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Analysis of Morpho-physiological and Biochemical Responses in *Brassica* Varieties Under Salt Stress

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Abstract

Abiotic stresses like salinity, water stress, high light intensity, heavy metals, etc. have a significant influence on the morphology, biochemical characteristics, and crop productivity, of *Brassica* species. Among these numerous abiotic stresses, stress from salinity is a significant issue, that demands immediate attention and resolution. Therefore, in this study, the tolerance of *Brassica carinata* and five different varieties of *Brassica campestris* including Agrani, Shafal, BINA Sharisha-6, BARI Sharisha-15 and Tori-7 were examined at varying concentrations of NaCl (0, 50, 100, 150, 200 mM). Different morpho-physiolwasal and biochemical properties including seed germination parameters, root/shoot ratio, proline content, H_2O_2 and MDA level, photosynthetic pigments, and Na^+/K^+ ion accumulation were measured and showed varying responses to the inhibitory impacts of NaCl. *B. carinata*, BINA Sharisha-6, Tori-7 and Shafal, with increasing NaCl concentrations resulted in the loss of germination percentages, root/shoot ratio, levels of photosynthetic pigments, proline, levels of H_2O_2 and MDA and the $Na^{+/}K^+$ ion accumulation in comparison to the other *Brassica* varieties. Among these varieties, Shafal showed the lowest values of the salt tolerant indicators. In contrast, variety Agrani and BARI Sharisha-15 exhibited high proline content, a good level of H_2O_2 and MDA and photosynthetic pigments, in response to salt stress, which enhanced their hydration status, enzyme activities and cellular functions.

Introduction

The *Brassica* genus belongs to the large plant family Brassicaceae and is a crucial edible oil seed crop in Bangladesh. Oilseed *Brassica* holds a position as one of the foremost sources of industrial oil, edible vegetable oil and protein-rich goods globally. It contains

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about 40-45% oil and 20-25% protein. It plays a significant role in human health especially for heart patients as it contains omega 3 and fatty acid compositions (linoleic and alpha linoleic acid, respectively) (Goswami et al. 2020).

Currently, *Brassica* ranks as the third most significant oil crop regarding the production area. In each year, the combined production of rapeseed and mustard seeds amounts to 63.04 million tons, which are collected from 34.33 million hectares of land globally (FAO 2013). Bangladesh, with a substantial demand for edible oil, particularly relies on *Brassica* and in terms of production area, *Brassica* holds the lead position among the oilseed crops cultivated in this country.

Crop quality and productivity are significantly influenced by the environment. The effects of climate change on crop growth are intricate and uncertain. Abiotic stresses, including salinity, heat, cold, scarcity of water, heavy metals, nutrient deficiency etc. have a great impact on world agriculture and are major contributors to yield reduction. Among these stresses, one of the most detrimental environmental factors is salinity, as it drastically impacts plant growth by altering metabolic activities (Rahim et al. 2020). Different physiological, morphological, and biochemical properties of plants are induced by salinity stress. It results in a notable reduction in germination rate, germination percentage, shoot and root length, and yield by inhibiting the different enzyme activities and photosynthesis that ultimately cause the plant to die (Gupta et al. 1993, Hussain et al. 2009, Islam et al. 2012, Jamil et al 2006).

Although *Brassica* is recognized for its resilience to salinity (Maas and Hoffman 1977) and the threshold values for salinity are greater (9.0 dS/m), the rate of yield decline beyond these thresholds is notably higher compared to other plant species (Mass 1993). The genotypes of *Brassica* which are salinity sensitive, unable to tolerate the faster accumulation of sodium ions (Na^+), experience leaf death and the plant will die eventually (Munns 2002).

In the early growth stages of different *Arabidopsis* genotypes, it was revealed that excessive salt harmed vegetative growth and germination frequency (Quesada et al. 2002). At different salinity level (0 mM to 150 mM), when choosing canola cultivars that are salt tolerant, factors including the rate of photosynthesis, proline concentration and mobility of intracellular or intercellular ion can all be helpful (Ulfat et al. 2007, Kumar et al. 2009).

To deal with abiotic stresses, plants use different kinds of osmotic regulations. Plants accumulate osmolytes to protect the intra-cellular activities against adverse environmental fluctuations. Soluble sugars and proline play a crucial role in eliminating free radicals originating from cells and mitigating the stress's effects on physiological processes, by elevating the concentration of osmolyte within the cell (TİryakÍ et al. 2016).

Photosynthetic pigments and carotenoids play an important role in photosynthesis. In neutralizing singlet oxygen radicals, the amounts of chlorophyll and carotenoid indicate the plant's relative stress tolerance (Dehnavi et al. 2017). Ahmed et al. (2013)

reported that under saline stress plants show a significant decrease in the number of photosynthetic pigments as well as photosynthesis rate and reduce the stomatal density and transpiration rate. Salinity stress also limits root and shoot elongation as well as decreases the fresh and dry weights. Therefore, to mitigate environmental challenges such as extreme salinity, it is essential to identify, select, and cultivate salt-tolerant cultivars (Ashraf and McNeilly 2004, Almodares et al. 2007, Islam and Karim 2010).

As a major source of vegetable oil, rape seeds (*Brassica campestris* L. and *B. napus* L.) and mustards (*B. juncea* (L.) Czern. and Coss, and *B. carinata*) are cultivated commercially around the world (Ashraf and McNeilly 2004). Among these *Brassica* species, *Brassica campestris* is grown for commercial purposes as well as a prominent oilseed crop, and *Brassica carinata* accounts for about 60% of the country's total oilseed production and occupies approximately 80% of Bangladesh's total oilseed area (Miah et al. 2015). According to the data of Bangladesh Agricultural Research Council (BARC), certain varieties of *Brassica campestris* have some special qualities such as Agrani and Shafal are high yielding yellow seeded varieties, while BARI Sharisha-15 and Tori-7, respectively, have oil contents of approximately 48-52% and 38-41%, and BINA Sharisha-6 is a salt tolerant variety. But it was observed that the level of high salinity drastically affected the growth and productivity of *Brassica* species (Francois et al. 1994). Soils affected by salinity are prevalent worldwide and there is no continent which is free from this issue (Brady et al. 2002). Salinity intrusion is a big challenge in coastal regions, especially in developing regions with low-lying topography (Nicholls et al. 2007). In Bangladesh, the coastal area spans approximately 47,201 square kilometers along the Bay of Bengal and includes 19 coastal districts that either directly face the bay or near it (Islam et al. 2006).

So, it is necessary to select *Brassica* varieties that exhibit tolerance to salt, which enables them to cultivate in a salty environment or in salinized waters. For these purposes, in the present study, *Brassica carinata* and 5 varieties of *Brassica campestris* (Agrani, Shafal, BARI Sharisha-15, BINA Sharisha-6 and Tori-7) were selected. Keeping in mind the significance of developing salt tolerant varieties in Bangladesh, the effects of varied NaCl concentrations (50 mM,100 mM 150 mM, and 200 mM) were studied on these six varieties of *Brassica* to understand the physiology of stress responses, different physiological parameters such as proline content, production of hydrogen peroxide (H_2O_2) in the peroxisome, level of malondialdehyde (MDA), amount of photosynthesis pigments etc. were examined under different concentration of NaCl treatments.

Materials and Methods

Plant materials and treatments: In this present investigation, *Brassica carinata* and 5 varieties of *Brassica campestris* including Agrani, Shafal, BINA Sharisha-6, BARI Sharisha-15 and Tori-7 were used as plant materials. Seeds of these varieties were previously collected from Oilseed division of Bangladesh Agricultural Research Institute (BARI), Gazipur. Then the materials were maintained under the supervision of Plant Breeding and

Biotechnology Laboratory, Department of Botany, University of Dhaka. In this study, seedlings were grown on petri plates that contained distilled water as the control and salt (NaCl) solutions at various concentrations (50, 100, 150 and 200 mM).

Germination conditions: In this experiment, seeds of each variety of *Brassica* were surface-sterilized. Filter paper was used to hold fifty representative seeds of each variety in a petri plate with distilled water (control) and 50,100,150 and 200 mM NaCl. The petri plates were carefully maintained at 22 ± 1°C temperature, for eight hours day length, in a humid room. To avoid evaporation, the petri plates were carefully wrapped with parafilm.

Germination rate: Based on the formula outlined by Asfaw et al. (2011), the average number of days required for the radical or plumule to emerge was calculated.

Germination percentage: After 7 days, germinated seedlings were counted, and the germination percentage calculated according to formula (Ania-tul-haq et al. 2012) as:

Germination percentage = (Number of seeds germinated / Total number of sowing seeds) *100

Seedling Root/shoot ratio: The ratio of seedlings root and shoot length was calculated as described by Kagan et al. (2010).

Determination of the Malondialdehyde (MDA) content: Thiobarbituric acid (TBA), trichloroacetic acid (TCA) method was used to measure malondialdehyde (MDA) concentration, which acts as an indicator of lipid peroxidation (Dhindsa et al. 1981). The procedure involved homogenizing 500 mg of frozen leaf samples in10% trichloroacetic acid (TCA) with 0.5% thiobarbituric acid (TBA), heating the mixture for 30 minutes at 100°C, cooling it to room temperature and centrifuging it for 15 minutes at 12,000 rpm at 4 °C. After collecting the supernatant, its specific absorbance was measured at 535 nm. This value was then adjusted by deducting the non-specific absorbance, which was recorded at 600 nm. Using the extinction coefficient (155 mM^{-1} cm^{-1}), the MDA concentration was calculated and reported as μmol/g FW.

Determination of proline content: 500 mg dried leaves were treated with 3% (w/v) sulfosalicylic acid for the extraction of proline. 2 ml of supernatant was collected and then combined with 2 ml of fresh acid-ninhydrin and 2 ml of glacial acetic acid and heated for one hour at 100°C. The mixture was then extracted using 4 ml of toluene, the toluene containing upper phase was separated, kept at room temperature for cooling and a spectrophotometer (Shimadzu UV-1800, Japan) was used to detect the absorbance at 520nm. After that, the concentration of proline was determined using standard L-proline (Bates et al. 1973).

Determination of photosynthetic pigments: 0.1 g leaf tissue was homogenized with 1 ml of 80% acetone and then maintained at room temperature in the dark for 15 minutes to assess the photosynthetic pigments. Following that, the samples were centrifuged for five minutes at 4°C at 3000 rpm. Subsequently, 100 µl of supernatant was extracted and diluted with 80% acetone to make 1 ml. After centrifugation, the absorbance of the supernatant was measured at 663, 647, and 470 nm using a Shimadzu UV-1800 spectrophotometer. The values for carotenoids and chlorophyll were estimated using the data of Arnon et al. (1949). The units were computed using the formula (mg g-1FW).

Determination of H_2O_2 content: Levels of hydrogen peroxide were measured in accordance with Sergiev et al. (1997). 500 mg of leaf tissues were homogenized with 5 ml of 0.1% (w/v) TCA in an ice bath. After centrifuging the homogenate for 15 minutes at 12,000×g, 0.5 ml of the supernatant was mixed with 1 ml of 1 M KI and 0.5 ml of 10 mM potassium phosphate buffer (pH 7.0). The supernatant's absorbency was measured at 390 nm.

Determination of Na^+/ K^+ ions: The ion contents determinations were followed by Rahman et al. (2016). Samples of plants were oven dried for a while at 80 °C until their weight remained consistent. Nitric acid and perchloric acid (5:1) were used to crush and digest 100 mg of dried root and shoot separately for 48 hours at 70 °C. After that, an atomic absorption spectrophotometer (Shimadzu UV-1800, Japan) was used to determine the ion contents.

Data analysis: At least five separate series of experiments were used to represent the results and the results were denoted as means ± standard error (3 observations each). Software Excel 2019 was used to examine morpho-physiological and biochemical data.

Results and Discussion

Effects on germination parameters: The seed germination parameters comprising percentage of germination, speed of the germination, relative germination rate and germination index showed a gradual decrease under salinity stress conditions across all varieties of *Brassica* (Table 1). High salinity levels in soil and irrigation water reduce the germination of seeds in many *Brassica* species. However, although germination takes place in certain *Brassica* species, the plants' growth and development are retarded (Kumar et.al 1995).

In the present investigation, six varieties of *Brassica* responded variably in terms of tolerance to different levels of salt treatments (0, 50, 100, 150, and 200 mM NaCl). Germination percentages declined drastically less than 25% with 150 and 200 mM NaCl treatments, whereas in control seeds, it was 100% germination. In the presence of higher NaCl concentrations (more than 100 mM), significant differences were observed among the investigated varieties. Out of the six varieties assessed, all exhibited a germination percentage higher than 50%, except for Shafal (*B. campestris*) at 100 mM NaCl treatment. Notably, *Brassica carinata* and *BARI Sharisha*-15 had more than 50% germination even at 150 mM NaCl treatment. In contrast, the remaining cultivars exhibited less than 25% germination at 150 mM, and some recorded 0% germination at the 200 mM salinity stress. Similarly, the other parameters such as the speed, rate, and germination index

Table 1. Impacts of salinity on six *Brassica* varieties morpho-physiological traits and germination parameters at varying NaCl concentrations. (Each value is an average of 3 replicates).

Varieties	Salinity level (mM)	Germination (%)	Speed of germination	Relative germination rate	Germination index	Root/ Shoot ratio
Brassica carinata	00	100	5.67	1	8.57	0.48
	50	100	5.4	1	8.28	0.35
	100	64.29	4.25	0.64	4.53	0.343
	150	55.29	4.20	0.04	0.39	0.373
	200	2.86	3.88	0.03	0.25	0.093
Agrani	00	78.57	5.83	1	7.00	0.183
(*B.campestris*)	50	64.29	5.13	0.82	5.14	0.157
	100	57.14	5.06	0.73	4.71	0.143
	150	10	3.33	0.13	0.6	0.10
	200	5.71	3.23	0.07	0.39	0
Shafal (*B.campestris*)	00	47.14	4.66	1	3.64	0.167
	50	38.57	4.4	0.82	2.78	0.127
	100	12.86	2.59	0.27	0.67	0.07
	150	10	2.3	0.21	0.57	0.1
	200	2.86	2.29	0.06	0.14	0.03
BINA Sharisha-6	00	100	5.46	1	8.35	0.43
(*B. campestris*)	50	65.71	5.27	0.66	5.35	0.287
	100	51.43	4.9	0.51	3.6	0.243
	150	14.29	4.21	0.14	1.14	0.097
	200	0	0	0	0	0
BARI Sharisha-15	00	100	5.5	1	8.39	0.177
(*B. campestris*)	50	92.86	5	0.93	8.32	0.167
	100	58.57	5.03	0.59	4.64	0.143
	150	50.71	4.76	0.36	2.71	0.10
	200	8.57	3.5	0.09	0.53	0.09
Tori-7	00	62.86	6.15	1	5.71	0.467
(*B. campestris*)	50	61.43	6.1	0.98	5.32	0.316
	100	58.57	6.08	0.93	4.64	0.257
	150	24.29	5.49	0.39	2.07	0.143
	200	5.71	5.25	0.09	0.5	0.063

exhibited a gradual decline compared to the control condition. Salt stress affects various morpho-biochemical and physiological processes including rate of development, photosynthetic contents, leaf area index, floral abortion, and ion contents (N+, K+ and P) in many *Brassica* species (Steinbrenner et al. 2012). Therefore, these findings can be effectively used to distinguish between moderately and highly salt-tolerant varieties.

The impact of salt stress on growth:

Seedling length: Under the unaltered condition (0 mM NaCl), all varieties exhibited the maximum root/shoot length ratio in comparison to different concentrations of salinity stress (Table 1). At excessive salinity levels, all varieties showed an abrupt reduction in growth and development. Moreover, salt stress resulted in a notable reduction of plant biomass accumulation (root and shoot) across all varieties. However, the impact of salinity was more extensive in root growth than shoot dry matter, which led to a reduction in the root/shoot ratio (Garg et al. 2013). The length of the seedlings decreased gradually with increasing the concentration of salt level. About 25 to 35% of plant biomass reduction was observed under high salinity stress (Ahmar et al. 2019). The detrimental effects of salt stress on *B. juncea* also altered the plant's biomass, root and shoot length, and the CO_2 absorption process (Bybordi et al. 2010). The increasing level of salinity adversely reduced the root/shoot ratio in three out of six varieties at 150 mM NaCl, and the ratio reached 0, at 200 mM NaCl treatment. Among the six varieties, *Brassica carinata*, BARI Sharisha-15 and Tori-7 showed a higher seedling root/shoot ratio at 100 mM to 150 mM salinity level.

Proline content: Accumulation of proline content was observed in all six varieties of *Brassica*, as the salt stress levels escalated gradually from low to high (Fig. 1) The proline content fluctuation was not significantly different at 50 mM and 100 mM salinity, compared to control in all varieties, except Shafal (*B. campestris*), where maximum proline content of 23.65 $\mu molg^{-1}$ was measured at 100 mM of salt treatment. Compared to the controlled condition, this amount has nearly tripled. At high levels of salinity (150 and 200 mM), maximum proline content of 36.12 and 36.31 $\mu mol\ g^{-1}$ in Tori-7, and 38.60 and 39.55 $\mu mol\ g^{-1}$ in BARI Sharisha-15 was observed, which is around 4-fold and 2-fold higher compared to their respective control. Similar observations of high levels of proline content in *B. campestris* were observed under water stress conditions (Alam et al. 2014). Deepak et al. (1995) also observed the presence of high proline content in *Brassica* leaves under water stress. At 200 mM salinity stress, *Brassica carinata* and Shafal (*B. campestris*) showed a moderate increase of proline concentration compared to the control. On the other hand, the accumulation of proline was very low in Agrani (*B. campestris*) and BINA Sharisha-6 at high salinity stress compared to the other varieties. Proline is necessary for signal transduction, preserving the right osmotic potential, and having antioxidant qualities (Bates et al. 1973, Nahar et al. 2013). Proline and soluble sugars were shown to be the primary factors in salt stress resistance because they keep leaves from drying out and lessen turgor loss (Bornare et al. 2013).

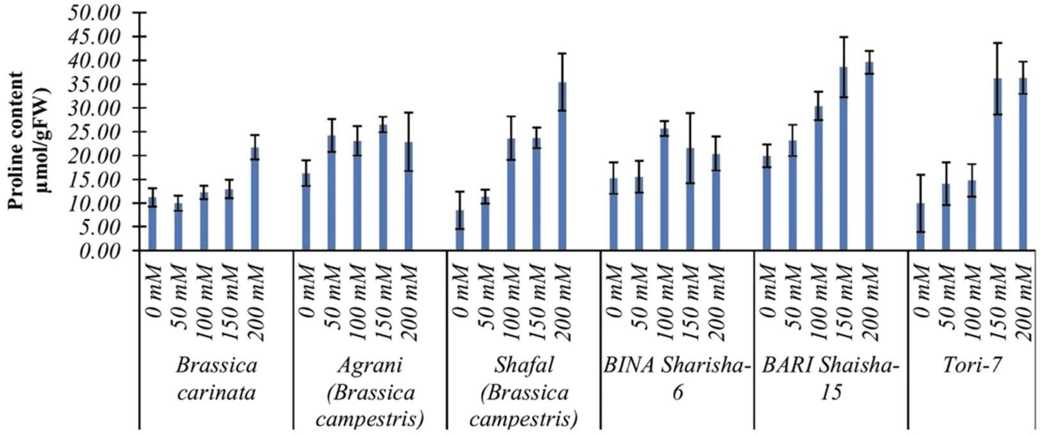

Fig. 1. Effects of salinity (0 mM, 50 mM, 100 mM, 150 mM and 200 mM NaCl) on proline content ($\mu molg^{-1}FW$), in six different varieties of *Brassica*. The average of three replicates is reflected in each value. Standard Error (SE) is represented as vertical bars.

Level of oxidative stress markers (H_2O_2 and MDA) under salt stress: Levels of MDA and H_2O_2 were used as indicators of oxidative stress to examine the consequences of salinity stress. The level of H_2O_2 exhibited a gradual increase, with rising the salinity levels. Agrani, Shafal, BINA Sharisha-6 and and BARI Sharisha-15 exhibited a progressive elevation in H_2O_2 content as the salinity level raised from 0 mM to 200 mM. Among them, Agrani (*B.campestris*) exhibited the highest H_2O_2 content compared to the control. Conversely, the other two varieties *B. carinata* and Tori-7, showed lower H_2O_2 accumulation under salt stress compared to the rest of the varieties (Fig. 2). Salt stress is complicated and imposed a broad spectrum of metabolic disorders which can lead to the accumulation of H_2O_2. H_2O_2 plays a critical role in various physiological processes of plants, encompassing photosynthesis, senescence, the progression of cells, stomatal activity, and biochemical responses over the course of a plant's growth (Deng et al. 2012). Increased levels of H_2O_2 have the potential to influence metabolic and antioxidant enzyme activities in a manner conducive to plant growth and development (Nilu and Liao 2016).

Lipid peroxidation is indicated by the MDA content, was consistently elevated as salinity levels increased across the cultivars. Agrani and BARI Sharisha-15 displayed the maximum accumulation of MDA content at 150 mM and 200 mM NaCl concentration, respectively, in comparison to the control. In contrast, Shafal and *B. carinata* showed the minimum level of MDA content at the 200 mM salinity level. The other two varieties represented the moderate accumulation of MDA content (Fig. 3). Previous studies showed that the primary cause of cell damage induced by salinity is the oxidative deterioration of cellular membranes, leading to lipid oxidation and the accumulation of malondialdehyde (MDA) (Ahmad et al. 2011, Das and Roychoudhury 2014). MDA is a widely used indicator of oxidative lipid injury induced by environmental stress.

According to the data of Jbir-Koubaa et al. (2015), shock and photo-oxidative stress are two possible effects of salinity which causes MDA to accumulate in leaves. These findings collectively highlight that H_2O_2 and lipid peroxidation are common occurrences in plants under stress, and they can serve as valuable indicators of the physiological status during plant growth.

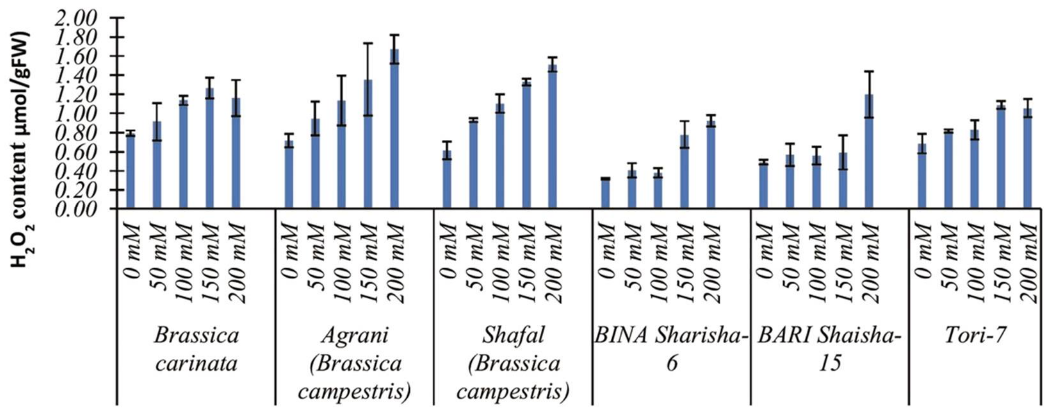

Fig. 2. Effects of salinity (0 mM, 50 mM, 100 mM, 150 mM and 200 mM NaCl) on H_2O_2 content (µmolg^{-1} FW), in six different varieties of *Brassica*. The average of three replicates is reflected in each value. Standard Error (SE) is represented as vertical bars.

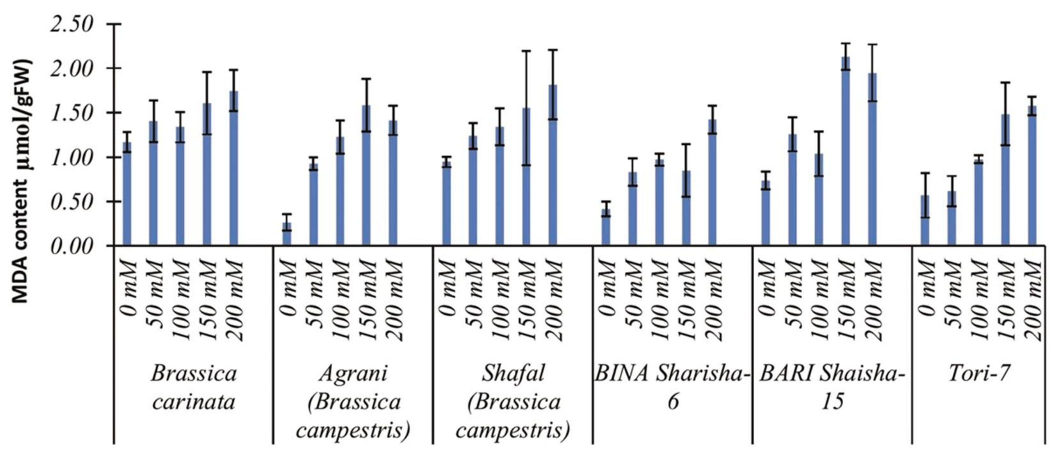

Fig. 3. Effects of salinity (0 mM, 50 mM, 100 mM, 150 mM and 200 mM NaCl) on MDA level (µmolg-1 FW), in six different varieties of *Brassica*. The average of three replicates is reflected in each value. Standard Error (SE) is represented as vertical bars.

Photosynthetic pigments in leaves: The impact of salt stress on chlorophyll contents and carotenoids was also observed in this investigation. Chlorophylls and other photosynthetic pigments, including carotenoids were significantly impacted by salinity stress. With the increase of the salt level, both total chlorophyll and carotenoids contents exhibited a gradual decrease. In this study, a consistent reduction in total chlorophyll contents and carotenoids was observed across all six varieties, with the continuous increase of salt levels from 0 mM to 200 mM (Fig. 4). Among the six varieties, Agrani showed the highest chlorophyll contents 0.39 µg/ml/gFW, and BINA Sharisha-6 exhibited the maximum carotenoids 0.16 µg/ml/gFW under 200 mM salinity stress, in comparison to the rest of the varieties. Conversely, variety Shafal recorded the minimum chlorophyll contents 0.20 ug/ml/gFW and the lowest carotenoids 0.02 µg/ml/gFW at the 200 mM salinity stress. Previous studies have indicated a connection between the degradation of essential photosynthetic pigments and the decrease in photosynthetic rate (Chaves et al. 2003, Reynolds et al. 2005). Under drought conditions, a low amount of chlorophyll a, b, and total was observed by Alam et al. (2014).

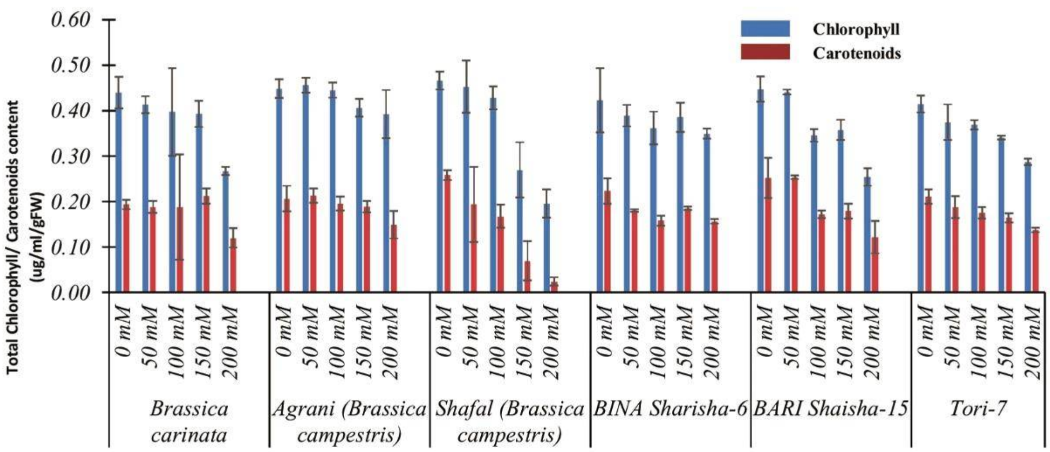

Fig. 4. Effects of salinity (0 mM, 50 mM, 100 mM, 150 mM and 200 mM NaCl) on Photosynthetic pigments (Chlorophyll a and b and carotenoid) (µg/ml/gFW), in six different varieties of *Brassica*. The average of three replicates is reflected in each value. Standard Error (SE) is represented as vertical bars.

Ion accumulation in roots: Ion accumulation is another phenomenon that occurs during different types of salt stress. All the varieties of *Brassica* displayed a unique response pattern in case of Na^+ and K^+ ion accumulation under different salt stresses. The Na^+ accumulation in roots showed a significant increase, with rising the level of salinity, whereas it inhibited the accumulation of K^+ ion in the roots. The ratio of Na^+ and K^+ ion accumulation exhibited a gradual increase across all the varieties. Among them, Agrani (*B. campestris*) showed the highest Na^+/ K^+ ion accumulation and *Brassica carinata* exhibited the lowest amount at 200 mM salinity level (Fig. 5). The Na^+/ K^+ ratio serves as a valuable indicator for evaluating plant tolerance to salt stress. Comparing salt-tolerant genotypes to salt-sensitive genotypes, the former showed an 8-fold higher K^+/Na^+ ratio and a strong attraction for K^+ over Na^+ (Ashraf et al. 2007).

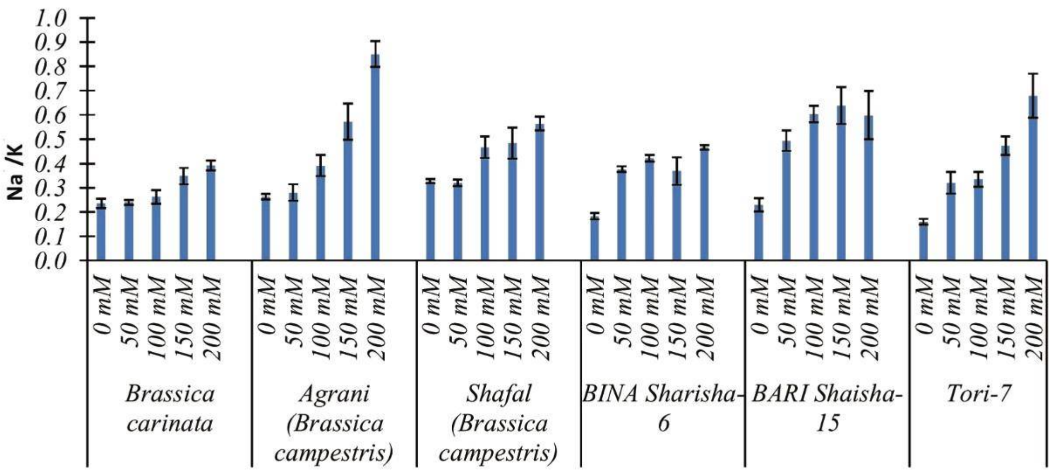

Fig. 5. Effects of salinity (0 mM, 50 mM, 100 mM, 150 mM and 200 mM NaCl) on Na^+ / K^+ ion accumulation, in six different varieties of *Brassica*. The average of three replicates is reflected in each value. Standard Error (SE) is represented as vertical bars.

The issue of salinity is one of the most significant global concerns. The normal growth of *Brassica* species is interrupted by saline, resulting in a significant decrease in their overall oilseed production. Therefore, a comparative study was performed among six varieties of *Brassica* species, which revealed their tolerance and sensitivity towards salinity. The findings of the study showed that Agrani and BARI Sharisha-15 were the most tolerant to high and moderate levels of salinity. As a result, these two varieties of *Brassica* species can be cultivated in a saline environment, which will lead to achieving the desired economic goals. Moreover, these methods can be applied to other important crops to find out their agronomic performances under salinity stress.

References

Ahmad P and **Umar S** (2011) Oxidative stress: role of antioxidants in plants. Studium Press, New Delhi during abiotic stress in plants. Bot. Res. Intern. **2**: 11-20.

Ahmar, Sunny, Liaqat N, Hussain M, Salim MA, Shabbir MA, Ali MY, Noushahi HA, Bilal M, Atta B and **Rizwan M** (2019) Effect of abiotic stresses on *Brassica* species and role of transgenic breeding for adaptation. Asian J. Res. Crop Sci. ***3***: 1-10.

Ahmed, Mosaddek I, Cao F, Zhang M, Chen X, Zhang G and **Wu F** (2013) Difference in yield and physiological features in response to drought and salinity combined stress during anthesis in Tibetan wild and cultivated barleys. Plos one **8**: e77869.

Alam MM, Nahar K, Hasanuzzaman M and **Fujita M** (2014) Trehalose-induced drought stress tolerance: A comparative study among different *Brassica* species. Plant Omics J. **7**: 271-283.

Alam SS, Khaleda L and **Al-Forkan M** (2013) An Efficient *in vitro* Regeneration System for Tori-7 (*Brassica campestris*). Glob. J. Science Frontier Res. Bio-Tech. Genet. **13**: 2249-4626.

Almodares A, Hadi MR and **Dosti B** (2007) Effects of salt stress on germination percentage and seedling growth in sweet sorghum cultivars. J. Biol. Sci. **7**: 1492-1495.

Ania-tul-haq, Vamil R and **Agnihotri RK** (2012) Effect of Osmotic Stress (PEG) on Germination and Seedling Survival of Lentil (*Lens culinaris*). M. Res. Agri. Sci. **1**: 201-202.

Arnon, Daniel I (1949) Copper enzymes in isolated chloroplasts, polyphenol oxidase in *Beta vulgaris*. Plant physiol. **24**:1.

Asfaw KG (2011) The Response of Some Haricot Bean (*Phaseolus vulgaris*) Varieties for Salt Stress during Germination and Seedling Stage. Curr. Res. J. Biol Sci. **3**: 282-288.

Ashraf M and **McNeilly T** (2004) Salinity tolerance in *Brassica* Oilseeds. CRC Crit Rev Plant Sci. **2**: 157- 174.

Ashraf M, Rahmatullah, Kanwal S, Tahir MA, Sarwar A and **Ali L** (2007) Differential salt tolerance of sugarcane genotypes. Pakistan J. Agr. Sci. **4**:85-89.

Bates LS, Waldren RP and **Teare ID** (1973) Rapid determination of free proline for water-stress studies. Plant Soil. **39**: 205-207.

Bornare SS, Prasad LC and **Kumar S** (2013) Comparative study of biochemical indicators of salinity tolerance of barley (*Hordeum vulgare* L.) with other crops: a review. Canadia J Plant Breed. **1**: 97-102.

Brady NC and **Weil RR** (2002) The Nature and Properties of Soils, 13th Edition, Agroforest. Syst. **54**: 249.

Bybordi A, Tabatabaei SJ and **Ahmadev A** (2010) Effects of salinity on fatty acid composition of canola (*Brassica napus* L.). J. Food, Agricul. Environ. **8**: 113-115.

Chaves MM, Maroco JP and **Pereira JS** (2003) Understanding plant responses to drought—from genes to the whole plant. Funct. Plant Biol. **30**: 239-264.

Dehnavi MM, Zarei T, Khajeeyan R and **Merajipoor M** (2017) Drought and salinity impacts on bread wheat in a hydroponic culture: a physiological comparison. J. Plant Physiol. Breed. **7**: 61–74.

Das K and **Roychoudhury A** (2014) Reactive Oxygen Species (ROS) and response of antioxidants as ROS-scavengers during environmental stress in plants. Front Environ. Sci. **2**: 1-13.

Deepak M, Wattal PN and **Mathue D** (1995) Influence of water stress on seed yield of Canadian mustard at flowering and role of metabolic factors. Plant Physiol. Bioch. **22**: 115-118.

Deng X, Cheng Y, Wu X, Kwak S, Chen W and **Egrinya A** (2012) Exogenous hydrogen peroxide positively influences root growth and metabolism in leaves of sweet potato seedlings. Aus. J. Crop Sci. **6**: 1572–1578.

Dhindsa RS, Plumb-Dhindsa P and **Thorpe TA** (1981) Leaf senescence: correlation with increased levels of membrane permeability and lipid peroxidation, and decreased levels of superoxide dismutase and catalase. J. Exp. Bot. **32**: 96-101.

FAO (Food and Agriculture Organization), (2013). FAO Production Yearbook. Food and Agriculture Organization of the United Nations, Rome 00100, Italy. **56**:118.

Francois LE, Grieve CM, Maas EV and **Lesch SM** (1994) Time of salt stress affects growth and yield components of irrigated wheat. Agron. J. **86**: 100-7.

Garg N and **Baher N** (2013) Role of arbuscular mycorrhizal symbiosis in proline biosynthesis and metabolism of *Cicer arietinum* L. (Chickpea) genotypes under salt stress. J. Plant Growth Regul. **32**: 767-778.

Goswami B, Hoque MI, Khan S and **Sarker RH** (2020) *In vitro* regeneration of three varieties of *Brassica campestris* L. grown in Bangladesh. Bangladesh J. Sci. Indust. Res. **55**: 181-188.

Gupta R and **Minhas PS** (1993) Managing salt affected waters for crop production. In: Singh S.D. (Ed.), Arid Land Irrigation and Ecological Management. Scientific Publishers, New Delhi: 159-198.

Hussain K, Majeed A, Nawaz K, Bhatti KH and **Nisar FK** (2009) Effect of different levels of salinity on growth and ion contents of black seeds (*Nigella sativa* L.). Curr. Res. J. Biol. Sci. **1**: 135-138.

Islam MM and **Karim MA** (2010) Evaluation of rice genotypes at germination and early seedling stage for their tolerance to salinity. Agriculturists. **8:** 57-65.

Islam MR (2006) Managing diverse land uses in coastal Bangladesh: institutional approaches. In Environment and livelihoods in tropical coastal zones: Managing agriculture-fishery-aquaculture conflicts, Wallingford UK: CABI, 237-248.

Islam MT, Jahan NA, Sen AK and **Pramanik MHR** (2012) Effects of salinity on morpho-physiological attributes and yield of Lentil genotypes. Int. J. Sustain. Crop Prod. **7**:12-18.

Jamil M, Lee DB, Jung KY, Ashraf M, Lee SC and **Rha ES** (2006) Effect of salt (NaCl) stress on germination and early seedling growth of four vegetables species. J. Cen. Europ. Agri. **7**: 273-282.

Jbir-Koubaa R, Charfeddine S, Ellouz W, Saidi MN, Drira N, Gargouri-Bouzid R and **Nouri-Ellouz O** (2015) Investigation of the response to salinity and to oxidative stress of interspecific potato somatic hybrids grown in a greenhouse. Plant Cell Tiss. Org. Cul. **120**: 933-947.

Kagan K, Karakoy T, Bakoglu A and **Akçura M** (2010) Determination of salinity tolerance of some lentil (*Lens culinaris* M.) varieties. J. Food. Agri. Envron. **8**: 140-14.

Kumar D (1995) Salt tolerance in oilseed brassicas-present status and future prospects. Plant Breed. **65**: 1439- 1477.

Kumar G, Purty RS, Sharma MP, Singla-Pareek SL and **Pareek A** (2009) Physiological responses among *Brassica* species under salinity stress show strong correlation with transcript abundance for SOS pathway-related genes. J. Plant Physiol. **166**: 507-520.

Miah MM, Afroz S, Rashid MA and **Shiblee SAM** (2015) Factors affecting the adoption of improved varieties of mustard cultivation in some selected sites of Bangladesh. Bangladesh j. agric. res. **40**: 363-379.

Maas EV and **Hoffman GJ** (1977) Crop salt tolerance: Current assessment. J. Irrigat. Drain. Div. **103**: 115-134.

Mashfiqur R, Mustafa KS, Tazreen SM, Shahriar K, Abu RM, Fadrus AN and **Harunor R** (2022). Performance of mustard varieties under saline prone areas of Bangladesh. African J. Agricult. Res. **18**: 608-616.

Mass EV (1993) Salinity and citriculture. Tree Physiol. **12**: 195- 216.

Munns R (2002) Comparative physiology of salt and water stress. Plant Cell Environ. **25**: 239-250.

Nahar K, Hasanuzzaman M, Alam MM and **Fujita M** (2013) Exogenous glutathione induced drought stress tolerance in *Vigna radiata* seedlings through enhanced antioxidant defense and methylglyoxal system. Interdrought IV Conference September.

Nicholls RJ, Wong PP, Burkett V, Codignotto J, Hay J, McLean R, Ragoonaden S, Woodroffe CD, Abuodha PAO, Arblaster J and **Brown B** (2007) Coastal systems and low-lying areas.

Niu L and **Liao W** (2016) Hydrogen peroxide signaling in plant development and abiotic responses: crosstalk with nitric oxide and calcium. Front Plant Sci. **7**: 230.

Quesada V, Garcia-Martinez S, Piqueras P, Ponce MR and **Micol JL** (2002) Genetic architecture of NaCl tolerance in *Arabidopsis*. Plant Physiol. **130**: 951-963.

Rahman A, Hossain MS, Mahmud JA, Nahar K, Hasanuzzaman M and **Fujita M** (2016) Manganese-induced salt stress tolerance in rice seedlings: regulation of ion homeostasis, antioxidant defense and glyoxalase systems. Physiol Mol Biol Plants**. 22**: 291-306.

Rahim QMAR and **Mohammed IQ** (2020) Effect of organic fertilizer and foliar application of humid and salicylic acids on reducing salinity stress of irrigation water on vegetative growth characteristics of broccoli (*Brassica oleracea*) Plant. Plant Arch. **20**: 2263-2268.

Reynolds MP, Mujeeb-Kazi A and **Sawkins M** (2005) Prospects for utilising plant-adaptive mechanisms to improve wheat and other crops in drought-and salinity-prone environments. Ann. appl. Biol. **146**: 239-259.

Sergiev I, Alexieva V and **Karanov E** (1997) Effect of spermine, atrazine and combination between them on some endogenous protective systems and stress markers in plants. Compt. Rend. Acad. Bulg. Sci. **51:** 121-124.

Steinbrenner AD, Agerbirk N, Orians CM and **Chew FS** (2012) Transient abiotic stresses lead to latent defense and reproductive responses over the *Brassica rapa* life cycle. Chemoecology. **22**: 239-250.

Tİryakİ İ (2016) Drought stress and tolerance mechanisms in alfalfa (*Medicago sativa* L.). Kahramanmaraş Sütçü İmam Üniversitesi Doğa Bilimleri Dergisi. **19**: 296-304.

Ulfat M, Athar HR, Ashraf M, Akram NA and **Jamil A** (2007) Appraisal of physiological and biochemical selection criteria for evaluation of salt tolerance in canola (*Brassica napus* L.). Pak. J. Bot. **39**: 1593-1608.

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