

EFFECT OF GIBBERELIC ACID APPLICATION ON THE GERMINATION AND SEEDLING GROWTH OF BARI HYBRID BHUTTA-16 UNDER SALINE CONDITION

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Abstract

The study was conducted at Agronomy Laboratory of Agrotechnology Discipline, Khulna University, Khulna, during August 2022, to assess the germination and seedling growth of BARI Hybrid Bhutta-16 under salinity stress with application of gibberellic acid. The experiment was carried out in a factorial design with two factors: four different concentrations of salt solutions and three different concentrations of gibberellic acid solutions. Salt concentrations were 10 dS/m, 20 dS/m, 30 dS/m and 0 dS/m as control and gibberellic acid concentrations 100 ppm, 150 ppm and 0 ppm as control. Three replications of the experiment were set up using a Completely Randomized Design. Data related to seed germination and seedling growth parameters were collected. Germination and seedling growth parameters decreased with increase in salinity levels but increased with increase in gibberellic acid levels. In case of interaction, there were inconsistent variation in germination parameters; germination percentage, germination capacity, germination energy, germination speed. Seedling growth parameters like root fresh weight and root dry weight showed better results; 436 to 480 mg and 55 to 67 mg, respectively, against different salinity levels with gibberellic acid application. Other parameters like root length, shoot length, shoot fresh weight, shoot dry weight and seedling vigor index were found better upto 20 dS/m salinity with 0, 100 and 150 ppm gibberellic acid but in higher salinity (30 dS/m) their performance was not good.

Key Word: Maize, Salinity stress, Growth regulator, Gibberellic Acid, Seedling growth, Shoot length, BARI Hybrid Bhutta-16.

Introduction

Maize (*Zea mays*) is a cereal grain first domesticated by indigenous people in southern Mexico about 10,000 years ago. A study by Matsuoka *et al.* (2002) and Vigouroux *et al.* (2003) demonstrated that, rather than the multiple independent domestication models, all maize arose from a single domestication in southern Mexico about 9,000 years ago. The study also showed that the highland maize

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varieties from Mexico are the oldest varieties still in existence. Maize becomes a staple food in many parts of the world, with the total production surpassing that of wheat or rice. It is an important C₄ plant from the Poaceae family and moderately sensitive to salt stress (Mass *et al.* 1983 and Solaimani *et al.* 2020). In 2021, total world production was about 1.2 billion tones led by the U.S.A with 31.0% and China 22.4% (FAOSTAT, 2018).

Maize is grown under a wide spectrum of soil and climatic conditions and it is an extremely versatile crop (Fernandez-Armesto, 2011). Soil salinity poses a severe threat to global production of maize for its moderate sensitivity to salt stress. Germination and stand establishment of maize are more sensitive to salt stress than later development stages (Solaimani *et al.* 2020). Symbiotically-interacting rhizosphere microorganisms, assist root plant transformation and mobilization of important nutrients such as nitrogen, phosphorus, potassium and micronutrients (Adal, 2023; Das and Haque, 2025 and Fu *et al.* 2025). High rhizosphere Na⁺ and Cl⁻ decrease plant uptake of N, P, Ca, Mg and Fe. Reduced grain weight and number is responsible for low grain yield in maize under salt stress. Poor kernel setting under salt stress is caused by sink restrictions and decreased acid invertase activity in growing grains. Exclusion of excessive Na⁺ or its compartmentation into vacuoles is an important adaptive strategy for maize under salt stress. Salt resistance in maize is mostly dependent on the up-regulation of genes involved in antioxidant defense and β -expansin proteins. Soils with excess soluble salts or exchangeable Na⁺ in the root zone are termed as salt-affected soils. More than 800 million hectares of land worldwide is affected by either salinity (397 million hectares) or sodicity (434 million hectares) (FAO, 2005). Osmotic stress caused by short-term salt stress on maize plants affects plant growth in the early stages of salt stress (Sümer *et al.* 2004) before lethal Na concentrations are reached (Fortmeier and Schubert, 1995).

Gibberellic acid is a phyto-hormone found in plants and fungi that stimulates cell growth and elongation. Gibberellic acid induces the production of mRNA molecules that code for hydrolytic enzymes in the cells of germination-initiating seeds. Applications of extremely low quantities can have a significant impact while too much will have the reverse result since gibberellic acid controls growth, they can stimulate rapid stem and root growth, induce mitotic division in the leaves of some plants and increase seed germination rates (Miriam, 1976). As a growth regulator or promoter, gibberellic acid has a great capacity of alleviation of salinity stress (Shaddad *et al.* 2013). Application of gibberellic acid significantly promotes plant length and plant fresh/dry biomass while markedly hindered by salinity stress (Hamayun *et al.* 2010).

In view of the above facts, the present research work has been undertaken in the Agronomy laboratory of Agrotechnology Discipline, Khulna University, Khulna,

with the objective to evaluate the germination and seedling growth of BARI Hybrid Bhutta-16 under salinity stress condition with the application of gibberellic acid.

Materials and Methods

BARI Hybrid Bhutta-16 is lodging tolerant due to its short-stature properties. The saline-tolerant BARI Hybrid Bhutta-16 can withstand salinities of up to 8 dS/m in the soil. The experiment was set through the glass petri-dishes of 9 cm in diameter. Seeds were soaked in 0.1% HgCl_2 for 10 minutes in a glass beaker for surface sterilization. After 10 minutes, seeds were rinsed by distilled water for three times. Then the surface sterilized seeds were air dried for 30 minutes. After drying, the seeds were soaked into gibberellic acid (0, 100 and 150 ppm) for 24h. The gibberellic acid treated seeds were placed on the filter papers soaked by 0, 10, 20 and 30 dS/m salt solutions on the petri-dishes. There were three layers of filter papers per petri-dish. In each petri-dish, 12 seeds were placed with the help of forceps. After five days of placement of seeds, salt solution was applied on a regular basis to prevent dehydration. The layer of filter papers could absorb approximately 6 ml of salt solution. It was ensured that there should not be any standing water. The salt solutions were applied as the absorbance level of the layer of filter papers. A Completely Randomized Design (CRD) was followed with three replications in the experiment. Each treatment combination was consisted of four petri-dishes. So, the whole study was performed in altogether 144 petri-dishes. After 15 days of planting the seeds, eight seedlings from each treatment were collected randomly for the measurement of the root length, shoot length, root fresh weight, shoot fresh weight, root dry weight, shoot dry weight and seedling vigor index. After measuring the root length, shoot length, root fresh weight and shoot fresh weight, the plant samples were oven dried for 24h. After 24h, root dry weight and shoot dry weight were recorded.

$$10 \text{ dS/m} = 6.4 \text{ g/L}$$

$$100 \text{ ppm} = 0.1 \text{ g/L}$$

$$\text{Germination Percentage (\%)} = \frac{\text{Total number of seeds germinated}}{\text{Total number of seeds set for test}} \times 100$$

$$\text{Germination Capacity (\%)} = \frac{\text{Number of seeds germinated at 168h}}{\text{Total number of seeds set for test}} \times 100$$

$$\text{Germination Energy (\%)} = \frac{\text{Number of seeds germinated at 72h}}{\text{Total number of seeds set for test}} \times 100$$

$$\text{Germination Speed (\%)} = \frac{\text{Number of seeds germinated at 72h}}{\text{Number of seeds germinated at 168h}} \times 100$$

$$\text{Seedling Vigor Index} = \text{Av. shoot length} + \text{Av. root length} \times \text{Germination percentage}$$

All the growth parameters of seedlings were recorded after 15 days of seed placement. The measurement of root length and shoot length in centimeter (cm) were taken from the collar region to the root tip and shoot tip. The measurement of root fresh weight and shoot fresh weight in gram (g) were recorded immediately after the collection of the seedling samples to prevent the moisture loss. The measurement of root dry weight and shoot dry weight in milligram (mg) were recorded after 24h of oven dry of the root and shoot collected from seedling samples. All the data collected on different parameters were statistically analyzed following the analysis of variance (ANOVA) technique using Statistix 10 computer package program. The mean differences were adjusted at 1% and 5% level of significance using the Duncan's New Multiple Range Test (DMRT).

Results and Discussion

Germination Percentage

Results showed that germination percentage differed from 79.17% to 91.67% (Table 1 to Table 3). The highest germination (91.67%) was found at salinity level of 0 dS/m and gibberellic acid level of 100 ppm and 150 ppm. The lowest germination (79.17%) was found in levels of 20 dS/m salinity and 0 ppm gibberellic acid (control). Gibberellic acid can mitigate moderate level of salinity stress in seed germination. It is not effective in high salinity condition.

Germination Capacity

Results showed that germination capacity differed from 79.17% to 91.67% (Table 1 to Table 3). The highest germination capacity (91.67%) was found at salinity level of 0 dS/m and gibberellic acid level of 100 ppm and 150 ppm. The lowest germination capacity (79.17%) was found in levels of 20 dS/m salinity and 0 ppm gibberellic acid (control). Germination capacity sharply decreased with the increase of salinity level and decrease of gibberellic acid level.

Germination Energy

Results showed that germination energy differed from 65.97% to 84.72% (Table 1 to Table 3). The highest germination energy (84.72%) was found at salinity level of 0 dS/m and gibberellic acid level of 150 ppm on the other hand the lowest germination energy (65.97%) was recorded in levels of 20 dS/m salinity and 100 ppm gibberellic acid. Germination energy sharply decreased with the increase of salinity level.

Germination Speed

Results showed that germination speed differed from 81.58% to 94.63% (Table 1 to Table 3). The highest germination speed (94.63%) was found at salinity level of

0 dS/m and gibberellic acid level of 0 ppm (control) on the other hand the lowest germination speed (81.58%) was found in levels of 30 dS/m salinity and 100 ppm gibberellic acid. Germination speed sharply decreased with the increase of salinity level.

Germination parameters like germination percentage, germination capacity, germination energy and germination speed were significantly affected by salinity levels showing a lower value with higher salinity that agreed with El-Kamar *et al.* (2013) and Ali and Hamza (2014). However, these parameters did not show significantly variable response due to different levels of gibberellic acid application.

Significant variation was found for the interaction between salinity and gibberellic acid levels in relation to germination percentage (Table 3). Interaction effect of salinity and gibberellic acid responded significantly on germination parameters but were not consistent and did not maintain a definite trend.

Table 1. Effect of salinity levels on seed germination parameters of BARI Hybrid Bhutta-16

Salinity level (EC dS/m)	Germination Percentage (%)	Germination Capacity (%)	Germination Energy (%)	Germination Speed (%)
S ₀	90.5 a	90.5 a	84.02 a	92.92 a
S ₁	87.5 ab	87.5 ab	80.78 ab	92.35 ab
S ₂	82.87 b	82.87 b	72.91 c	87.83 ab
S ₃	84.49 b	84.49 b	73.61 bc	86.74 b
CV (%)	6.37	6.37	9.85	6.47
Level of Significance	*	*	*	*

Table 2. Effect of gibberellic acid levels on seed germination parameters of BARI Hybrid Bhutta-16

GA ₃ level (ppm)	Germination Percentage (%)	Germination Capacity (%)	Germination Energy (%)	Germination Speed (%)
G ₀	85.59	85.59	78.12	91.16
G ₁	85.41	85.41	75.17	87.67
G ₂	88.02	88.02	80.2	91.0658
CV (%)	6.37	6.37	9.85	6.47
Level of Significance	NS	NS	NS	NS

Table 3. Interaction effect of salinity and gibberellic acid levels on seed germination parameters of BARI Hybrid Bhutta-16

Salinity level (EC dS/m)	GA ₃ level (ppm)	Germination Percentage (%)	Germination Capacity (%)	Germination Energy (%)	Germination Speed (%)
S ₀	G ₀	88.19 a-c	88.19 a-c	83.333 ab	94.63 a
S ₀	G ₁	91.67 a	91.67 a	84.03 a	91.653 ab
S ₀	G ₂	91.67 a	91.67 a	84.72 a	92.49 ab
S ₁	G ₀	86.11 a-c	86.11 a-c	79.167 a-c	91.95 ab
S ₁	G ₁	86.8 a-c	86.8 a-c	81.943 ab	94.34 a
S ₁	G ₂	89.583 ab	89.583 ab	81.25 a-c	90.783 a-c
S ₂	G ₀	79.86 c	79.86 c	70.833 b-d	88.943 a-c
S ₂	G ₁	79.17 c	79.17 c	65.97 d	83.123 bc
S ₂	G ₂	89.587 ab	89.587 ab	81.943 ab	91.443 ab
S ₃	G ₀	88.19 a-c	88.19 a-c	79.170 a-c	89.107 a-c
S ₃	G ₁	84.03 a-c	84.03 a-c	68.747 cd	81.58 c
S ₃	G ₂	81.25 bc	81.25 bc	72.917 a-d	89.547 a-c
CV (%)		6.37	6.37	9.85	6.47
Level of Significance		*	*	*	*

The figures having same letter(s) in a column are not significantly different by DMRT.

When compared to non-stimulated seed, seedlings from stimulated with gibberellic acid exhibited greater germination and other characteristics. They also had lower ratios of salt to potassium and higher chlorophyll content, two chemical indications of increased plant tolerance to salinity that agreed with Ali and Hamza (2014).

Root Length: Results showed that root length of seedlings differed from 8.88 cm to 19.75 cm (Table 4 to Table 6). The highest root length of seedlings (19.75 cm) was found at salinity level of 0 dS/m and gibberellic acid level of 0 ppm (control), which was statistically similar to that of salinity level of 0 dS/m and gibberellic acid level of 150 ppm. The lowest root length of seedlings (8.88 cm) was found in salinity levels of 30 dS/m and 100 ppm gibberellic acid. Root length of seedlings sharply decreased with the increase of salinity level and decrease of gibberellic acid level.

Shoot Length: Results showed that shoot length of seedlings varied significantly and ranged from 21.92 cm to 42.63 cm (Table 4 to Table 6). The highest shoot length of seedlings (42.63 cm) was found at salinity level of 0 dS/m and gibberellic acid level of 150 ppm. The lowest shoot length of seedlings (21.92 cm) was

recorded in levels of 30 dS/m salinity and 0 ppm gibberellic acid (control). Shoot length of seedlings sharply decreased with the increase of salinity level and decrease of gibberellic acid level.

Root Fresh Weight: Results showed that root fresh weight of seedlings differed from 0.32 g to 0.54 g (Table 4 to Table 6). The highest root fresh weight of seedlings (0.54 g) was found at salinity level of 30 dS/m and gibberellic acid level of 0 ppm (control) that was similar with the salinity level of 0 dS/m and gibberellic acid level of 0 ppm (0.53 g). The lowest root fresh weight of seedlings (0.32 g) was found in levels of 0 dS/m salinity (control) and 100 ppm gibberellic acid.

Shoot Fresh Weight: Results showed that shoot fresh weight of seedlings differed from 0.389 g to 0.861 g (Table 4 to Table 6). The highest shoot fresh weight of seedlings (0.861 g) was found at salinity level of 0 dS/m (control) and gibberellic acid level of 150 ppm. The lowest shoot fresh weight of seedlings (0.389 g) was found in levels of 30 dS/m salinity and 100 ppm gibberellic acid. Shoot fresh weight of seedlings sharply decreased with the increase of salinity level and decrease of gibberellic acid level.

Root Dry Weight: Results showed that root dry weight of seedlings differed from 52 mg to 74 mg (Table 4 to Table 6). The highest root dry weight of seedlings (74 mg) was found at salinity level of 0 dS/m (control) and gibberellic acid level of 0 ppm (control). The lowest root dry weight of seedlings (52 mg) was found in levels of 30 dS/m salinity and 100 ppm gibberellic acid.

Shoot Dry Weight: Results showed that shoot dry weight of seedlings differed from 46 mg to 79 mg (Table 4 to Table 6). The trend is similar to that of shoot fresh weight. The lowest shoot dry weight of seedlings (46 mg) was found in level of 20 dS/m salinity and 150 ppm gibberellic acid. Shoot dry weight of seedlings sharply decreased with the increase of salinity level.

Seedling Vigor Index: Results showed that seedling vigor index of seedlings differed from 748.6 to 1775.2 (Table 4 to Table 6). The highest seedling vigor index of seedlings (1775.2) was found at salinity level of 0 dS/m (control) and gibberellic acid level of 0 ppm (control). The lowest seedling vigor index of seedlings (748.6) was found in levels of 30 dS/m salinity and 150 ppm gibberellic acid. Seedling vigor index of seedlings sharply decreased with the increase of salinity level.

Seedling growth parameters were significantly affected due to increase in salinity levels with an exception in root fresh weight. Gibberellic acid application influenced some seedling growth parameters like shoot length, root fresh weight, root dry weight and shoot dry weight that agreed with Shahzad *et al.* (2021) but differed with Khan and Ansari (1998).

Results showed significant variation in the interaction between salinity and gibberellic acid levels in relation to root length of seedlings (Table 6). Interaction effect of salinity and gibberellic acid showed significant variation on all the seedling growth parameters.

Remarkable improvement was not found in root length, shoot length, shoot fresh weight, shoot dry weight and seedling vigor index due to gibberellic acid application in high salinity level (30 dS/m) but gibberellic acid improved the salinity effect on root fresh weight and root dry weight.

In our study we applied high levels of salinity (0-30 dS/m) and it was observed that gibberellic acid application can alleviate salinity effect on seedling growth upto 20 dS/m and in case of higher salinity (30 dS/m) it was ineffective.

Table 4. Effect of salinity levels on seedling growth parameters of BARI Hybrid Bhutta-16

Salinity level (EC dS/m)	Root Length (cm)	Shoot Length (cm)	Root Fresh Weight (g)	Shoot Fresh Weight (g)	Root Dry Weight (mg)	Shoot Dry Weight (mg)	Seedling Vigor Index
S ₀	17.72 a	38.65 a	0.48	0.83 a	67 a	73 a	1641.41 a
S ₁	12.61 b	35.43 b	0.45	0.69 b	63 ab	70.4 a	1144.4 b
S ₂	10.06 c	28.05 c	0.438	0.48 c	55 b	49.8 b	861.38 c
S ₃	9.42 c	24.19 d	0.436	0.43 c	61 ab	49.5 b	817.97 c
CV (%)	14.08	6.7	15.83	13.17	13.16	13.4	15.7
Level of Significance	*	*	NS	*	*	*	*

Table 5. Effect of gibberellic acid levels on seedling growth parameters of BARI Bhutta-16

GA ₃ level (ppm)	Root Length (cm)	Shoot Length (cm)	Root Fresh Weight (g)	Shoot Fresh Weight (g)	Root Dry Weight (mg)	Shoot Dry Weight (mg)	Seedling Vigor Index
G ₀	13.13	26.39 b	0.49 a	0.61	66 a	57 b	1156.94
G ₁	12.15	34.08 a	0.4 b	0.59	58 b	64 a	1083.72
G ₂	12.09	34.28 a	0.45 ab	0.64	62 ab	62 a	1108.21
CV (%)	14.08	6.70	15.83	13.17	13.16	13.40	15.70
Level of Significance	NS	*	*	NS	*	*	NS

Table 6. Interaction effect of salinity and gibberellic acid levels on seedling growth parameters of BARI Hybrid Bhutta-16

Salinity level (EC dS/m)	GA ₃ level (ppm)	Root Length (cm)	Shoot Length (cm)	Root Fresh Weight (g)	Shoot Fresh Weight (g)	Root Dry Weight (mg)	Shoot Dry Weight (mg)	Seedling Vigor Index
S ₀	G ₀	19.75 a	31.16 c	0.53 a	0.85 a	74 a	67.1 ab	1775.2 a
S ₀	G ₁	16.06 b	42.17 a	0.32 c	0.79 ab	60.4 a-c	75.8 a	1517.17 a
S ₀	G ₂	17.35 ab	42.63 a	0.46 ab	0.861 a	69 ab	79 a	1631.903 a
S ₁	G ₀	12.15 c-e	28.9 cd	0.44 ab	0.68 b	65 a-c	60 bc	1083.98 b-d
S ₁	G ₁	13.08 c	37.77 b	0.482 ab	0.66 b	61 a-c	76 a	1176.284 b
S ₁	G ₂	12.63 cd	39.6 ab	0.458 ab	0.76 ab	63 a-c	75 a	1172.956 bc
S ₂	G ₀	10.1 d-f	23.5 ef	0.483 ab	0.49 c	57 bc	50 cd	828.6 de
S ₂	G ₁	10.6 c-f	31.98 c	0.439 ab	0.52 c	58 bc	54 b-d	876.247 de
S ₂	G ₂	9.48 ef	28.6 cd	0.394 bc	0.47 c	53 c	46 d	879.35 c-e
S ₃	G ₀	10.5 c-f	21.92 f	0.54 a	0.446 c	67 ab	51 cd	940.096 b-e
S ₃	G ₁	8.88 f	24.4 ef	0.398 bc	0.389 c	52 c	48.8 cd	765.187 e
S ₃	G ₂	8.9 f	26.3 de	0.51 ab	0.47 c	64 a-c	49.2 cd	748.6 e
CV (%)		14.08	6.7	15.83	13.17	13.16	13.4	15.7
Level of Significance		*	*	*	*	*	*	*

The figures having same letter(s) in a column are not significantly different by DMRT.

EC = Electrical Conductivity, S₀ = 0 dS/m, S₁ = 10 dS/m, S₂ = 20 dS/m, S₃ = 30 dS/m, G₀ = 0 ppm, G₁ = 100 ppm, G₂ = 150 ppm, CV = Coefficient of Variation, * Significant variation shown by DMRT.

Conclusion

Results indicated that germination and seedling growth parameters decreased with the increased effect of salinity levels. It also indicated that germination and seedling growth parameters increased with the increased effect of gibberellic acid levels. It further indicated that salinity decreases germination and seedling growth. Gibberellic acid showed great influence on shoot length increase but does not enhance root growth. So, the interaction effect of salinity levels and gibberellic acid levels showed no variation. Gibberellic acid was effective to alleviate salinity stress up to 10 dS/m on germination and seedling growth parameters. Seedling shoot length was found better with 100 ppm of gibberellic acid for 20 dS/m of salinity. Over application of gibberellic acid might be the reason behind deterioration of the growth of the seedlings that supports Zhu *et al.* (2019), as the growth regulators have trace amount of necessity for the plants. As BARI Hybrid

Bhutta-16 can tolerate up to 8 dS/m salinity level, the performance in 30 dS/m salinity level is very poor. At the level of 20 dS/m salinity stress, the seedlings of BARI Hybrid Bhutta-16 can show some better performance with the help of 100 ppm gibberellic acid concentration by alleviating the salinity stress.

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