

## AMELIORATING THE TOXIC IMPACTS OF SALINITY ON SUDAN GRASS BY EXOGENOUS APPLICATION OF SEAWEED EXTRACT, AMINO ACIDS AND NUTRIENTS

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**Abstract.** A pot experiment was conducted to investigate the effect of seaweed, amino acid and nutrient solution on growth and physiological performance of Sudan grass growing under salinity stress. The experiment consisted of different levels of salinity stress 0 mM, 50 mM, 100 mM, 150 mM, 200 mM, and 250 mM and exogenous application of seaweed extract, amino acids and nutrients; control, soil application of seaweed extract, amino acids and nutrients and foliar application of seaweed extract, amino acids and nutrients. Soil application of seaweed, amino acid and nutrient solution was found best in ameliorating the toxic effect of salinity on root length, shoot fresh weight, root fresh weight, number of leaves, number of roots, number of tillers, leaf area, leaf width, leaf length, shoot dry weight and root dry weight. The same treatment also significantly improved in chlorophyll a, chlorophyll b, total chlorophyll, carotenoids, proline, phosphate and sulphate contents. On the other hand, foliar spray of seaweed extract, amino acids and nutrients resulted in maximum phenolics, flavonoids, nitrates, shoot potassium and

calcium as compared to control. Hence, soil application of seaweed, amino acids and nutrient solution was found effective in mitigating salinity impacts.

**Keywords:** *amino acids, biomass production, nutrient concentration, phenolics, secondary metabolites, Sudan grass*

## Introduction

Salinity stress affects 20% of irrigated land globally, which results in severe yield losses and is a major environmental problem that limits agricultural output globally (Shrivastava and Kumar, 2015). In many arid and semi-arid environments, salinity stress is a key abiotic stress factor that inhibits agricultural yield (Munns and Tester, 2008). Salinity stress affects plants in a number of different ways, including reduced development, decreased photosynthesis, ion toxicity, and oxidative stress (AbdElgawad et al., 2018).

Phytoplankton are microscopic plant-like organisms, while seaweed refers to macroscopic marine algae (Raghunandan et al., 2019). Thallophyta aquatic plants flourish in intertidal and subtidal areas with 0.1% photosynthetic light (Dhargalkar et al., 2001; Arioli et al., 2015). Seaweeds are ecologically and commercially significant aquatic creatures that are adaptable, abundant, and tenacious. Seaweeds have been used for food, feed, industrial gums, and medicines since ancient times, but are often underused (Dhargalkar and Pereira, 2005). Many studies have shown that seaweeds promote plant growth (Russo and Berlyn, 1990; Zodape et al., 2011; Rao and Chatterjee, 2014) are a novel source of antioxidants, plant hormones, osmo-protectants, plant nutrients, and other bioactive metabolites of pharmaceutical and industrial importance (Raghunandan et al., 2019).

Amino acids, and seaweed extracts have shown the promising results to improve the plant performance and mitigate the toxic effects of salinity stress (Rouphael and Colla, 2020; Hussein et al., 2021). Phytohormones, amino acids, and vitamins are just a few of the bioactive components found in seaweed extracts (Hamouda et al., 2022) that have been proven to boost plant growth and development and enhance plant tolerance to abiotic challenges, including salt stress (Khan et al., 2021). Plants rely on amino acids for a wide variety of metabolic processes, including osmotic adjustment, reactive oxygen species scavenging, and gene expression control (Huang et al., 2020). Nutritional solutions, including potassium and calcium, help plants maintain their cellular ion balance in the face of salt stress, perhaps mitigating its negative effects on growth (Wu et al., 2019).

Seaweed extract applications have been shown to improve seed germination and establishment, crop performance and yield and biotic and abiotic stress resistance (Khan et al., 2009). Seaweed extract foliar spray is a common method for increasing above-ground biomass. Improved organ development and drought and disease resistance are agronomic benefits (Digruher et al., 2018). Biostimulants improve plant development by enhancing photosynthetic pigments (Voko et al., 2022), carbohydrates, proteins, phytohormones, and phytochemistry (Nemahunguni et al., 2019, 2020; Gupta et al., 2021).

Chlorophyll a, b, and carotenoids are the major photosynthesis pigments. The changes in quantity of pigment and photosynthesis under salt stress are utilized to choose tolerant and sensitive cultivars (Zhong and Cheng, 2017; Houimli et al., 2010). Salinity increases stomatal resistance and disrupts photosynthetic pigment production, affecting membrane functions and photosynthesis (Karabal et al., 2003; Sairam, 2005). Nutritional components may be applied foliarly as well as soil. Nutritional spraying accelerates plant development by reducing the delay between element uptake and utilization (Taiz and Zeiger, 2002). Amino acids have an amine group, a carboxylic acid

group, and a variable side chain. These molecules include carbon, hydrogen, oxygen, and nitrogen (Mecha et al., 2023). Amino acids improve nutrient absorption, translocation, leaf photosynthesis, and biomass production (Souri and Hatamian, 2019). Applying amino acids, particularly glycine, may improve plant growth and quality in several crops (Aghayie Noroozlo et al., 2019; Garcia et al., 2011; Keutgen and Pawelzik, 2008; Souri et al., 2017).

Agricultural crops are susceptible to abiotic stresses like salinity. Salinity in agricultural soils is a widespread issue, adversely affecting crop growth and yield. This dual challenge poses a significant threat to food security and agricultural sustainability. Thus, there is a pressing need to develop strategies that enhance salinity tolerance in plants. The goal of this study to determine the effect of seaweed, amino acids and nutrient solution in mitigating toxic effects of salinity stress in Sudan grass.

## **Materials and methods**

### ***Growth conditions and plant material***

A pot experiment was conducted at Islamia University of Bahawalpur, Bahawalnagar Campus (30.012764° N, 73.278286° E) Punjab, Pakistan in between April 2022 to August 2022 using CRD with three replicates. Sudan grass seeds were sown in pots (5 kg capacity) using loamy soil (pH 8.0; organic matter 0.89%; total N 0.07%; available P 10 ppm; available K 115 ppm; saturation 23% and electrical conductivity 3.8  $\text{dsm}^{-1}$ ) as growth medium.

### ***Experimental details***

A pot experiment conducted to evaluate commercially available product “Tipple Boost” containing seaweed, amino acid and nutrients (Biostimulant) against NaCl stress on Sudan grass. The experiment consisted of different levels of salinity stress 0 mM, 50 mM, 100 mM, 150 mM, 200 mM, and 250 mM and exogenous application of seaweed extract, amino acids and nutrients; control, soil application of seaweed extract, amino acids and nutrients and foliar application of seaweed extract, amino acids and nutrients. After 7 days of salinity treatment, soil and foliar application of seaweed extract, amino acids and nutrients (1000 ppm) was done to only experimental group. All other agronomic practices were kept constant.

### ***Measurement of growth traits***

The heights of three randomly selected plants from each pot was measured and averaged. Likewise, root and shoot lengths of same plants were measured and averaged.

### ***Determination of photosynthetic pigments***

Photosynthetic pigments were measured using fresh leaf tissue extracted in 80% acetone. The concentration of chlorophyll *a*, chlorophyll *b*, total chlorophyll and carotenoids were determined by the methods as described by Lichtenthaler (1987).

### ***Determination of amino acids and secondary metabolites***

Free amino acid concentration was determined by adding 1 mL of a buffered crude extract to 1 mL of pyridine and 1 mL of ninhydrin in test tubes. The volume of the

sample was made to 25 mL, and the test tubes were heated in a water bath at 90°C for 30 min and absorbance was noted at 570 nm (Hamilton et al., 1943).

### ***Phenolic contents***

To determine phenolic contents, we added 1 ml aliquot of gallic acid extracts to a 25 ml volumetric flask with 9 ml distilled water. A reagent blank was made using distilled water then this mixture was shaken by adding one milliliter of Folin-Ciocalteu phenol reagent. Thereafter, the mixture was added with 10 ml of 7% Na<sub>2</sub>CO<sub>3</sub> and absorbance was noted at 550 nm.

### ***Flavonoid content***

Flavonoid content was determined by aluminum chloride colorimetric test described by Zhishen et al. (1999). 1 ml aliquot of quercetin extracts was added to a 10 ml volumetric flask with 4 ml distilled water. The flask received 0.30 ml of 5% NaNO<sub>2</sub> and 0.3 ml of 10% AlCl<sub>3</sub> after 5 min. After 5 min, 2 ml of 1 M NaOH was added and volume was made to 10 ml and absorbance was noted at 510 nm.

### ***Proline contents***

Proline was determined by procedure described by Bates et al. (1973). 0.5 g of leaf samples were taken and extract was obtained and mixed with 3% (w/v) sulphosalicylic acid and filtered and heated at 100°C for 1 h in water bath. Then this mixture was added with acid ninhydrin and glacial acetic acid and absorbance was noted at 520 nm.

### ***Ionic concentration***

Ion measurements were performed according to a procedure described by Carrillo et al. (2011). Samples of the Sudan grass plant were dried in an oven at 65 degrees Celsius and then powdered. After that, 0.5% of the pulverized material was mixed with two acids (hydrochloric acid, HCl, and nitric acid, HNO<sub>3</sub>) in a 1:2 ratio and heated to 180°C for digestion. After that, water was added to dilute the samples and they were filtered again. The chloride concentration was measured using a chloride analyzer, while sodium, calcium, magnesium, and potassium were measured by a flame photometer.

### ***Statistical analysis***

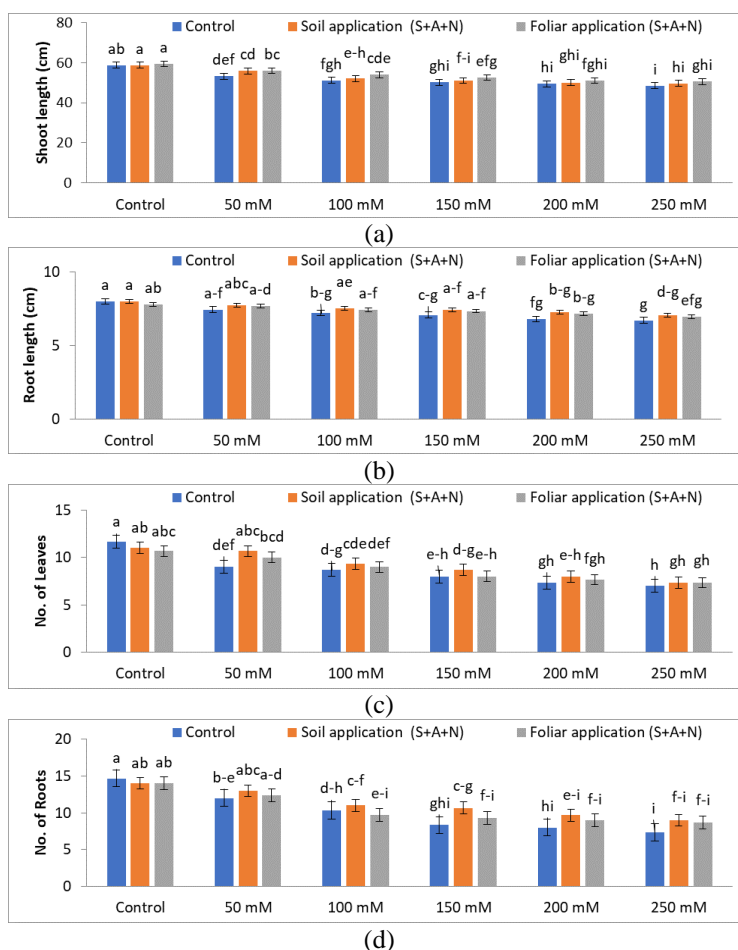
The pot experiment was conducted using completely randomized layout (CRD) with three replications. All data will be subjected to analysis of variance via CoStat Software (version 6.2; CoHort Software, Monterey, CA, USA) and differences among treatments was compared by using LSD test ( $P < 0.05$ ).

## **Results**

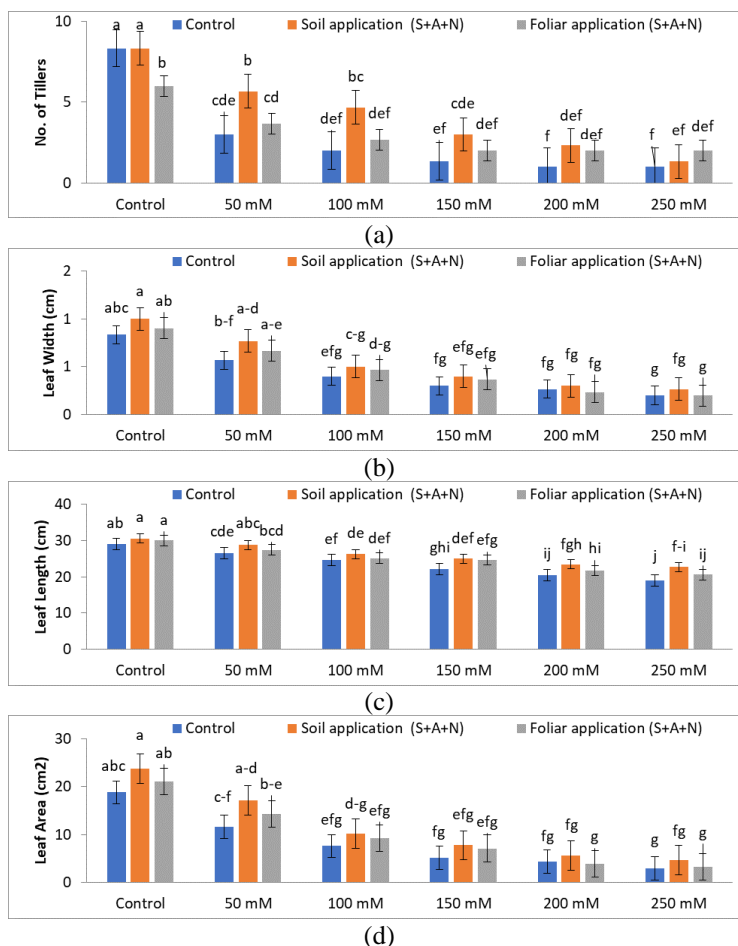
### ***Growth parameters***

Salinity seems to have a large effect on plant development, as shown by the analysis of variance (ANOVA), which revealed significant reduction ( $P < 0.05$ ) for salinity across all evaluated growth parameters. The control treatment with no salt significantly increased shoot length, whereas the application of 250 mM NaCl

significantly decreased shoot length. There was found gradual reduction in shoot length as NaCl level increased (*Fig. 1*). Shoot length was increased in salt conditions by the foliar application of seaweed, amino acids, and nutrients (S + A + N), followed by the soil application (*Fig. 1*). The control without (S + A + N) treatment significantly decreased shoot length when subjected to salt (*Fig. 1*). The length of roots, the number of leaves, the number of roots, the number of tillers, the width of leaves, the length of leaves, and the area of leaves all followed similar patterns (*Figs. 1 and 2*). Under salt conditions, foliar application of seaweed, amino acids, and nutrients (S + A + N) was the most significant results in boosting root length, number of leaves, number of roots, number of tillers, leaf width, leaf length, and leaf area, followed by soil application (*Figs. 1 and 2*). In conclusion, salinity has significant effect in reduction of plant growth, whereas (S + A + N) variation and its combination with salinity exhibited only a moderate effect. In order to increase plant development under salt conditions, foliar and soil application of seaweed, amino acids, and nutrients (S + A + N) was shown to be the most effective remedy. These results highlight the need to regulate salt and nutrients in comparable agricultural or environmental situations to maximize plant growth and yield.



**Figure 1.** The impact of seaweed, amino acids and nutrient solution (1000 ppm) application on shoot length (A), root length (B), leaves (C) and number of roots (D) of Sudan grass plant under salinity 0 mM, 50 mM, 100 mM, 150 mM, 200 mM, and 250 mM (NaCl). Data is the mean value of three replication ( $n = 3$ ) along with standard errors (SE). The same letters with different values showing no significant difference (LSD test)

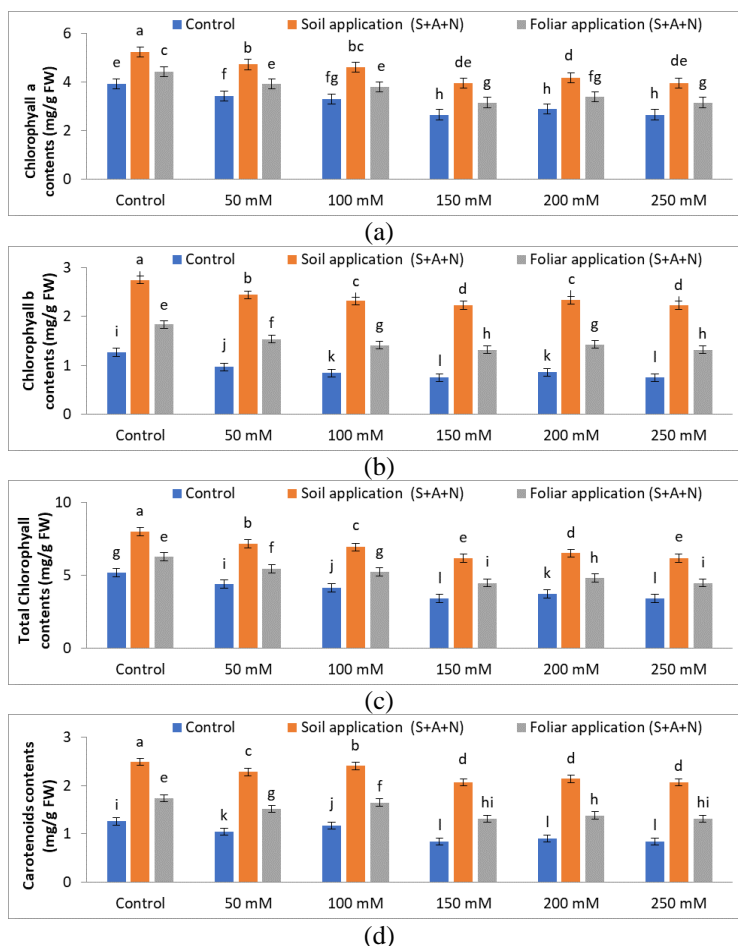


**Figure 2.** The impact of seaweed, amino acids and nutrient solution (1000 ppm) on tillers (A), leaf width (B), leaf length (C) and leaf area (D) of Sudan grass plant under salinity 0 mM, 50 mM, 100 mM, 150 mM, 200 mM, and 250 mM (NaCl). Data is the mean value of three replication ( $n = 3$ ) along with standard errors (SE). The same letters with different values showing no significant difference (LSD test)

### Photosynthetic pigments

The synthesis of photosynthetic pigments was significantly ( $P < 0.05$ ) decreased under salinity stress (Fig. 3). However, seaweed, amino acids and nutrient solution reversed the toxic effects salinity and markedly improved the synthesis of chlorophyll and carotenoids under normal and stress conditions. Chlorophyll a, b, and total chlorophyll, were all found highly significant in the control treatment without salt, whereas they were all found to lowest at 250 mM NaCl. Chlorophyll a, b, total chlorophyll, and carotenoid concentrations all decreased gradually as NaCl concentrations increased. Soil application of seaweed, amino acids, and nutrients (S + A + N) was the most efficient remedy for boosting chlorophyll a, chlorophyll b, total chlorophyll, and carotenoids under salinity conditions. The least effective response in all these pigments under salinity was shown in the control treatment without (S + A + N). Soil application of (S + A + N) results highly significant chlorophyll b, total chlorophyll, and carotenoids relative to the control and foliar spray treatments across all salinity levels. Soil applications of (S + A + N) was found highly significant,

the results were in following order from highest to lowest at concentrations of photosynthetic pigments 0 mM, 50 mM, 200 mM, 150 mM, 100 mM, and 250 mM NaCl respectively. The results of this experiment showed that seaweed, amino acids, and nutrients (S + A + N) application and salinity have significant effects on plant growth variables such as chlorophyll a and b concentrations, as well as total chlorophyll and carotenoid content. Soil application of seaweed, amino acids, and nutrients (S + A + N), followed by foliar application, was the most effective method to increase chlorophyll and carotenoid content in plants grown in salty environments. The results highlight the significance of seaweed, amino acids, and nutrients (S + A + N) and the use of specialized remedies in ameliorating the detrimental effects of salt on plant pigments and, followed by plant health and growth.



**Figure 3.** The impact of seaweed, amino acids and nutrient solution (1000 ppm) on chl a (A), chl b (B), total chl (C) and carotenoids (D) of Sudan grass plant under salinity 0 mM, 50 mM, 100 mM, 150 mM, 200 mM, and 250 mM (NaCl). Data is the mean value of three replication (n = 3) along with standard errors (SE). The same letters with different values showing no significant difference (LSD test)

### Biomass production

The exogenous application of seaweed, amino acid and nutrients solution appreciably improved the root and shoot growth and biomass production under control and salinity

stress conditions. Soil application of seaweed, amino acid and nutrient solution significantly increased root and shoot growth as compared to control (*Table 1*). The control, soil and foliar application of seaweed, amino acid and nutrient solution treatments results were in following order soil  $\geq$  foliar  $\geq$  control in terms of biomass production (*Table 1*).

**Table 1.** Fresh and dry weights (g) of shoots of Sudan grass plants under application seaweed, amino acids, nutrients and salinity

		SFW (g)	RFW (g)	SDW (g)	RDW (g)
Control	Control	7.22 <sup>ab</sup>	0.75 <sup>abc</sup>	2.73 <sup>a</sup>	0.25 <sup>a</sup>
	50 mM	0.71 <sup>c</sup>	0.73 <sup>abc</sup>	2.48 <sup>a</sup>	0.12 <sup>de</sup>
	100 mM	8.00 <sup>a</sup>	0.72 <sup>abc</sup>	2.43 <sup>a</sup>	0.11 <sup>ef</sup>
	150 mM	8.33 <sup>a</sup>	0.71 <sup>abc</sup>	2.41 <sup>a</sup>	0.08 <sup>i</sup>
	200 mM	7.33 <sup>a</sup>	0.69 <sup>bc</sup>	2.37 <sup>a</sup>	0.04 <sup>k</sup>
	250 mM	5.10 <sup>b</sup>	0.68 <sup>c</sup>	2.35 <sup>a</sup>	0.04 <sup>k</sup>
Soil application (S + A + N)	Control	8.02 <sup>a</sup>	0.76 <sup>a</sup>	2.67 <sup>a</sup>	0.25 <sup>a</sup>
	50 mM	7.76 <sup>a</sup>	0.75 <sup>ab</sup>	2.59 <sup>a</sup>	0.16 <sup>b</sup>
	100 mM	7.60 <sup>a</sup>	0.74 <sup>abc</sup>	2.53 <sup>a</sup>	0.13 <sup>d</sup>
	150 mM	7.49 <sup>a</sup>	0.73 <sup>abc</sup>	2.50 <sup>a</sup>	0.10 <sup>fg</sup>
	200 mM	7.37 <sup>a</sup>	0.72 <sup>abc</sup>	2.46 <sup>a</sup>	0.09 <sup>h</sup>
	250 mM	7.24 <sup>a</sup>	0.71 <sup>bc</sup>	2.41 <sup>a</sup>	0.07 <sup>i</sup>
Foliar application (S + A + N)	Control	8.06 <sup>a</sup>	0.75 <sup>ab</sup>	2.69 <sup>a</sup>	0.24 <sup>a</sup>
	50 mM	7.68 <sup>a</sup>	0.74 <sup>abc</sup>	2.56 <sup>a</sup>	0.14 <sup>c</sup>
	100 mM	7.55 <sup>a</sup>	0.73 <sup>abc</sup>	2.52 <sup>a</sup>	0.11 <sup>e</sup>
	150 mM	7.44 <sup>a</sup>	0.72 <sup>abc</sup>	2.48 <sup>a</sup>	0.09 <sup>gh</sup>
	200 mM	7.36 <sup>a</sup>	0.71 <sup>abc</sup>	2.45 <sup>a</sup>	0.06 <sup>j</sup>
	250 mM	7.24 <sup>a</sup>	0.70 <sup>bc</sup>	2.41 <sup>a</sup>	0.06 <sup>j</sup>

Data is the mean values of three replication (n = 3). Comparison of means was performed using LSD test at 5% level. SFW = shoot fresh weight; RFW = root fresh weight; SDW = shoot dry weight; RDW = root dry weight

### ***Amino acids and secondary metabolites***

Among amino acids and secondary metabolites proline contents were found highly significant in soil application of seaweed, amino acid and nutrient solution as compared to control and foliar application of seaweed, amino acid and nutrient solution. The contents significantly decreased as concentration of salinity increased in all experimental units. Phenolics and flavonoids, contents were found highly significant in foliar application of seaweed, amino acid and nutrient solution as compared to control and soil application of seaweed, amino acid and nutrient solution. The phenolics contents increased as salinity level increased. Total free amino acids contents found significant in control conditions as compared to soil and foliar application of seaweed, amino acid and nutrient solution. Total free amino acids in control conditions and foliar application of seaweed, amino acid and nutrient solution increased as salinity level increased while in soil application of seaweed, amino acid and nutrient solution declined as salinity level increased (*Table 2*).



**Table 2.** Phenolics, proline, TFAA and Flavonoids in Sudan grass plants under application seaweed, amino acids, nutrients and salinity

		<b>Phenolics (mg AE)</b>	<b>Proline (mol g<sup>-1</sup> FW)</b>	<b>TFAA (mg g<sup>-1</sup> proteins)</b>	<b>Flavonoids (mg GAE)</b>
Control	Control	31.92 <sup>c</sup>	30.52 <sup>ab</sup>	130.73 <sup>ef</sup>	6.88 <sup>a</sup>
	50 mM	32.57 <sup>c</sup>	30.15 <sup>b</sup>	141.64 <sup>de</sup>	6.99 <sup>a</sup>
	100 mM	35.71 <sup>c</sup>	33.85 <sup>ab</sup>	153.34 <sup>bc</sup>	7.56 <sup>a</sup>
	150 mM	36.03 <sup>c</sup>	35.51 <sup>ab</sup>	134.28 <sup>e</sup>	7.58 <sup>a</sup>
	200 mM	35.99 <sup>c</sup>	35.23 <sup>ab</sup>	136.35 <sup>e</sup>	7.43 <sup>a</sup>
	250 mM	36.22 <sup>c</sup>	36.57 <sup>ab</sup>	159.10 <sup>ab</sup>	7.54 <sup>a</sup>
Soil application (S + A + N)	Control	35.99 <sup>c</sup>	32.49 <sup>ab</sup>	160.77 <sup>a</sup>	6.92 <sup>a</sup>
	50 mM	38.08 <sup>c</sup>	35.83 <sup>ab</sup>	156.49 <sup>ab</sup>	7.00 <sup>a</sup>
	100 mM	38.56 <sup>c</sup>	36.94 <sup>ab</sup>	104.57 <sup>i</sup>	7.53 <sup>a</sup>
	150 mM	36.19 <sup>c</sup>	37.84 <sup>ab</sup>	109.86 <sup>gh</sup>	7.54 <sup>a</sup>
	200 mM	43.27 <sup>b</sup>	39.07 <sup>a</sup>	112.87 <sup>g</sup>	7.45 <sup>a</sup>
	250 mM	43.36 <sup>b</sup>	39.46 <sup>a</sup>	110.06 <sup>gh</sup>	7.57 <sup>a</sup>
Foliar application (S + A + N)	Control	44.35 <sup>b</sup>	31.10 <sup>ab</sup>	106.51 <sup>hi</sup>	7.17 <sup>a</sup>
	50 mM	44.62 <sup>b</sup>	34.22 <sup>ab</sup>	140.37 <sup>e</sup>	7.79 <sup>a</sup>
	100 mM	49.79 <sup>a</sup>	35.79 <sup>ab</sup>	146.05 <sup>d</sup>	7.67 <sup>a</sup>
	150 mM	50.93 <sup>a</sup>	36.69 <sup>ab</sup>	127.72 <sup>f</sup>	8.76 <sup>a</sup>
	200 mM	52.19 <sup>a</sup>	37.91 <sup>a</sup>	151.14 <sup>c</sup>	8.46 <sup>a</sup>
	250 mM	51.82 <sup>a</sup>	38.53 <sup>a</sup>	157.89 <sup>ab</sup>	8.26 <sup>a</sup>

Data is the mean values of three replication (n = 3). Comparison of means was performed using LSD test at 5% level. TFAA = total free amino acids

### ***Ionic contents***

Among ionic contents parameters phosphate and sulphate contents were found significant in soil application of seaweed, amino acid and nutrient solution as compared to control and foliar application of seaweed, amino acid and nutrient solution. The contents decreased significantly as concentration of salinity increased in all experimental units. Nitrates, shoot potassium and calcium contents were found highly significant in foliar application of seaweed, amino acid and nutrient solution as compared to control and soil application of seaweed, amino acid and nutrient solution. Sodium contents found significant in control conditions as compared to soil and foliar application of seaweed, amino acid and nutrient solution. Sodium contents increased in all experimental units as level of salinity increased (*Table 3*).

### **Discussion**

The present study conducted to investigate the ameliorating the impact of salinity by novel seaweed, amino acid and nutrient solution on growth and physiological components of Sudan grass. In this experiment application of commercially available product “Tipple Boost” containing seaweed, amino acid and nutrients as soil and foliar application against salinity stress and salinity independently observed on growth and physiological components of Sudan grass. The results of present investigation revealed

that salinity stress reduced the Sudan grass growth (*Figs. 1 and 2*). The similar findings also showed by AbdElgawad et al. (2018) the detrimental influence of salt on plant development was highlighted by the fact that the greatest values for all growth metrics and pigment levels were continuously found in the control treatment without salinity. The relevance of a low-salt environment for plant development is shown by this control condition. Similar findings were also discussed by many researchers as salinity stress affects plants in a number of different ways, including reduced development, decreased photosynthesis, ion toxicity, and oxidative stress. Salinity stress also induced ionic toxicity (*Table 3*) which damaged proteins, lipids, membranes and disturbed nutrient uptake, and caused a marked reduction in plant growth (Aouz et al., 2023) Among growth parameters root length, shoot fresh weight, root fresh weight, number of leaves, number of roots, number of tillers, leaf area, leaf width, leaf length, shoot dry weight and root dry weight were found best in soil application of seaweed, amino acid and nutrient solution against salinity and shoot length was found best in foliar application of seaweed, amino acid and nutrient solution.

**Table 3.** Nitrate, phosphate, sulphate, shoot potassium, calcium and sodium contents of Sudan grass plants under application seaweed, amino acids, nutrients and salinity

		NO <sub>3</sub> (mg g <sup>-1</sup> DW)	PO <sub>4</sub> (mg g <sup>-1</sup> DW)	SO <sub>4</sub> (mg g <sup>-1</sup> DW)	KS (mg g <sup>-1</sup> DW)	Ca (mg g <sup>-1</sup> DW)	Na (mg g <sup>-1</sup> DW)
Control	Control	15.15 <sup>ab</sup>	9.73 <sup>bc</sup>	9.39 <sup>abc</sup>	23.38 <sup>bcdef</sup>	8.69 <sup>a</sup>	19.82 <sup>gh</sup>
	50 mM	14.39 <sup>ab</sup>	9.37 <sup>c</sup>	9.12 <sup>abc</sup>	23.11 <sup>cdef</sup>	8.64 <sup>a</sup>	20.48 <sup>gh</sup>
	100 mM	14.43 <sup>ab</sup>	9.44 <sup>c</sup>	7.77 <sup>abc</sup>	23.11 <sup>def</sup>	8.64 <sup>a</sup>	23.76 <sup>g</sup>
	150 mM	14.37 <sup>ab</sup>	9.32 <sup>c</sup>	7.18 <sup>c</sup>	22.28 <sup>ef</sup>	8.49 <sup>a</sup>	30.33 <sup>de</sup>
	200 mM	14.09 <sup>ab</sup>	8.51 <sup>c</sup>	7.47 <sup>c</sup>	22.56 <sup>ef</sup>	8.54 <sup>a</sup>	33.61 <sup>bx</sup>
	250 mM	12.49 <sup>b</sup>	8.27 <sup>c</sup>	7.12 <sup>c</sup>	21.19 <sup>f</sup>	8.29 <sup>a</sup>	36.24 <sup>a</sup>
Soil application (S + A + N)	Control	15.41 <sup>a</sup>	14.57 <sup>a</sup>	10.01 <sup>a</sup>	24.20 <sup>bcde</sup>	8.84 <sup>a</sup>	18.51 <sup>h</sup>
	50 mM	14.57 <sup>ab</sup>	14.21 <sup>a</sup>	9.74 <sup>ab</sup>	23.93 <sup>bcde</sup>	8.79 <sup>a</sup>	19.16 <sup>h</sup>
	100 mM	14.68 <sup>ab</sup>	14.28 <sup>a</sup>	8.28 <sup>abc</sup>	23.93 <sup>bcde</sup>	8.79 <sup>a</sup>	21.79 <sup>g</sup>
	150 mM	14.55 <sup>ab</sup>	14.16 <sup>a</sup>	7.80 <sup>abc</sup>	23.11 <sup>ef</sup>	8.64 <sup>a</sup>	27.04 <sup>f</sup>
	200 mM	14.35 <sup>ab</sup>	13.35 <sup>ab</sup>	7.96 <sup>abc</sup>	23.38 <sup>cdef</sup>	8.69 <sup>a</sup>	30.33 <sup>de</sup>
	250 mM	12.67 <sup>b</sup>	13.11 <sup>ab</sup>	8.54 <sup>abc</sup>	22.01 <sup>f</sup>	8.44 <sup>a</sup>	33.61 <sup>bc</sup>
Foliar application (S + A + N)	Control	15.70 <sup>a</sup>	10.05 <sup>ab</sup>	10.06 <sup>a</sup>	27.76 <sup>a</sup>	9.49 <sup>a</sup>	17.85 <sup>h</sup>
	50 mM	14.93 <sup>ab</sup>	9.69 <sup>c</sup>	9.78 <sup>a</sup>	26.66 <sup>ab</sup>	9.29 <sup>a</sup>	18.51 <sup>h</sup>
	100 mM	14.97 <sup>ab</sup>	9.76 <sup>bc</sup>	8.44 <sup>abc</sup>	26.12 <sup>bc</sup>	9.19 <sup>a</sup>	21.79 <sup>g</sup>
	150 mM	14.91 <sup>ab</sup>	9.70 <sup>c</sup>	7.84 <sup>abc</sup>	25.29 <sup>bcd</sup>	9.04 <sup>a</sup>	28.36 <sup>ef</sup>
	200 mM	14.63 <sup>ab</sup>	8.83 <sup>c</sup>	8.14 <sup>abc</sup>	25.29 <sup>bcd</sup>	9.04 <sup>a</sup>	31.64 <sup>cd</sup>
	250 mM	13.03 <sup>b</sup>	8.59 <sup>c</sup>	7.78 <sup>abc</sup>	24.75 <sup>bcd</sup>	8.94 <sup>a</sup>	34.27 <sup>b</sup>

Data is the mean values of three replication (n = 3). Comparison of means was performed using LSD test at 5% level

Digruber et al. (2018) also described that seaweed extract foliar spray increase above-ground biomass; improve organ development and drought and disease resistance. All morphological parameters in control, foliar and soil application against salinity declined as concentration of salinity increased. AbdElgawad et al. (2018) also described similar results as Root and leaf biomass both declined dramatically when NaCl concentration was increased, with the greatest reduction occurring at the highest (150 mM) NaCl concentration. Soil application of seaweed, amino acid and nutrient solution was found maximum growth. The control, soil and foliar application of seaweed, amino acid and nutrient solution in following order soil ≥ foliar ≥ control.

Similar results also described by many studies shown that seaweeds promote plant growth (Zodape et al., 2011; Rao and Chatterjee, 2014; Ali et al., 2023).

Among physiological parameters including Chlorophyll a, Chlorophyll b, Total Chlorophyll, Carotenoids, Proline, Phosphate and Sulphate contents were found highest values in soil application of seaweed, amino acid and nutrient solution as compared to control and foliar application of seaweed, amino acid and nutrient solution. In above mentioned parameters values declined as concentration of salinity increased in all experimental units. Phenolics, flavonoids, nitrates, shoot potassium and calcium contents were found highest performance in foliar application of seaweed, amino acid and nutrient solution as compared to control and soil application of seaweed, amino acid and nutrient solution. Similar findings also found in previous studies as Applying amino acids, particularly glycine, may improve plant growth and quality in several crops (Aghayie Noroozlo et al., 2019; Garcia et al., 2011; Keutgen and Pawelzik, 2008; Souri et al., 2017). The phenolics contents increased as salinity level increased. There are two parameters total free amino acids and sodium contents whose values found highest in control conditions as compared to soil and foliar application of seaweed, amino acid and nutrient solution. Total free amino acids in control conditions and foliar application of seaweed, amino acid and nutrient solution increased as salinity level increased while in soil application of seaweed, amino acid and nutrient solution declined as salinity level increased. Similar results were also discussed as Sodium contents increased in all experimental units as level of salinity increased. Previous studies also found similar results as amino acids improve nutrient absorption, translocation, leaf photosynthesis, and biomass production (Souri and Hatamian, 2019).

Overall, the findings suggest that salinity is a significant factor affecting plant growth and chlorophyll content. In this experiment, seaweed, amino acids, and nutrients (S + A + N) considerably mitigate salt's harmful effects. To improve plant development and chlorophyll content in salty environments, the application of seaweed, amino acids, and nutrients to the soil showed better results. To increase plant growth and production in saline environments, and so contribute to sustainable agriculture and environmental management, a thorough understanding of the interaction between seaweed, amino acids, and nutrients (S + A + N) and salt is essential. Spinelli et al. (2010) also describe similar results, increases in vegetative growth (10%), leaf chlorophyll content (11%), stomatal density (6.5%), photosynthetic rate (27%), fruit output (27%), and berry weight (27%) were all seen after biostimulant application. The most notable effect was a rise in plant biomass, with increases of up to 27% in shoot dry matter and 76% in root dry matter.

## Conclusion

Application of seaweed, amino acid and nutrient solution enhanced the growth of Sudan grass plants under salinity and by enhancing photosynthetic pigments, antioxidant activities, nutrient homeostasis, and reducing the uptake of toxic ions. Hence the use of seaweed, amino acid and nutrient solution as soil and foliar application can be an effective approach to mitigate the toxic effects of and salinity. The better vegetative growth and a higher yield indicate that, in the treated plants, more mineral nutrients and photo assimilates are available for the different plant organs.

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