Table 3: Effect of HBL and EBL on mannitol content (μ mol g^{-1} FW) of 30 days old plant of *Zea mays* subjected to salt stress.

BRs Treatment	0 mMNaCl	40mM NaCl	60mM NaCl	80mM NaCl	100mM NaCl
0M	12.32±0.64	12.59±1.194	29.37±2.610	39.88±5.291	36.64±3.124
10^{-10}M HBL	19.97±1.399	25.11±3.520	37.23±3.649	46.50±3.149	36.84±3.540
10^{-8}M HBL	15.84±0.745	25.28±2.927	45.91±2.322	56.36±3.550	33.58±2.812
10^{-6}M HBL	17.33±0.621	15.23±1.547	39.30±2.033	40.21±4.930	50.30±2.590
Treatment Dose Treatment× Dose F-ratio 88.205* 12.93* 4.960*					
10^{-10}M EBL	14.96±2.579	29.38±1.572	37.09±3.471	38.36±4.416	53.60±2.997
10^{-8}M EBL	27.36±3.670	31.67±1.889	35.13±2.309	50.90±5.226	56.60±3.20
10^{-6}M EBL	33.41±4.130	19.16±5.078	26.89±2.886	46.58±4.931	58.91±4.212
Treatment Dose Treatment× Dose F-ratio 123.46* 12.903* 2.388					

*Indicate statistically significant differences from control at p≤0.05

Table 4: Effect of HBL and EBL on total sugar content (μ mol g^{-1} FW) of 30 days old plant of *Zea mays* subjected to salt stress

BRs Treatment	0 mMNaCl	40mM NaCl	60mM NaCl	80mM NaCl	100mM NaCl
0M	**1.543±0.045**	**1.687±0.095**	**1.428±0.054**	**1.745±0.051**	**2.137±0.057**
10^{-10}M HBL	1.640±0.051	2.424±0.286	1.661±0.045	1.944±0.029	3.160±0.075
10^{-8}M HBL	1.738±0.057	2.718±0.091	1.849±0.061	2.455±0.123	3.518±0.172
10^{-6}M HBL	1.905±0.080	1.915±0.035	1.565±0.032	2.029±0.129	3.014±0.125
Treatment Dose Treatment× Dose F-ratio 119.29* 51.542* 6.163*					
10^{-10}M EBL	2.075±0.110	3.537±0.288	1.900±0.073	1.741±0.251	2.578±0.166
10^{-8}M EBL	2.484±0.242	2.611±0.132	1.678±0.144	2.459±0.235	3.283±0.217
10^{-6}M EBL	1.560±0.201	2.279±0.171	1.811±0.022	2.656±0.083	2.763±0.296
Treatment Dose Treatment× Dose F-ratio 384.57* 53.367* 42.660*					

***Indicate statistically significant differences from control at p≤0.05**

Table5: Effect of HBL and EBL on proline content (μ mol g^{-1} FW) of 60 days old plant of *Zea mays* subjected to salt stress.

BRs Treatment	0 mMNaCl	40mM NaCl	60mM NaCl	80mM NaCl	100mM NaCl
0M	**27.73±4.967**	**46.06±4.561**	**39.23±5.108**	**56.31±6.511**	**60.61±6.162**
10^{-10}M HBL	45.25±1.276	64.60±4.366	46.01±3.790	74.21±3.478	59.73±6.217
10^{-8}M HBL	35.50±2.754	56.60±3.271	61.00±1.159	49.97±6.569	74.61±2.628
10^{-6}M HBL	28.29±3.429	44.63±2.623	64.42±4.384	64.71±3.908	73.91±2.475
Treatment Dose Treatment× Dose F-ratio 13.592* 11.899* 4.956*					
10^{-10}M EBL	42.80±3.696	60.63±5.748	48.48±4.444	74.15±2.899	63.59±2.976
10^{-8}M EBL	33.66±3.484	55.96±1.820	37.31±3.693	76.48±7.626	73.54±3.111
10^{-6}M EBL	26.83±3.339	62.26±3.712	57.38±4.400	62.37±3.903	63.56±3.859
Treatment Dose Treatment× Dose F-ratio 49.188* 11.672* 2.306					

*Indicate statistically significant differences from control at p≤0.05

Table 6: Effect of HBL and EBL on glycine betaine content (μ mol g^{-1} FW) of 60 days old plant of *Zea mays* subjected to salt stress

BRs Treatment	0 mMNaCl	40mM NaCl	60mM NaCl	80mM NaCl	100mM NaCl
0M	**23.57±1.767**	**25.01±2.068**	**35.14±2.424**	**29.02±5.248**	**47.84±1.278**
10^{-10}M HBL	42.96±3.358	34.92±7.346	58.57±4.598	43.66±4.507	58.67±4.261
10^{-8}M HBL	32.90±5.360	58.35±4.592	62.30±3.543	65.20±2.637	73.51±2.385
10^{-6}M HBL	24.95±1.464	49.31±5.511	48.28±4.344	54.37±4.573	64.73±2.892
Treatment Dose Treatment× Dose F-ratio 29.377* 47.361* 6.387*					
10^{-10}M EBL	44.68±2.610	46.78±3.878	50.07±4.808	43.08±3.342	61.53±2.461
10^{-8}M EBL	22.39±1.654	46.64±3.176	45.31±2.648	28.40±4.518	53.88±2.20
10^{-6}M EBL	20.66±2.328	30.06±5.794	39.22±0.704	36.13±2.197	49.41±3.104

	Treatment	Dose	Treatment× Dose
F-ratio	41.70*	23.042*	4.953*

*Indicate statistically significant differences from control at p≤0.05

Table 7: Effect of HBL and EBL on mannitol content (μ mol g^{-1} FW)of 60 days old plant of *Zea mays* subjected to salt stress.

BRs Treatment	0 mMNaCl	40mM NaCl	60mM NaCl	80mM NaCl	100mM NaCl
0M	**25.41±2.647**	**54.44±2.234**	**40.95±5.750**	**42.21±3.784**	**59.56±5.424**
10^{-10}M HBL	30.25±1.732	56.32±3.605	58.89±2.351	65.84±4.72	66.33±3.452
10^{-8}M HBL	36.25±1.732	67.16±4.102	59.75±5.893	75.87±2.834	76.61±1.836
10^{-6}M HBL	29.02±4.394	59.36±7.809	56.64±3.704	62.57±5.114	62.81±4.915
Treatment Dose Treatment× Dose F-ratio 76.898* 19.442* 5.252*					
10^{-10}M EBL	32.84±2.930	45.15±2.707	50.33±0.671	64.51±1.709	60.65±3.675
10^{-8}M EBL	28.39±4.285	45.69±3.640	56.68±3.901	69.54±4.21	71.92±1.820
10^{-6}M EBL	44.43±2.909	56.32±3.605	48.00±4.785	59.96±4.42	66.39±3.932
Treatment Dose Treatment× Dose F-ratio 153.19* 22.087* 10.637*					

*Indicate statistically significant differences from control at p≤0.05

Table 8: Effect of HBL and EBL on total sugar content (μ mol g^{-1} FW) of 60 days old plant of *Zea mays* subjected to salt stress.

BRs Treatment	0 mMNaCl	40mM NaCl	60mM NaCl	80mM NaCl	100mM NaCl
0M	**2.579**±0.143	**3.785±0.145**	**3.254±0.143**	**4.486±0.236**	**3.058±0.108**
10^{-10}M HBL	3.228±0.241	3.344±0.264	4.261±0.222	5.484±0.238	5.185±0.092
10^{-8}M HBL	3.250±0.145	3.817±0.117	6.714±0.203	7.663±0.181	4.457±0.118
10^{-6}M HBL	3.355±0.213	5.185±0.219	5.625±0.119	4.933±0.064	3.650±0.142
Treatment Dose Treatment× Dose F-ratio 125.04* 105.79* 22.61*					
10^{-10}M EBL	3.247±0.173	4.287±0.175	3.820±0.528	6.581±0.225	4.049±0.090
10^{-8}M EBL	3.940±0.393	4.834±0.438	5.349±0.373	5.450±0.263	3.903±0.473
10^{-6}M EBL	4.294±0.179	4.047±0.553	2.454±0.230	5.566±0.264	4.249±0.338
Treatment Dose Treatment× Dose F-ratio 27.313* 14.75* 4.991*					

***Indicate statistically significant differences from control at p≤0.05**

4. Results and Discussion

Salt stress imposition resulted in increase of osmolytes content (proline, glycine betaine, mannitol and total sugar content) in both 30 and 60 days old *Zea mays* plants. In 30 days old plants, maximum proline content was recorded in 100 mM salt stressed plants (22.60 μ mol g^{-1} FW) in comparison to control plants (16.90 μ mol g^{-1} FW). Further treatment of HBL (10^{-8} M) along with NaCl (60mM) showed the maximum increase of proline content (1.48 times) as compared to NaCl alone. Similarly application of EBL (10^{-10}M) in conjunction with NaCl (40mM) enhanced the proline content 1.40 times as compared to NaCl alone (Table 1). However in 60 days old plants, osmolytes content were found to increase as compared to 30 days old plants. Application of HBL (10^{-8}M) and EBL (10^{-8}M) both ameliorated the toxic effect of salt stress by enhancing the proline content (1.23 & 1.35 times respectively) under 100 mM and 80 mM NaCl alone respectively (Table 5).

Thus from above results it was reported that application of BRs (HBL and EBL) enhanced the proline accumulation under stress conditions which reveals that it play an important role in osmotic adjustment. It induces the expression of genes responsible for the biosynthesis of proline and overcome the salinity stress by regulating the proline content in plants which provide protection to the sub-cellular structures by reducing the oxidative damage caused due to free radicals in response to salt stress [27, 28, 29, 30, 31]. BRs application increased the proline content in rice plants subjected to drought stress by overcoming the deleterious effects of salt [32]. Similarly Hayat *et al.* [33] also reported the enhanced proline accumulation with the treatment of BRs in *Lycopersicon esculentum* when subjected to cadmium stress.

Glycine betaine and mannitol content was also found to increase under salt stress as compared to control. In 30 days old plant, glycine betaine (2.73 folds) and mannitol content (3.23 times) was observed to be increased under 100 and 80 mM salt stress respectively in comparison to control. However supplementation of (10^{-8} M) HBL with NaCl stress maximally enhanced the glycine betaine (1.69 fold) content under 100 mM salt stress and mannitol content (1.41 times) under 80 mM salt stress as compared to NaCl alone. Similarly treatment of EBL also enhanced the glycine betaine content 1.68 times and mannitol content 1.60 times under 100mM salt stress (Table 2, 3).

In 60 days old plants, application of HBL (10^{-8}M) enhanced the glycine betaine content (2.24 folds) under 80 mM salt stress and application of EBL (10^{-10}M) increased the glycine betaine content (1.28 folds) under 100 mM salt stress. Similarly maximum increase of mannitol content was recorded 1.28 folds with the application of HBL (10^{-6}M) and 1.20 folds with EBL (10^{-8}M) application along with 100mM salt stress (Table s6, 7).

Glycine betaine and mannitol both as osmoprotectant play important role in stress alleviation. Glycine betaine occurs abundantly in chloroplast where it provides protection to the thylakoid membranes and maintains the photosynthetic efficiency [34]. Mannitol serves as free radical scavenger and also stabilizing sub cellular structures and plays role in storage of carbon and energy [35]. BRs application enhanced the accumulation of osmoprotectents which is a means to counter the adverse effect of stress [36]. In the present study, BRs application enhanced the both glycine betaine and mannitol content under stress condition. Ali and Abdel Fattah, [37] reported that BRs treatment increased the glycine betaine content under salt stress in *Phaseolus vulgaris* and *Hordeum vulgare* by activating the enzyme betaine aldehyde dehydrogenase (BADH) which catalyse the synthesis of glycine betaine from the choline and enhanced the accumulations.

Sugar content was also found to increase under salt stress in both 30 and 60 days old plants. Supplementation of 30 days old plant with HBL (10^{-8}M) plus NaCl showed the enhancement of sugars content (1.64 times) under 100 mM NaCl stress whereas application of EBL (10^{-10}M) increased the sugar content about 2.09 times under 40mM salt stress (Table 4). In 60days old plants, presoaking treatment of HBL along with NaCl stress increased the sugar content 1.70 folds and EBL application enhanced the sugar content 1.46 times both under 80 mM salt stress (Table 8). Enhancement of sugars with the application of BRs provides tolerance against salt stress. Verma *et al.* [38] reported an increased sugar level with the treatment of BRs in *Arachis hypogaea*. Similarly Vardhini *et al.* [39] also reported the increased carbohydrate fractions like reducing sugars and starch in the radish roots with the treatment of BRs.

5. Conclusion

It was concluded from present study that osmolytes production during salt stress is considered as very important in view of its role in stress tolerance. Further BRs application overcome the salinity stress by enhancing the osmoytes accumulation and thus developed the tolerance against salinity stress.

6. Future Scope

Plants often experience various abiotic and biotic stresses like drought, high or low temperature, flooding, salinity, metal toxicity, UV-radiations, herbicides and pathogen stress which adversely affected the crop production and yield. Plants adopted various strategies to adopt the stress conditions and osmolytes accumulation is one of among defensive stratregies. Application of BRs at appropriate dose further develops the stress tolerance by enhancing the accumulation of osmolytes production which helps the plants to overcome the stress conditions. Thus our study will further helpful to study the complex and fine mechanisms of osmolytes participation in the creation of resistant plants and help to explore the fundamental signaling mechanism of BRs induced plant stress protection in abiotic stressed plants.

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