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The Effect of Algal Biochar in Improving Wheat *Triticum aestivum* L Performance under Salinity Stress

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Abstract

Salinity is one of the major abiotic factors in global food security. However, algal biochar (ABC) could be particularly well suited to improve plant growth under salinity stress conditions. Therefore, the present study was conducted to evaluate the effect of Algal Biochar (ABC) on the growth and yield of wheat *Triticum aestivum* L grown under salinity stress. Pots experiment with and without combinations of algal biochar (ABC) under saline and non-saline conditions (0 and 120mM NaCl) was performed. Wheat grains were sowed in plastic pots filled with four kilograms of sandy soil and mixed with 3% of algal biochar. The experiment was designed in a completely randomized design (CRD) with four replications. The study found that 3% of ABC mitigated the negative effects of salinity stress and improved wheat performance. The result indicated that the application of ABC significantly ($P< 0.05$) increased plant height by 10%, tiller number plant^{-1} by 40%, spike number plant^{-1} by 42%, spikelet number spike^{-1} by 11%, grain number plant^{-1} by 25%, plant dry weight by 8%, 1000 grain weight by 35%, harvest index by 56%, and grain yield plant^{-1} by 69%. The study suggests that the application of ABC could be an effective strategy to improve the growth and yield of wheat crops under salinity stress conditions.

Keywords: Wheat *Triticum Aestivum* L; Salinity Stress; Algal Biochar; Growth; Yield

INTRODUCTION

Wheat *Triticum aestivum* L occupies 30% of world cereal production (770 million tons) from 220 million hectares (FAOSTAT, 2021). The world population is expected to reach nine billion people in 2050; therefore, there is an urgent need to increase the production of wheat crops to feed the population (UN, 2022). However, due to climate change, global wheat production faces several challenges, including some biotic and abiotic stresses (Liu et al., 2019; Zahra et al., 2023). Heat, drought, and salinity are considered among the most important environmental stresses facing wheat production worldwide (Ehtaiwesh, 2016; Fatima et al., 2020). Globally, salinity stress in wheat is a growing concern, and its impact on crop production has been predicted to increase because of current climate changes. Salinity is a major abiotic stress that adversely affects the growth and yield of wheat crops (Ehtaiwesh, 2019; Sadak and Dawood, 2023). However, there are many efforts to develop some strategies to mitigate the negative effects of salinity on the growth and productivity of wheat crops. Examples of these strategies include the application of some compounds such as plant hormones, humic acid, jasmonic acid, acetylsalicylic acid, and dry yeast solution which have been shown to minimize the toxic effect of salt stress (Atia et al., 2018; Ayub et al., 2020; Saidimoradi et al., 2019; Aliet al., 2022; Ehtaiwesh, 2022a; Ehtaiwesh, 2023). In addition, another strategy to mitigate salinity is by growing salt-tolerant genotypes (Mohanavelu et al., 2021; Hossain et al., 2021; Ehtaiwesh, 2022b, Ehtaiwesh et al., 2024). Moreover, biochar addition is suggested as an effective strategy to improve crop performance under saline stress (Parkash and Singh, 2020; Kul et al., 2021).

Biochar is a carbon-rich charcoal reproduced by the process of pyrolysis, which involves heating biomass in an oxygen-limited atmosphere (Weber and Quicker, 2018). The biochar is low in surface area and cation exchange capacity, and high in ash and nutrients (Wang and Wang, 2019). Many studies indicated that biochar is used as a catalyst, soil amendment, fuel cell, heavy metal ions and contaminant adsorbent, gas storage and separation, wastewater treatment, and plant growth enhancer (Xiang et al., 2020; Zhang et al., 2020; Joseph et al., 2021; Lehmann et al., 2021; Liu et al., 2022; Qian et al., 2023). Currently, there are interests in using biochar produced from algae in agricultural fields, such as improving soil properties and using algae as an organic and biofertilizer (Baweja et al., 2019; Ammar et al., 2022). Algae are considered one of the biological species that are efficient in producing biomass and phytochemicals, due to their high efficiency in photosynthesis, in addition to their high growth rate compared to terrestrial plants (Abideen et al., 2022). Some studies mentioned that the application of algal bi-char is a sustainable solution that has the potential to improve agricultural productivity by increasing crop production (Santos and Pires, 2018; Shanmugam et al., 2018). Algal biochar is considered a soil ameliorant that may significantly improve the retention of nutrients in the soil and could rebuild organic matter in the soil (Mona et al., 2021). Many studies concluded that algal biochar has a very high exchangeable nutrient content (N, P, K, Ca, Mg, and Mo) and may add essential nutrients to the soils (Joseph et al., 2021). In addition, biochar has the potential to decrease soil acidity, increase cationic exchange capacity, and optimum nutrient availability (Pshenovschi et al., 2022). Recently, some studies indicated that the use of algal biochar has contributed to improving bio characterization, growth, and productivity of many plants such as soybean (Zhang et al., 2020), stevia plant (Abd el Aal et al., 2020), maize (Ullah et al., 2020), and tomato (Kul et al., 2021). In addition, some studies have indicated the ability of biochar to mitigate some of the negative effects of some abiotic stresses such as drought (Zhang et al., 2020), salinity (Kanwal et al., 2018), high temperature (Fahad et al., 2015) and improving overall agricultural sustainability (Abideen et al., 2022). In Libya, the calcareous sandy soils suffer from poor productivity due to their low organic matter content; their low water retention, and their deficiency of nutrients elements including nitrogen, phosphorus, potassium, and other micronutrients besides to salinity of the soils and irrigation water (Nwer et al., 2021). For these reasons, and in order to increase the productivity of these soils, it is preferable to use biochar as an organic amendment element. Therefore, the aim of this work was to study and evaluate the effect of algal biochar on salinity stress mitigation in wheat (*Triticum aestivum* L.).

MATERIALS AND METHODS

The experiment was conducted during the wheat growing season of fall/ winter of 2020 in Jodaam farm, to investigate the effects of algal biochar (ABC) on the performance of wheat crops under salinity stress conditions. Bread wheat (Salambo) seeds were obtained from National Libyan Gen-Bank (NLGB) in Tripoli.

Algae collection

Cystoseira barbata moss was collected from the beach of the Al-Muttarid area in August 2020. At the coordinates (12◦ 48.889 East, 32◦ 47.965 North) approximately 10 meters away from the beach and from a depth of 1-1.5 meters, the algae were washed well with seawater to get rid of impurities and plankton. Algae samples were washed with fresh water three times to get rid of salinity, and then the samples were dried in the shade for 4 days.

Algal biochar preparation

Biochar was prepared from dried algae material and then via pyrolysis at a temperature of 400°C for 1hr (Ullah et al.,2020). Then, the biochar was grounded and the resulting material was passed through a sieve (2 mm). The algal biochar was subjected to some analyses to estimate some chemical and physical properties such as acidity (pH), electrical conductivity (EC), ash percentage, and determination of some elements such as nitrogen, phosphorus, and potassium (NPK).

Estimating the electrical conductivity of biochar and its pH was done (Bird et al., 2011) by preparing a solution with a ratio of 10:1 (sample: water) and placing it in the autoclave for 35 minutes at a temperature of 121°C and at atmospheric pressure (1atm). Then the solution is filtered and the EC and pH are measured using an EC and pH meter respectively.

The Ash of the biochar was also estimated by taking 5 g of biochar sample in a crucible and placed in a combustion furnace at a temperature of 800°C for 4 hours, then cooled in a glass container. The ash was estimated from the following equation:

$$Ash\ percentage = \frac{weight\ of\ ash\ sample}{weight\ of\ dry\ ash} * 100$$

The total NPK in the biochar was estimated following the method mentioned in (George et al., 2013) using a DR 3900™ Spectrophotometer at a wavelength of 410 nm. The algal biochar properties are shown in Table 1. Mixing of biochar in soil was done in a specific concentration (3% of soil) before sowing of seeds.

Table (1). Physicochemical traits of algal biochar

Parameters	Algal biochar
PH	10
EC(ds m^1)	1.9670
Ash content (%)	45
Total nitrogen (%)	1.790
Total phosphorous (%)	5.340
Total potassium (%)	1.920

Experimental details

The experiment was designed in a completely randomized design (CRD) with four replicates for each treatment. Before sowing, the soil was mixed with biochar and sieved with a 2 mm mesh. Then, plastic pots with a capacity of 5kg were filled with 3% biochar-treated and untreated soils. Untreated soil pots served as control. Ten seeds of wheat (*Triticum aestivum* L. cv. Salambo) were sown in each pot, and seedlings were allowed to germinate for 2 weeks, then seedlings were thinned to three seedlings per pot. At the booting stage, the pots in each treatment, biochar-treated soil and untreated were divided into two groups, and each group represented a salinity treatment (0mM and 120mM). Salinity stress was applied by irrigating wheat plants with tap water with electrical conductivity (EC) of 1.7ds m^{-1}, and 120mM NaCl solution with electrical conductivity (EC) of 8.5 ds m^{-1} throughout the treatment period, which lasted for 3 weeks. Irrigation was applied manually two times a week. After salinity treatment, all plants were irrigated with tap water as needed and well managed until wheat plants reached physiological maturity. Grain maturity was visually estimated according to the complete loss of green color from grains.

Data collection

At harvesting, four plants were collected from each treatment one plant from each replicate. Wheat plants were hand-harvested by cutting them at the soil level. Data on shoot length (cm), number of tillers plant^{-1} both fertile (with spikes) and non-fertile (without spike), and number of spikes plant^{-1} were recorded, Then plants were oven dried at 60 °C for 73 h to record plant dry weight (g). Then main spikes were hand threshed to separate grains, grain number spike^{-1} was counted manually and grain yield spike^{-1}, grain yield plant^{-1} (g), 1000-grain weight (g), and harvest index were calculated.

Statistical analysis

The experiment was conducted in a completely randomized design with four replications. Biochar was the main plot factor (two levels with (+ABC) and without Biochar (-ABC)), and salinity was assigned to subplots (two levels 0mM and 120mM NaCl). Data were analyzed using the GLM procedure in statistical SPSS software for mean and standard error estimation. Separation of means was carried out using the least significant differences (LSD; $P < 0.05$).

RESULTS

The values in Table 2 represent the probability values of the effect of algal biochar (ABC) treatment, salinity stress(S), and the interaction between Algal biochar and salinity on some growth and yield traits of wheat plants. The effect of algal biochar had a very significant ($P<0.01$) effect on all growth and yield traits of wheat plants. In addition, the effect of salinity had a highly significant ($P<0.001$) effect on the growth and yield traits of wheat plants. In the same way, there was a significant ($P<0.05$) effect of the interaction of algal biochar treatment and salinity stress on the growth and yield traits of wheat plants table2.

Table (2). Probability values of the effects of biochar (ABC), salinity (S), and ABC x S interaction on various growth and yield traits of wheat.

Traits	Algal biochar (ABC)	Salinity (S)	ABC x S
Plant height (cm)	.002	<.001	0.043
Tiller number plant^{-1}	.006	<.001	0.033
Spike number plant^{-1}	.007	<.001	0.049
Spikelet number spike^{-1}	.003	<.001	0.023
Grain number plant^{-1}	<.001	<.001	0.041
Grain yield plant^{-1} (g)	<.001	<.001	0.034
1000 grain weight (g)	0.002	<.001	0.007
Dry weight plant^{-1} (g)	<.001	<.001	0.045
Harvest index (%)	<.001	<.001	0.008

Table 3 showed the main effect of algal biochar treatment on the growth and yield of wheat plants, which indicated that the addition of algal biochar to the soil had increased wheat growth and yield traits. All growth and yield traits included in this study were increased with the addition of algal biochar, and this percentage increase ranged from 4% to 18% table3.

Table (3). The main effect of algal biochar treatments on various growth and yield traits of wheat plants. Means were estimated using the GLM procedure in SPSS.

Traits	- ABC	+ ABC
Plant height (cm)	61.5	64.4(+5%)
Tiller number plant^{-1}	5.6	6.5(+16%)
Spike number plant^{-1}	4.8	5.5(+15%)
Spikelet number spike^{-1}	16.8	17.6(+5%)
Grain number plant^{-1}	209	228(+9%)
Grain yield plant^{-1} (g)	7.7	8.5(+18%)
1000 grain weight (g)	31.3	36(+15%)
Dry weight plant^{-1} (g)	21.1	22.1(+5%)
Harvest index (%)	0.31	0.37(+17%)

Values in parenthesis indicate the percent increase from the control treatment (without ABC) to the ABC treatment (with ABC).

Table 4 shows the main effect of salinity stress on the growth and yield traits of wheat. Salinity stress had a negative effect on all growth and yield traits of wheat plants. This negative effect of salinity resulted in a decrease in all the traits studied in this experiment, and this is evident from the percentage decrease in all the traits, which ranged from -24% to -70% percent reduction over the control table3.

Table (4). The main effect of salinity on various growth and yield traits of wheat plants. Means were estimated using the GLM procedure in SPSS.

Traits	0mM NaCl	120mM NaCl
Plant height (cm)	77.3	48.7(-37%)
Tiller number plant^{-1}	7.6	4.5(-41%)
Spike number plant^{-1}	6.6	3.6(-45%)
Spikelet number spike^{-1}	19.6	14.8(-24%)
Grain number plant^{-1}	314	123(-61%)
Grain yield plant^{-1} (g)	12	3.6(-70%)
1000 grain weight (g)	38.4	29(-25%)
Dry weight plant^{-1} (g)	25.5	17.7(-31%)
Harvest index (%)	0.472	0.203(-57%)

Values in parenthesis indicate the percent differences from the control treatment (0mM NaCl) to the (120mM NaCl).

The changes in the growth and yield traits of the wheat plants under the interaction of different salinity and algal bio char applications are shown in Figures 1-3. Salt treatment significantly decreased all investigated growth and yield traits. Biochar application had a positive effect on all observed traits at both salinity levels (0mM and 120mM NaCl) compared to the respective untreated ones.

Salinity stress had a significantly negative effect on wheat growth traits such as plant height. However, at both salinity levels (0mM and 120mM NaCl), algal biochar treatment had significant ($P< 0.05$) positive effect on wheat plants, which was indicated by increasing plant height in wheat plants treated with algal biochar as compared to wheat plants without algal biochar treatment (Fig.

1a). The same trend was found with other growth trait such as tiller number, the result indicated that salinity stress had significantly negative effect on tiller number (plant^{-1}). Nevertheless, when algal biochar was added to the growth media of wheat plants at both salinity levels (0mM and 120mM NaCl), the tiller number (plant^{-1}) was increased as compared to the untreated plant (Fig1 b). Similarly, salinity stress significantly reduced above ground plant dry weight trait. Though, at both salinity levels (0mM and 120mM NaCl), algal biochar treatment had significantly increased plant dry weight as compared to untreated plants (Fig1 c).

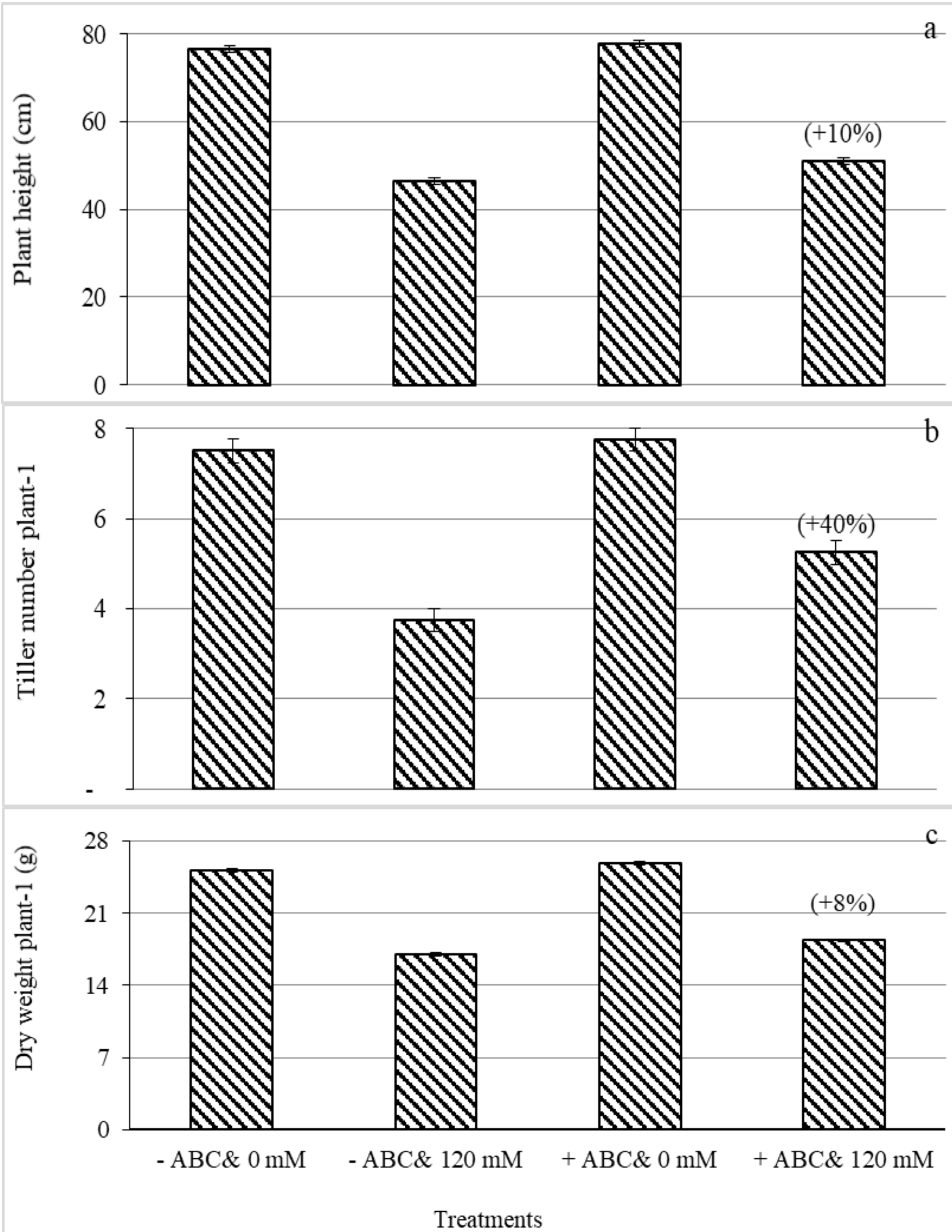

Figure (1): Effect of algal biochar and salinity treatments on (a) plant height (cm), (b) tiller number (plant $^{-1}$), and (c) total dry weight (g plant^{-1}) of wheat. Each datum indicates the mean value and vertical lines on top of bars indicate standard error of means (n = 4). Values in parenthesis indicate the percent increase from without the addition of ABC treatment to with the addition of ABC treatment under salinity stress (120mM NaCl)

The effects of algal biochar and salinity on spike number (plant^{-1}) (cm), spikelet number (spike $^{-1}$), and grain number (plant $^{-1}$) of wheat plants are shown in Fig. 2. It was found that salinity significantly (P< 0.05) decreased spike number (plant^{-1}), however when analyzed across the two salinity levels there was a tendency of increasing in spike number (plant $^{-1}$), with algal bio char addition as compared to the non-biochar treatment (Fig 2a). In addition, salinity stress significantly decreased (P< 0.05) and decreased spikelet number (spike $^{-1}$) for all wheat plants. However, this decrease was moderated by the addition of algal biochar, which resulted in a significant interactive effect of salinity and algal biochar on spikelet number (Fig. 2b). Moreover, salinity had significantly (P< 0.05) decreased grain number (plant^{-1}) of wheat plants. However, adding algal biochar had a positive effect on the number of grins (plant^{-1}), which resulted in an increase in the number of grains (plant^{-1}) compared to plants that were not treated with alga biochar (Fig. 2c).

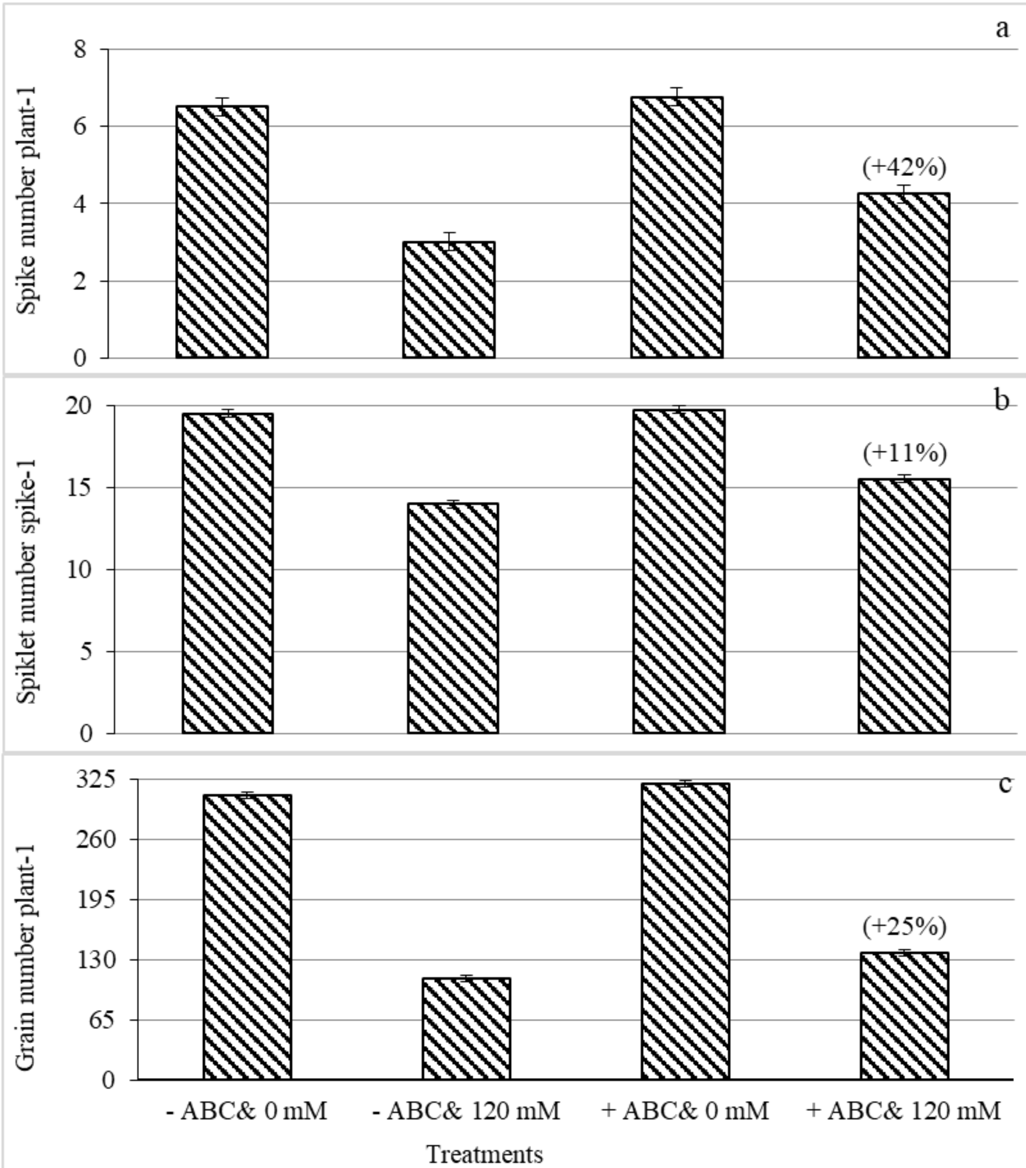

Figure (2): Effect of algal biochar and salinity treatments on (a) spike number (plant $^{-1}$), (b) spikelet number (spike $^{-1}$), and (c) grain number (plant $^{-1}$) of wheat. Each datum indicates the mean value and vertical lines on top of bars indicate standard error of means (n = 4). Values in parenthesis indicate the percent increase from without the addition of ABC treatment to with the addition of ABC treatment under salinity stress (120mM NaCl)

Both algal biochar and salinity treatments significantly ($P< 0.05$) influenced grain yield (g/plant[1]). The result showed that salinity stress decreased grain yield ($plant^{-1}$) while algal biochar addition significantly increased grain yield in both salinity levels as compared to respective non-algal bio-char treatment (Fig. 3a). Fig. 3b revealed that 1000 grain weight was significantly affected by salinity stress as well as algal biochar application. Under 120mM Nacl salinity level 1000 grain weight was decreased. However biochar addition alleviated the negative effect of salinity and resulted in an increased 1000 grain weight as compared to untreated plants (Fig. 3b). On one hand, the results showed that salinity stress negatively affected harvest index and resulted in decreased harvest index trait. On the other hand, the biochar addition significantly influenced harvest index traits under saline and non-saline conditions compared to respective non-biochar treatment (Fig. 3c).

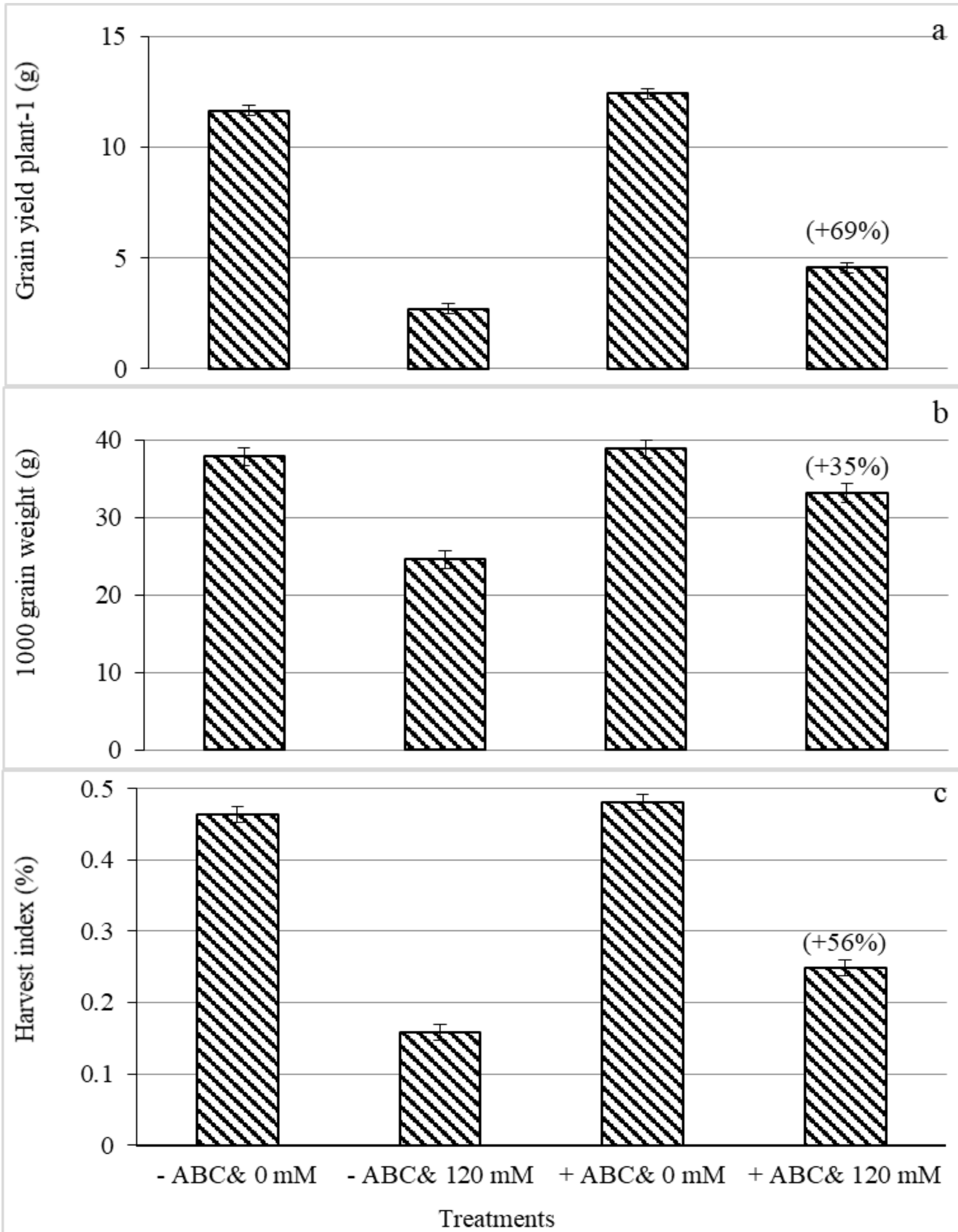

Figure (3): Effect of algal biochar and salinity treatments on (a) grain yield (g/plant[1]) (b) 1000 grains weight (g) and (c) harvest index (%) of wheat. Each datum indicates the mean value and vertical lines on top of bars indicate standard error of means (n = 4). Values in parenthesis indicate the percent increase from without the addition of ABC treatment to with the addition of ABC treatment under salinity stress (120mM NaCl)

DISCUSSION

Salinity is considered one of the abiotic environmental stresses that limit the production of many agricultural crops, due to the huge losses it causes because of its negative impact on many morphological, physiological, and biochemical processes of the plant (Kibria and Hoque, 2019, Ehtaiwesh et al., 2024). Therefore, the development and use of modern technologies and nature-based solutions are urgently needed, it is suggested that algal biochar amendment could be an effective approach for sustaining crop production in salinity-affected soils (Mona et al., 2021; Kenneth et al., 2022). As aforementioned, the aim of the current study was to evaluate the ability of algal biochar to mitigate the negative effects of salinity on some growth and yield traits of wheat plants. The results here indicated that salinity significantly ($P< 0.001$) decreased the growth and yield traits of wheat, while the application of algal biochar mitigated the negative effects of salinity on wheat performance. This outcome agreed with previous studies, which reported that the application of biochar alleviated the negative effect of stress on crops, wheat (Akhtar et al, 2015), maize (Ullah et al., 2020; Ali et al., 2021), barley (Gul et al., 2023), rice (Chen et al., 2021), potato (Liu et al., 2017) and tomato (Almaroai and Eissa. 2020; Calcan et al., 2022). Salinity affects plant growth because it increases the osmotic pressure of the soil solution (Munns et al., 2020).

A high concentration of total salts in the soil leads to an increase in its osmotic pressure, which causes the plant to suffer when it acquires water from the soil (Zahra et al., 2020). In the present study, salinity treatment limited plant growth and yield, this reduction in growth and yield traits under salinity stress could be attributed to reduced water uptake, osmotic potential, and ions toxicity to the plants (Safdar et al., 2019; Munns et al., 2020). Soil salinity occurs when there is an accumulation of dissolved salts in the area of root growth at a concentration high enough to hinder the ideal growth of the plants. Dissolved salts in this study consist mainly of Na^+ and Cl^-, when these ions are presented in high concentrations in the root growth zone; they cause toxic effects on plant growth. It also causes an imbalance in the availability of essential nutrients, for example, the competition of Na^+ with K^+ ions. However, the application of algal biochar reduced the negative effect of salinity stress, which could be due to the adsorbing of toxic ions such as Na^+ and Cl^- from the soil solution. This finding agrees with early studies on potato plants (Akhtar et al., 2015; Ehtaiwesh, 2022c). Another possibility of the ability of biochar to minimize the negative effect of salinity stress may be due to the role of biochar in increasing the cation exchange capacity (Domingues et al, 2020; Mohamed et al., 2021; Ghassemi-Golezani and Rahimzadeh, 2024). Cation exchange capacity is a critical soil property that directly stimulates nutrient availability and plant growth (Antonangelo et al., 2024). Also, some studies have reported that adding biochar helped in nutrient and water retention, thus reducing irrigation needs, improving agricultural efficiency, and increasing crop yields (Fischer et al., 2019; Semida et al., 2019; Cui et al., 2022). In addition, many studies suggested that biochar is added to agricultural sandy and or acidic soil because it is used as a soil improver due to its unique ability to retain water and increase soil fertility, which increases agricultural production (Dai et al., 2020; Abideen et al., 2022).

CONCLUSION

Algal biochar is a sustainable and eco-friendly approach for improving plant performance under stressed conditions. The experiment was conducted to investigate the potential effects of algal biochar applied to wheat grown under saline and non-saline conditions. The study concluded that algal biochar positively enhanced wheat growth and yield indicated by better performance under both conditions. The main result overall is that algal biochar application had a major impact on the growth and yield traits of wheat compared with the controls either with, or without salinity stress. The study suggested that algal biochar could provide a significant revenue stream as a soil ameliorant and fertilizer under saline and non-saline conditions. However, more studies are required to explore the effect of algal biochar on both salt-tolerant and non-salt-tolerant plants in salt-affected soils.

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