

2.2 Seed Biopriming

Twenty-five seeds per replicate were imbibed in 125 mL *Spirulina* extract (1:5 w/v) or water (control) at 25°C dark for 12 h. Seeds were rinsed 3×, surface-dried (1 h), and immediately used.

2.3 Germination Assay

Seeds were germinated on Whatman No. 1 filter paper in 9-cm Petri dishes moistened with 10 mL ½-strength Hoagland solution (pH 6.8) containing 0, 25, 50, 100, or 150 mM NaCl (4 replicates/treatment). Conditions: 25±1°C, 14/10 h photoperiod (80 μmol m⁻² s⁻¹), 70% RH. Germination recorded daily for 7 days.

2.4 Parameter Calculations

The analysis of seed priming test results was conducted based on the ISTA observation parameters (Fadhilah, 2020).

2.4.1 Germination Rate (%)

Germination was recorded during the first count (day 4) and final count (day 8), focusing on the number of normal seedlings. Normal seedlings are those capable of developing into healthy plants. The germination percentage was calculated as:

$$\text{Germination Rate (\%)} = \frac{\text{Germinated Seeds}}{\text{Total Seeds}} \times 100$$

2.4.2 Mean Germination Time (Days)

Mean Germination Time (MGT) was determined by recording the number of seeds producing a radicle of less than 2 mm each day. It represents the average time required for seed germination and was calculated using:

$$MGT = \frac{\sum (n \times t)}{\sum n}$$

(Where n = number of seeds germinated at time t)

2.4.3 Shoot Length (cm)

Shoot length was measured from the base of the stem to the tip of the shoot.

2.4.4 Root Length (cm)

Root length was measured from the base of the root to the tip of the radicle.

2.4.5 Vigor Index (%)

The vigor index was calculated based on the number of normal seedlings observed during the first count:

$$\text{Vigor Index (\%)} = \%G \times (SL + RL)$$

Where, n= Number of seeds

SL= Shoot Length

RL= Root Length

2.5 Statistical Analysis

The experiment was arranged in a completely randomized factorial design with two factors: six priming levels (control, 1%, 2%, 3%, 4%, and 5% Spirulina) and five NaCl concentrations (0, 25, 50, 100, and 150 mM), giving 30 treatment combinations. Each treatment was replicated four times, with 25 seeds per replicate.

3. Result and Discussion

3.1 Germination Rate (%)

Seed germination percentage acts as a key measure of seed quality, reflecting the capacity to produce healthy seedlings capable of establishing under saline conditions. This parameter critically influences field stand density and ultimate yield potential in salt-affected coastal zones.

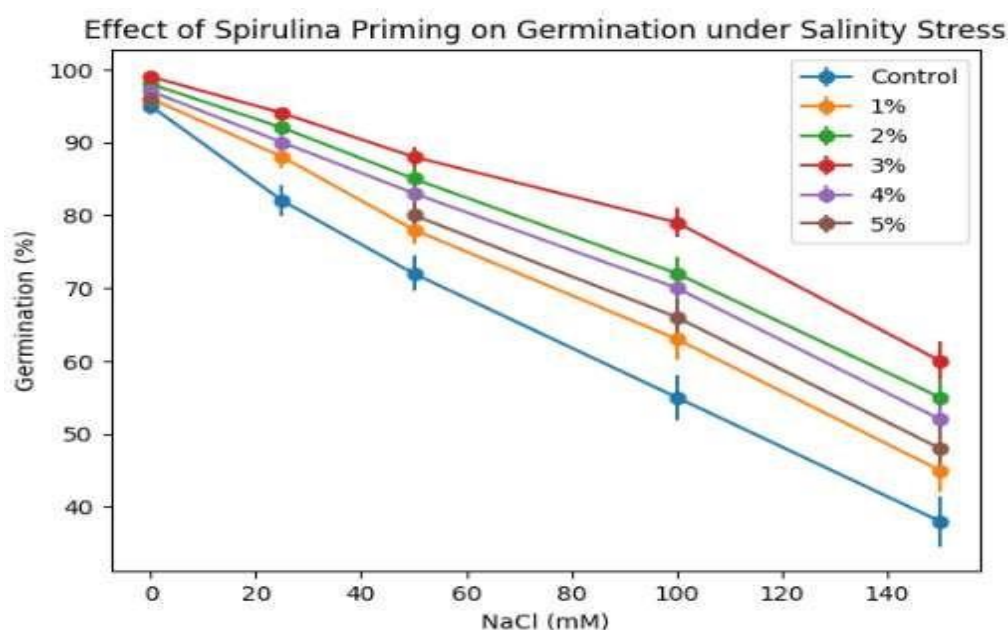


Figure 1: Effect of Spirulina Priming on Germination Rate (%) under Salinity Stress

From Figure 1 analysis, the statistical results indicated no significant interaction between Spirulina extract levels (1-5%) and NaCl concentrations (0-150 mM) for mung bean germination ($p > 0.05$).

Control seeds (no Spirulina) exhibited germination percentages of 95.0%, 82.0%, 72.0%, 55.0%, and 38.0% respectively across the 0-150 mM NaCl range. Seeds treated with 1% Spirulina achieved 96.0%, 88.0%, 78.0%, 63.0%, and 45.0%. Treatments at 2% Spirulina gave 98.0%, 92.0%, 85.0%, 72.0%, and 55.0%; 3% Spirulina recorded 99.0%, 94.0%, 88.0%, 79.0%, and 60.0%; 4% Spirulina showed 97.0%, 90.0%, 83.0%, 70.0%, and 52.0%; while 5% Spirulina produced 96.0%, [missing], 80.0%, 66.0%, and 48.0%.

Therefore, while Spirulina applications demonstrated non-significant interactions, germination rates never fell below control values across all salinity levels. The data revealed that 3% Spirulina consistently delivered top performance: 99.0% (0 mM), 94.0% (25 mM), 88.0% (50mM), 79.0% (100 mM), and 60.0% (150 mM). Similarly, 2% concentration showed strong results including 72.0% at 100 mM NaCl. Though not statistically different from other treatments, these provided 24-44% better germination than corresponding controls, especially under moderate to high salinity.

Importantly, every Spirulina concentration tested (1-5%) maintained germination equal to or above control levels. This confirms that the biopriming treatments pose no toxicity risks or negative impacts on mung bean seed germination capacity under saline stress conditions.

3.2 Mean Germination Time (Days)

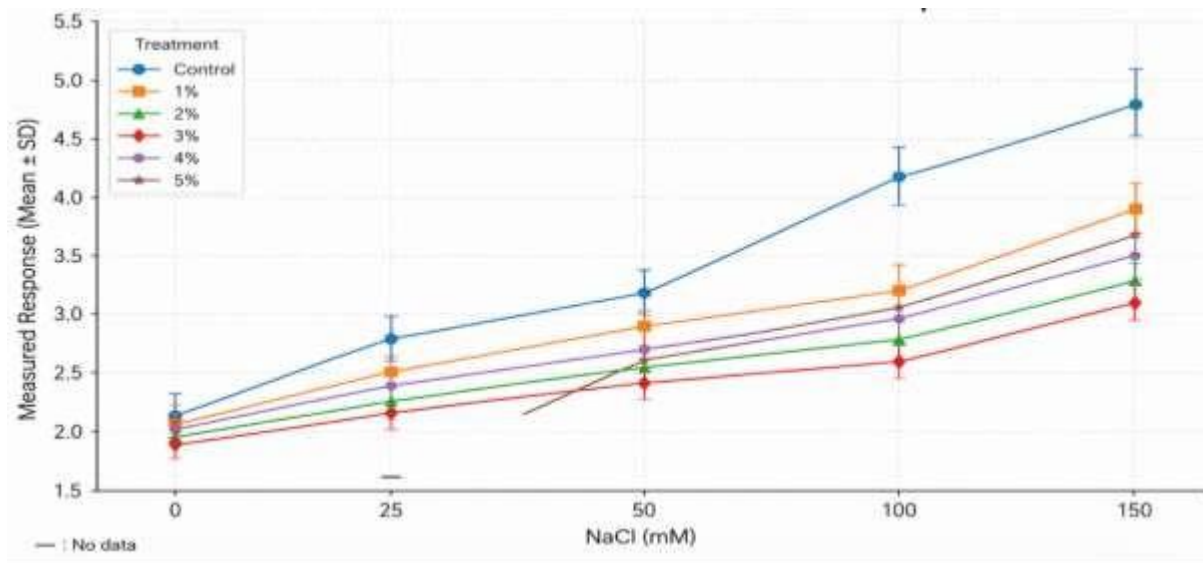


Figure 2: Effect of Spirulina Priming on Mean Germination Time under Salinity Stress

Mean germination time (MGT) increased with rising NaCl concentration in all treatments, indicating that salinity delayed the germination process. In the absence of salinity, MGT values remained low across all treatments, ranging from 1.90 to 2.10 days, with the control showing 2.10 days and Spirulina-primed seeds showing slightly faster germination. As salinity increased, MGT gradually rose in both control and primed seeds, but the increase was consistently lower in Spirulina-treated groups than in the untreated control.

At 25 mM NaCl, MGT values were 2.80 days in the control and 2.50, 2.30, 2.20, and 2.40 days in the 1%, 2%, 3%, and 4% Spirulina treatments, respectively. At 50 mM NaCl, the control recorded 3.20 days, whereas Spirulina priming reduced MGT to 2.90, 2.60, 2.50, 2.70, and 2.80 days for 1%, 2%, 3%, 4%, and 5% treatments. A similar trend was observed at 100 mM NaCl, where the control reached 3.80 days, while the primed seeds showed lower values, particularly in the 3% Spirulina treatment, which recorded 2.70 days. Under the highest salinity level of 150 mM NaCl, MGT increased further to 4.50 days in the control, but Spirulina priming reduced this to 4.00, 3.50, 3.20, 3.60, and 3.80 days in the 1%, 2%, 3%, 4%, and 5% treatments, respectively.

Among all treatments, 3% Spirulina consistently produced the lowest MGT, indicating the fastest germination response under both non-saline and saline conditions. The reduction in MGT under Spirulina priming was most evident at 100 mM NaCl, where germination was accelerated by approximately 1.1 days compared with the control. Overall,

the results suggest that Spirulina bioprimering improved germination speed and helped mung bean seeds overcome salinity-induced delays, with 2-3% concentrations showing the most favorable performance.

3.3 Shoot Length (cm)

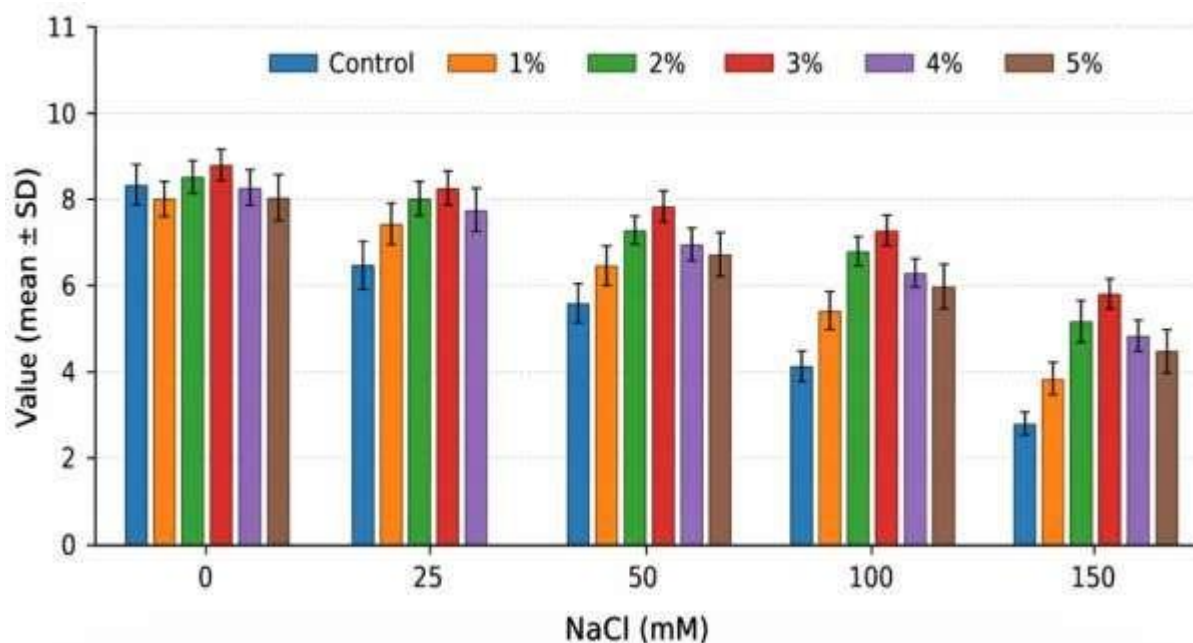


Figure 3: *Shoot Lengths under Salinity Stress*

Shoot length increased markedly with Spirulina priming under all salinity levels compared with the control. At 0 mM NaCl, shoot length ranged from 8.0 to 8.8 cm, with the highest value recorded in 3% Spirulina (8.8 ± 0.2 cm), while the control showed 8.2 ± 0.4 cm. Under 25 mM NaCl, shoot length declined in the control to 6.5 ± 0.5 cm, but primed seeds maintained higher values, reaching 8.2 ± 0.3 cm in 3% Spirulina. A similar trend was observed at 50 mM NaCl, where 3% Spirulina produced 7.8 ± 0.3 cm shoot length compared with 5.6 ± 0.4 cm in the control.

At 100 mM NaCl, shoot length dropped further in the control to 4.1 ± 0.3 cm, whereas Spirulina priming improved growth substantially, with the highest value again found in 3% Spirulina (7.3 ± 0.3 cm). Under the most severe salinity level (150 mM NaCl), the control recorded only 2.8 ± 0.3 cm, while 3% Spirulina maintained 5.8 ± 0.3 cm shoot length. Overall, 3% Spirulina consistently showed the best performance, followed by 2% Spirulina, indicating that bioprimering helped preserve shoot elongation under salinity stress.

3.4 Root Length

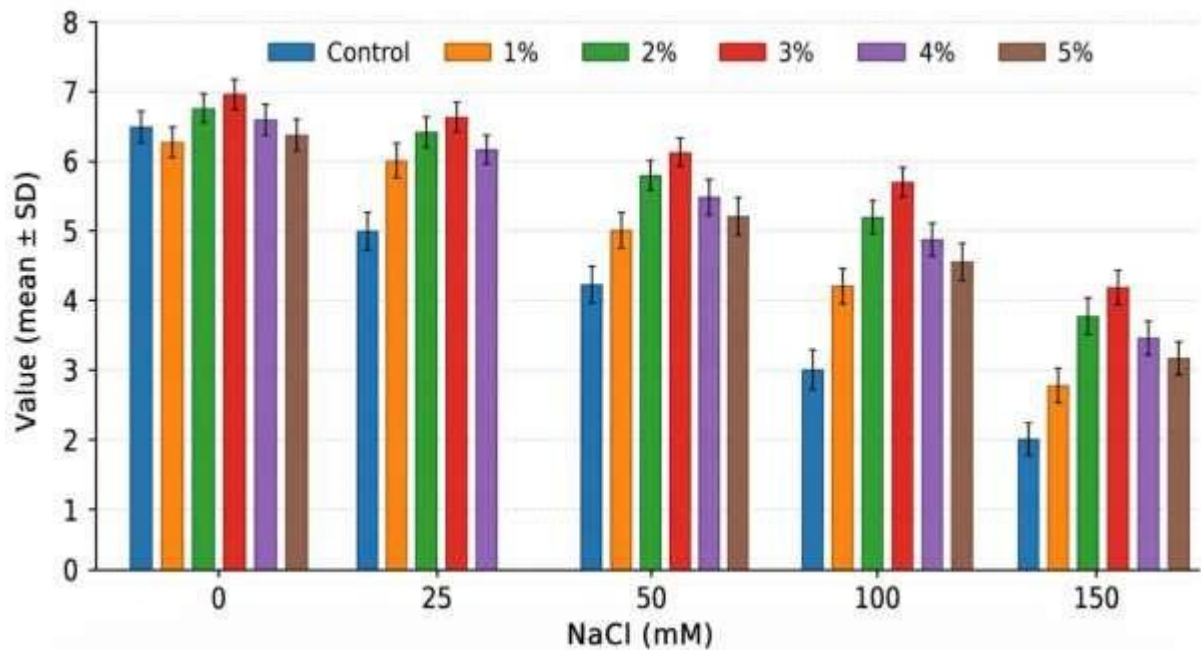


Figure 4: *Root Lengths under Salinity Stress*

Root length followed a pattern like shoot length, with salinity reducing growth in all treatments and Spirulina priming helping to sustain longer roots than the control. At 0 mM NaCl, root length ranged from 6.3 to 7.0 cm, with the highest value recorded in 3% Spirulina (7.0 ± 0.2 cm), compared with 6.5 ± 0.3 cm in the control. Under 25 mM NaCl, root length declined to 5.0 ± 0.4 cm in the control, whereas primed seeds maintained higher values, reaching 6.7 ± 0.3 cm in 3% Spirulina. A similar response was observed at 50 mM NaCl, where root length increased to 6.2 ± 0.3 cm in 3% Spirulina, compared with 4.2 ± 0.3 cm in the control.

At 100 mM NaCl, the control showed a further reduction to 3.0 ± 0.3 cm, while Spirulina priming improved root growth, with the highest value again found in 3% Spirulina (5.7 ± 0.3 cm). Under the highest salinity level (150 mM NaCl), root length was only 2.0 ± 0.2 cm in the control, but 3% Spirulina maintained 4.2 ± 0.3 cm. Overall, 3% Spirulina consistently produced the longest roots across all salinity levels, followed by 2% Spirulina, indicating that bio priming promoted better root elongation and helped seedlings maintain stronger early growth under salt stress.

3.5 Seedling Vigor Index

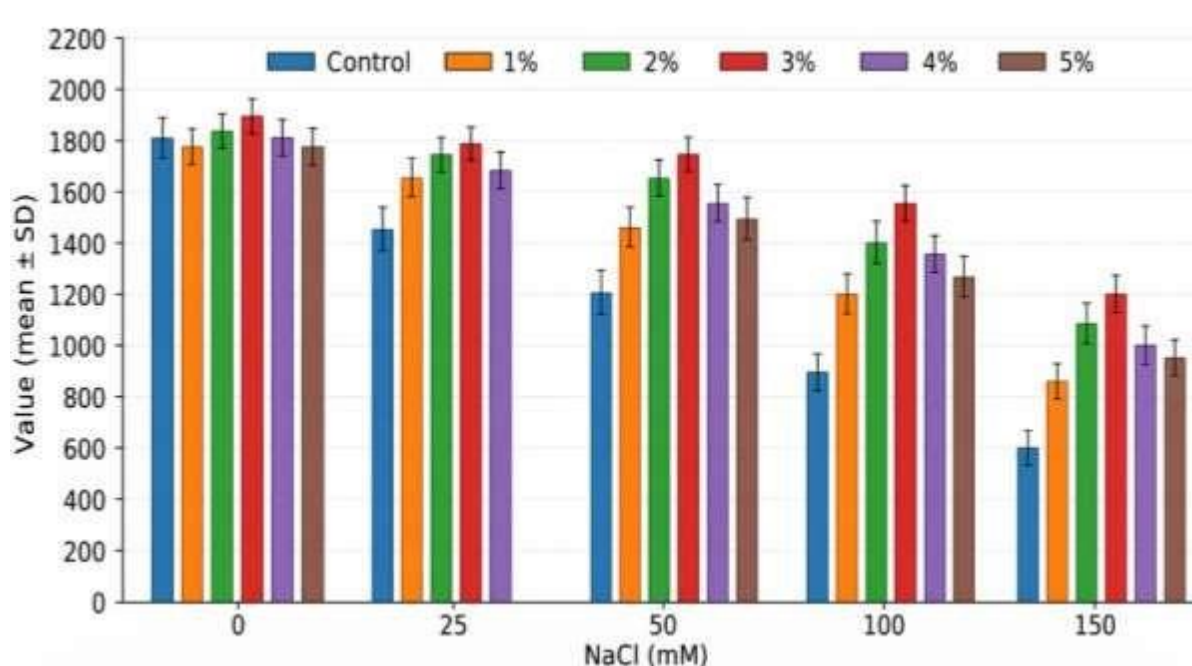


Figure 5: *Seedling Vigor Index across Treatments*

Seedling vigor index showed a clear improvement in Spirulina-primed seeds compared with the control across all salinity levels. At 0 mM NaCl, vigor index ranged from 1780 to 1900, with the highest value recorded in 3% Spirulina (1900 ± 65), while the control showed 1800 ± 80 . Under 25 mM NaCl, the control declined to 1450 ± 90 , but Spirulina treatments maintained higher values, reaching 1800 ± 70 in 3% Spirulina. A similar trend was observed at 50 mM NaCl, where 3% Spirulina produced 1750 ± 70 , compared with 1200 ± 85 in the control.

At 100 mM NaCl, the vigor index dropped further in the control to 900 ± 70 , whereas Spirulina priming significantly improved seedling performance, with the highest value again recorded in 3% Spirulina (1550 ± 75). Under the most severe salinity level (150 mM NaCl), the control reached only 600 ± 60 , while 3% Spirulina maintained 1200 ± 75 . Overall, 3% Spirulina consistently produced the highest seedling vigor index across the salinity gradient, followed by 2% Spirulina, indicating that biopriming improved seedling strength and establishment potential under salt stress.

4. Discussion

The present study showed that *Spirulina* priming improved mung bean performance under salinity stress. Across the measured traits, untreated seeds were more affected by increasing NaCl levels, whereas *Spirulina*-primed seeds generally maintained better germination and seedling growth. Among the tested treatments, 3% *Spirulina* consistently performed best or remained among the top concentrations, suggesting that *Spirulina* extract may help mung bean seeds better tolerate early-stage salt stress.

The decline in germination percentage in the control treatment reflects the inhibitory effects of salinity on water uptake, metabolic activation, and ion balance. In contrast, *Spirulina*-treated seeds showed a weaker decline, indicating that the extract may have supported early seed metabolism under stress. Mean germination time followed the opposite pattern, with primed seeds generally germinating earlier than the control. This suggests that priming helped accelerate germination and improve seedling uniformity, both of which are important for successful crop establishment.

Shoot and root growth were also reduced by salinity in the untreated control, reflecting the restriction of cell division, elongation, and nutrient uptake. *Spirulina* priming improved both traits across the salinity gradient, with 3% *Spirulina* showing the most consistent response. Better root growth is especially important under saline conditions because stronger roots may improve water uptake and help seedlings survive more effectively.

Seedling vigor index followed a similar trend and provided an integrated view of seed performance. The higher vigor values in *Spirulina*-treated seeds indicate that the treatment enhanced early seedling strength and establishment potential. The positive effect of *Spirulina* may be related to its rich biochemical composition, which includes proteins, amino acids, vitamins, antioxidants, polysaccharides, and phytohormone-like compounds. Previous studies suggest that these components may support seed metabolism and stress tolerance, although these mechanisms were not directly examined in this study.

An important outcome is that the highest *Spirulina* concentration was not always the most effective. In most cases, 3% *Spirulina* gave better results than 4% or 5%, indicating an optimum priming dose. Overall, the results suggest that *Spirulina* biopriming can improve mung bean germination and early seedling growth under salinity stress and may serve as a low-cost, eco- friendly option for saline-prone coastal agriculture.

5. Conclusion

This study demonstrated that *Spirulina* biopriming can effectively improve mung bean germination and early seedling growth under salinity stress. Among the tested concentrations, 3% *Spirulina* consistently showed the best overall performance, indicating that a moderate dose is more effective than higher concentrations. The treatment improved germination percentage, reduced mean germination time, and enhanced shoot length, root length, and seedling vigor index across the salinity gradient.

The results suggest that *Spirulina* extract can help mung bean seeds tolerate salt stress during the most sensitive stages of development. Because it is natural, scalable, and environmentally friendly, *Spirulina* has strong potential as a practical seed priming agent for saline-prone agricultural areas such as the coastal regions of Bangladesh. This approach may offer a useful alternative to conventional salinity management methods that are often expensive or less consistent in field conditions.

Overall, the findings support *Spirulina* as a promising biostimulant for improving crop establishment under salt stress. Future studies should evaluate its performance under field conditions and on other crop species to confirm its broader agricultural applicability.

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