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Research Article

Salt Stress Alleviation Potential of Endophytic Fungi on Okra Plant Growth

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ABSTRACT

Saline environments for plants have a negative impact on their biochemical, molecular, and physiological features, resulting in a loss in plant growth and yield in most parts of the world. Hormonal imbalance, ionic toxicity, ROS production, impaired nutrient mobilization, and osmotic stress all contribute to plant growth loss. The symbiotic connection of several endophytes with their host plants has improved plant tolerance to various abiotic challenges in recent years. So, the current study aimed to see how *Aspergillus japonicus* and salinity affected growth parameters, plant biochemistry, antioxidant enzyme status, endogenous IAA, and various anion and cation concentrations in okra. According to the findings, applying salt lowered numerous growth properties, including Chlorophyll a and b, total chlorophyll, and endogenous IAA. Enhanced values of a/b (chlorophyll), total carotenoids, total proteins, total carbs, total lipids, total proline, total phenol, total flavonoids, total tannin, lipid peroxidation, catalase, and ascorbate peroxidase were seen at similar salt levels. In okra (Sanaf green), salt application resulted in salt accumulation and higher levels of Cl, Na, Ca/K, and Na/K ratio, while low contents of K, P, Mg, Ca, O, N, and C were found. The results of this study show that using *Aspergillus japonicus* promotes plant resistance to saltwater environments, as evidenced by the fact that using the fungal strain increased the values of many growth parameters, metabolites, anti-oxidant enzyme activity, and endogenous IAA. In the same way, using *Aspergillus japonicus* lowered the amount of lipid peroxidation in the okra plant. The accumulation of salts in okra after application of *Aspergillus japonicus* resulted in an increase in Ca, Mg, N, C, K, Cl, O, and P values, as well as a decrease in the Ca/K and Na/K ratios, Cl, and Na values. According to the findings, salt stress causes stunted development and altered biochemistry in okra, while the presence of *Aspergillus japonicus* aids in plant salt tolerance. As a result, this strain is an excellent candidate for testing as a biostimulant/biofertilizer to improve the growth of essential crops in Pakistan's salt-affected areas.

Keywords: *Aspergillus japonicus*, seedling growth, Okra, Salt stress, catalase, IAA, ionomic status, metabolic status

INTRODUCTION

Waterlogging, low and high temperatures, drought, salinity, soil pH, pesticides, and other environmental stressors all had a detrimental impact on crop yields and were regarded as limiting variables in agricultural productivity (Shrivastava and Kumar, 2015). Soil and irrigation water salinity is the world's most serious problem, resulting in ongoing reductions in cultivable land and plant output. This element has a significant impact on 20 percent of arable land and 33 percent of irrigated land around the world (Ali et al., 2014). Soil salinization is increasing by 10% per year due to rock erosion, bad agricultural practices, and

saline water irrigation (Shrivastava and Kumar, 2015). Salinity causes some common gene responses related to plant metabolism, physiology, and morphology to be elicited, which leads to a reduction in the germination, seedling growth, flower setting, and fruit setting processes (Abreu et al., 2013). Under prolonged exposure to high salt concentrations in soil, sodium and chloride ions accumulate in the soil, resulting in limited potassium uptake (Xiong et al., 2002). This factor also disrupts the cell's water potential and osmotic balance, causes oxidative stress, and causes oxidative damage to nucleic acids, proteins, and membrane lipids (Chawla et al., 2013).

Abelmoschus esculentus is a Malvaceae plant that is farmed for its young leaves and soft fruits in America, Asia, South Europe, and Africa, particularly in Nigeria (Khomsug et al., 2010). This plant is grown for human consumption in several parts of the world. This crop is high in minerals, calcium, and vitamins, particularly vitamin C, all of which are deficient in the diets of people in underdeveloped nations. This plant is slightly salt tolerant and is planted in dry summers with a lot of irrigation, primarily in Mediterranean nations (Habib et al., 2016). It's also known as okra, but it's also known as lady's fingers or gumbo in many English-speaking nations. It is a popular vegetable in Nigeria because of its delicate fruits and young leaves. It can be found in Africa, Asia, Southern Europe, and America (Khomsug et al., 2010). The principal components of okra include vitamins, mineral salts, and calcium, which are inadequate in the diet of individuals living in underdeveloped countries in some locations. High salt levels in the root zone of this crop have a negative impact on a variety of processes, including yield, growth, physiological processes, root turgor pressure, density, and growth, as well as reduced water absorption (Achour, 2016; Ayub et al., 2018).

Various tactics and initiatives, such as adopting salt-resistant cultivars, organic matter conditioners, plant genetic engineering, and salt stress mitigation compounds, are now being employed to mitigate the negative effects of salt stress on plant development and productivity (Zhang et al., 2000). Scientists are finding it difficult to design effective and less expensive techniques because of this stress element. Salt-tolerant microorganisms have been shown to be the best option for improving crop growth and productivity under salt stress. After association, plant growth promoting endophytes appear beneficial and assist plants cope with various abiotic and biotic challenges (El-Awady et al. 2015; Zhao et al. 2016). These endophytes improve the antioxidant system, osmolyte production, plant nutrient uptake efficiency, and phytohormonal signaling to increase salt tolerance in the host plant (Kushwaha et al., 2020). Different plant growth-promoting activities are involved in beneficial interactions formed between plants and different plant growth-promoting microorganisms (Anwar et al., 2016), such as nitrogen fixation, different volatile compounds and hormones, nutrient solubilization, biocontrol of disease organisms, production of siderophore, and induction of systemic resistance (Anwar et al., 2016; Vejan et al., 2016). This study aimed to emphasize salinity's negative impacts on okra development, biochemistry, and ionomic state, as well as how to mitigate these effects using endophytic fungus (*Aspergillus japonicus*).

MATERIALS AND METHODS

Collections and purification of Endophytic fungi and preparation of spore suspension:

The Plant-Microbe Interaction Laboratory, Department of Botany, Abdul Wali Khan University Mardan, provided *Aspergillus japonicus* (EU-26), which was previously identified by Ismail et al. (2018). After repeated sub-culturing on PDA, the strain was purified and cultured at 25°C for two weeks to yield sufficient spores (conidia). Using a sterile No. 21 blade, the mycelia and spores generated on the PDA media were scraped and deposited in a 50 mL sterile conical tube. For spore dispersion, 20 mL sterile water was poured to a 50 mL conical tube and vortexed for 5 minutes. Then, using Watman filter paper No. 2, this mixture (which

contained mycelia and spores) was filtered. To obtain spore suspension, filter paper was folded and placed in a flask, and the mixture was filtered. For the experiment, the final concentration of spores in water was adjusted to 5 x10^7 spores/mL.

Characterization of *Aspergillus japonicus*

7 days of fungal culture filtrate were filtrated and centrifuged at 3000rpm for 10 mins. The fungal culture supernatant was used to analyze various hormones and metabolites. The estimation of indole-3-acetic acid (IAA) in fungal filtrate was carried out by a colorimetric technique using the Salkowski reagent (Benizri *et al*., 1998). The two parts of the supernatant and one part of the Salkowski reagent (2 mL of 0.5 $FeCl_3$ in 49ml of 70% $HCLO_4$ solution) were added and allowed to incubate for 30 min in a dark chamber. The O.D was determined at 540 nm. Salicylic acid was estimated using the spectrophotometric method of Warrier et al., 2013. 100 µL of the fungal filtrate was mixed with 0.1% of freshly prepared ferric chloride, and the volume of the reaction mixture was made up to 3 mL. The complex formed between Fe^{3+} ions and SA, which was violet in color, was measured spectrophotometrically (PerkinElmer Lambda 25 double beam spectrophotometer) at 540 nm.

The total phenol content of fungal filtrate was recorded by applying the method proposed by Mallick and Singh (1980). The reaction mixture consists of 2 mL filtrate and 0.5 ml of Folin-Ciocalteau reagent. The mixture was incubated for 4 minutes at 25, and 2 ml of a 20% Na_2CO_3 solution was added and the tubes were then placed in a boiling water bath for 1 minute. The tubes were cooled down and absorbance was measured at 650 nm in a spectrophotometer (PerkinElmer Lambda 25 double beam spectrophotometer). To determine the total flavonoid content in fungal filtrate, the aluminum chloride method was used (Mervet et al., 2009). 0.5ml of filtrate was taken, mixed with 0.1ml of 10% $AlCl_3$, followed by the addition of 0.1ml of 10% potassium acetate and 4.8 ml of methanol (80%). The test solution was shaken vigorously. After 30 minutes of incubation, the absorbance at 415 nm was recorded. Aqueous ethanol (80%) was used as blank.

Experimental setup

The effect of *Aspergillus japonicus* (EU-26) inoculation on okra growth was studied in a pot experiment. The inoculation of *Aspergillus japonicus* endophyte was part of a completely randomized scheme (EU-26). ARI (Agriculture Research Institute) Tarnab, Peshawar provided seeds of okra var. sanaf green. Before beginning the experiment, the seeds were tested for viability. Seeds of equal size were sterilised for 60 seconds with 0.1 percent mercuric chloride and washed three times with distilled water. A total of 12 plastic pots (8.5 cm diameter and 12.5 cm depth) with a basal outlet for leaching were placed and filled with 300 grams of sandy loam soil. Before filling the pots, soil samples were collected and analyzed for different physical and chemical properties (Table 1). The disinfected seeds were germinated in pots, and spore suspension of fungi @1ml/gm was applied in the necessary set at the seedling stage. After five days of spore suspension, a 200 mM NaCl salt solution was administered twice a week, whereas control sets were irrigated with tap water. The experimental design included four treatment sets, three replications, and five plants per pot in each duplicate.

Set I: Okra (Var. Sanaf green) seedlings watered with 0 mM NaCl.

Set II: Okra (Var. Sanaf green) seedlings irrigated with 200 mM NaCl.

Set III: Okra (Var. Sanaf green) seedlings irrigated with 0 mM NaCl and treated with fungal spores (*Aspergillus japonicus*, EU-26).

Set IV: Okra (Var. Sanaf green) seedlings irrigated with 200 mM NaCl and treated with fungal spores (*Aspergillus japonicus*, EU-26).

Okra plants were collected after the 35-day experiment was completed. Shoot and root systems were separated, and roots were thoroughly rinsed with tap water to eliminate soil

particles clinging to them. The fresh weights of the shoots and roots were measured, then oven-dried for 72 hours at 60 degrees Celsius, and weighed to obtain the dry weights.

Table 1. Physical and chemical properties of soil used in the experiment.

Soil Variables	Content
Texture	Sandy-loam
Sand (%)	72.9
Silt (%)	12.7
Clay (%)	14.1
CEC (dS/cm)	4.3
ECe (dS/m)	1.34
pH	7.3
Organic matter (%)	1.25
Organic Carbon (%)	4.26
Carbonates (meq/l)	1.41
Bicarbonates (meq/l)	2.87
Chlorides (meq/l)	1.32

Assay for Chlorophyll and carotenoids

Maclachlan and Zalik (1963) established a method for estimating and calculating total chlorophyll and carotenoid content in fresh-leaf samples. The fresh-leaf sample was powdered in 3ml of 80% acetone before being centrifuged at 1000 rpm for 5 minutes. Plant material pellets were centrifuged three times, and the supernatant was collected in a 7 ml final volume of acetone (80%). Optical densities for chlorophyll-a were measured at 663 nm, 645 nm for chlorophyll-b, and 480 and 510 nm for carotenoids.

Assay for Total Carbohydrate

This work utilized the Anthron method to extract and estimate total sugars in fresh-leaf samples (Yemm and Willis 1954). In 10ml D.H_2O, 0.5 grams of fresh leaves were crushed. The centrifugation operation took 5 minutes at 3000 rpm. For estimating, 0.1 mL of supernatant was mixed with 1 mL of phenol (80%), and the sample was incubated in an incubator for 10 minutes at a time. The mixture was then incubated for another hour with 5 mL concentrated - H_2SO_4. At 485 nm, the optical density of the solution was measured.

Assay for Total Proteins

Plant total proteins were quantified using a well-defined method described by Bradford (1976). A 5 ml phosphate buffer (pH= 7, 0.1M Potassium Phosphate) was used to homogenize a 0.1-gram leaf sample in an ice-cold pestle and mortar. After putting the extract in the freezer for 20 minutes, it was centrifuged at 12,000 rpm for 20 minutes. With the help of the buffer, the final volume of supernatant was increased to 5 ml, and then 4.8 ml buffer was added to 0.2 ml of the extract for dilution. In a clean test tube, 0.1 ml of diluted extract was added to 5 ml Bradford reagent to estimate total proteins. The O.D. was then measured using a spectrophotometer at 595 nm.

Assay for Total Lipids

The total lipid content was determined using Van Handel's (1985) well-established technique. 0.2 g of leaf sample was powdered in a chloroform: methanol (2: 1 v/v) mixture. After rapidly shaking the tube until it was completely dissolved, add 0.8 mL of 0.73 percent NaCl. Following

the completion of the process, three layers containing various chemicals were separated, with the lower layer containing lipids being collected through a separating funnel. After adding 0.1 mL sulfuric acid and shaking the mixture, it was heated at 100 ^{o}C for 10 minutes. Allow the sample to cool before adding 2.4 mL of vanillin reagent. When the pink color appeared, the absorbance was measured at 490 nm.

Assay for Total Proline

The method published by Bates et al. (1973) for extracting and estimating proline in plants was employed in this study. The leaf (0.5 g) was powdered in a 10-ml sulphosalicyclic acid (3%) solution and centrifuged for 5 minutes at 3000 rpm. 2 ml glacial acetic acid and 2 ml freshly made ninhydrin reagent were added to 2 ml supernatant. The mixture was heated to 100 °C in a glass tube for an hour. After cooling the tube, 4 mL of toluene was added to the mixture. After adding toluene, the layer was separated, and the optical density was measured at 520 nm.

Assay for Total Phenols

The Malick and Singh (1980) procedure was used for total-phenol extraction and estimation. With the help of D.H_2O, the leaf sample extract was separated in a glass tube, and the volume was increased to 3 ml. The extract was combined with 0.5 mL Folin ciocalteau reagent and 2 mL Na_2CO_3, and the absorbance of the solution was measured at 650 nm.

Assay for Total Flavonoids

El Far and Taie (2009) described a method for measuring total flavonoids in plants. A 5 g leaf sample was macerated in 50 mL of 80 percent ethanol for extract production. After a one-day incubation period, the extract was centrifuged at 10,000 rpm for 15 minutes. 250 µl of extract were combined with 1.25 µl of D.H_2O and 7 µl of a 5% $NaNO_2$ solution. After 5 minutes of incubation, add 150 µl of 10% $AlCl_3$.H_2O and incubate for 6 minutes in the incubator. 500 µl of 1M NaOH and 275 l D.H_2O are added to the solution, and the optical density is measured at 415 nm. Standard curves with varied levels of quercetin (15 g – 500 g) were utilized to calculate flavonoids, whereas ethanol (80%) was employed as a reagent blank.

Assay for Total Tannins

Akindahunsi and Oyetayo's (2006) method for determining total plant tannins was used. A 0.5-gm leaf sample was homogenized in 100 mL acetone (70%) and incubated for 6 hours in a shaking incubator. From a stock solution of tannic acid (50 g tannin acid diluted in 70% ethanol), serial dilutions were prepared (100 mL). Mix 950 l D. H_2O with 50 l extracts, then add 20 % Na_2CO_3 (2.5 ml) and the reagent (0.5 ml folin-phenol) to the solution. At 510nm, the absorbance of the solution was measured against a reagent blank.

Assay for Lipid peroxidation

Heath and Packer (1968) published a lipid peroxidation measurement method in plants using the thiobarbituric acid (TBARS) assay used in this study. 0.1 g of plant material was macerated in 2.5 ml of 0.1 percent tricholoro acetic acid and centrifuged at 3000 rpm for 20 minutes. 1 mL extract was combined with 2.25 mL thiobarbituric acid and 20% tricholoro acetic acid. The solution was heated to 95°C for 30 minutes, then cooled in an ice bath before being centrifuged for 10 minutes at 3000 rpm. The optical density of the solution was measured at 532nm and 600nm against reagent-blank.

Assay for Catalase Activity

Chandlee and Scandalios (1984) used a methodology to assess catalase enzyme activity in fresh leaves. 0.1 mL enzyme extract + 1 mL potassium phosphate buffer (100 mM, pH 7.0) + 0.4

mL H_2O_2 (200 mM) were added to the reaction mixture (final volume was 1.5 ml). The breakdown of H_2O_2 causes a reduction in absorbance, which is measured at 240nm after 30-second intervals (extinction-coefficient = 0.036-mM / cm).

Assay for Ascorbate Peroxidases Activity

Asada (1987) described a method for determining ascorbate peroxidase activity in plants. The mixture consisted of 0.1 ml enzyme extract + 0.2 mM ascorbic acid + 50 mM potassium phosphate buffer + 0.2 mM EDTA + 20 mM H2O2 (the final volume was 1 ml). At 290nm, the reduction in optical density was monitored every 30 seconds for up to 7 minutes.

Assay for Endogenous IAA Level

A technique established by Gordon and Weber (1951) was used to determine IAA in plant material. A 0.5-gm leaf sample was ground in 10 ml dH_2O to make the extract. Salkowski reagent (2 ml) was combined with 1 ml supernatant, and the solution was incubated in the dark for 30 minutes before being measured at 540nm.

Analysis of different Minerals

Plant samples were taken to test for several cations (K^+, Na^+, Mg^{+2}, and Ca^{+2}). The ash weight was calculated using 0.5 grams of dried plant material. The ash was mixed with 50 mL of deionized water, and the solution was used to make dilutions using deionized water for cation analysis. An atomic absorption spectrophotometer was used to measure the concentration of cations in various samples.

Statistical Analysis

Using SPSS, data from various factors were evaluated for the analysis of variance (one-way ANOVA) (Ver. 21). At the P0.05 level, the means of different treatments were examined and compared using DMRT (Duncan's multiple range test).

RESULTS

Characterization of *Aspergillus japonicus*

Culture filtrate analysis of *Aspergillus japonicus* showed a considerable level of IAA (19.19 µg/ml), salicylic acid (63.11 µg/ml), flavonoids (5.56 µg/ml), and total phenols (4.4 µg/ml) (Ismail et al., 2018).

Seedling establishment

According to the present investigation, when okra plants were subjected to salinity stress, plants showed significant ($P<0.001$) inhibition in shoot length (-46%), root length (-63%), Number of leaves (-43%), fresh weight (-58%) and dry weight (-66%) while comparing with control plants (Table 1). Under the same project, an association of *Aspergillus japonicas* with okra under both saline and normal conditions cause significant ($P<0.001$) and considerable changes in studied parameters. Under saline medium, endophytic association causes slightly lower values of reduction, as -41% in shoot length, -59% in root length, -9% in number of leaves, -50% in fresh weight while -53% in dry weight as compared to non-saline medium.

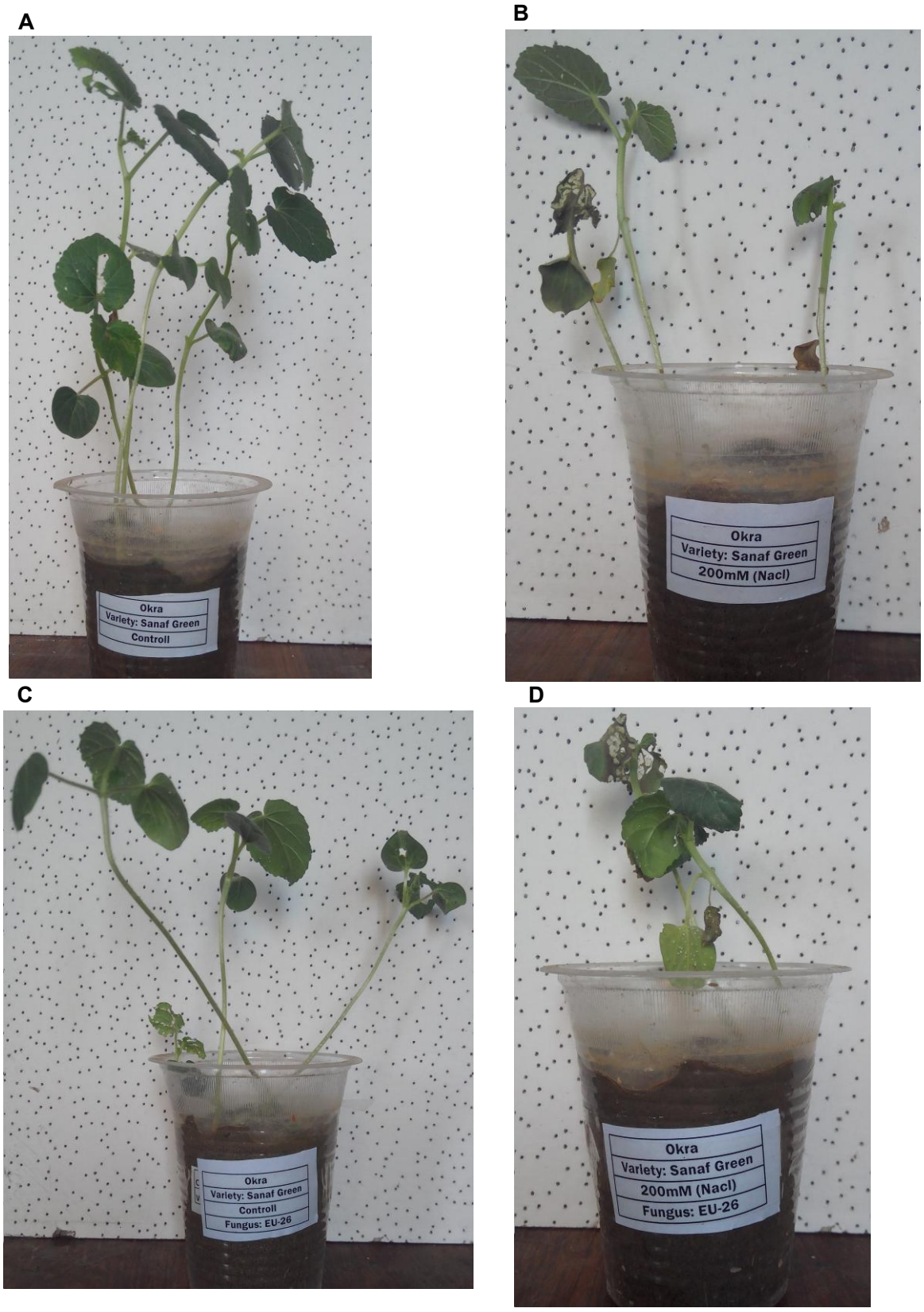

Figure 1. Effect of salt stress on Okra growth before and after association with *Aspergillus japonicus*.

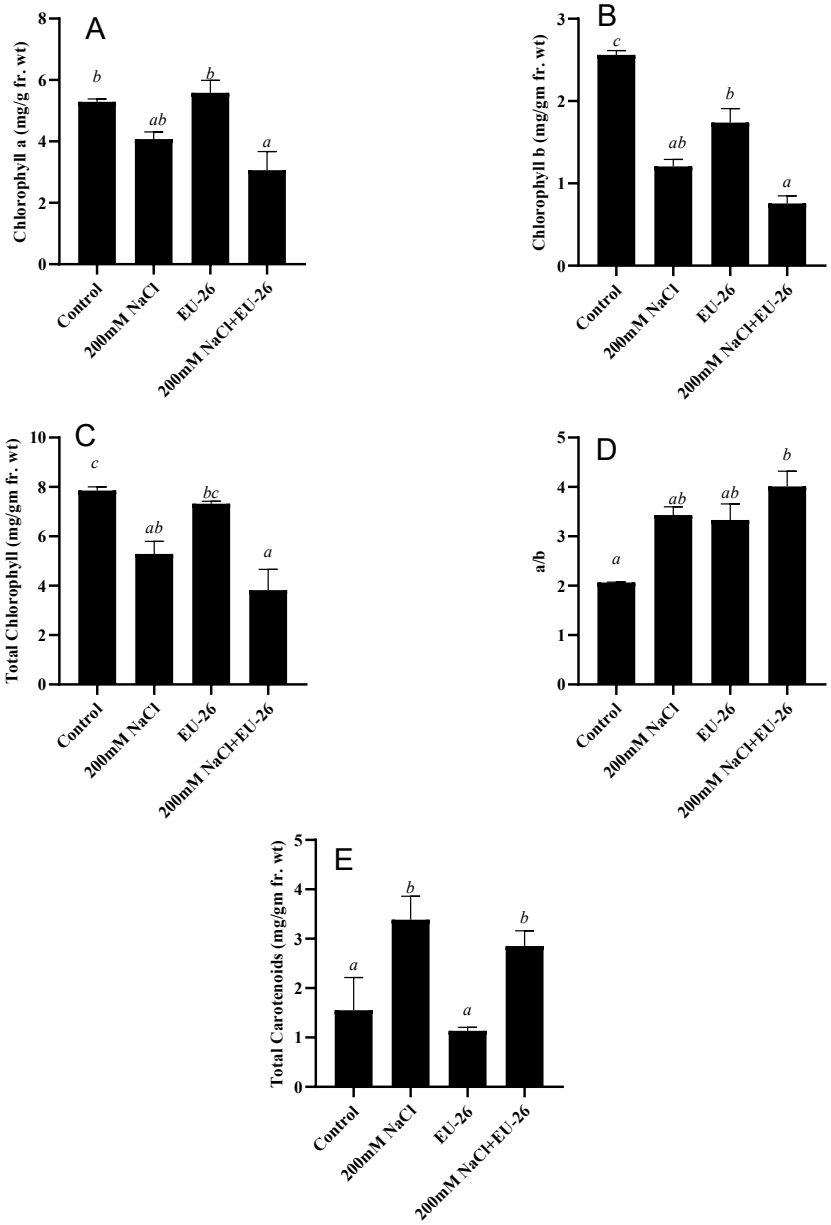

Figure 1. Beneficial effect of *Aspergillus japonicus* (EU-26) on Okra growth under normal and salt stress conditions. (**A**) chlorophyll a, (**B**) chlorophyll b, (**C**) total chlorophyll, (**D**) a/b (chlorophyll a and ratio) and (**E**) total carotenoids. Data bars are means ± SE (n = 3). Columns with italic letters denote the significant difference among treatments calculated by Duncan's multiple range test P<0.05.

Photosynthetic apparatus activity

According to the present investigation, when okra plants were subjected to salinity stress, plants showed significant (P<0.01) inhibition in chlorophyll a (-45%), chlorophyll b (-56%), total chlorophyll (-51%), while exhibited promotion in a/b ratio (66%) and total carotenoids (151%) while comparing with control plants (Figure 1). Under the same project, an association of *Aspergillus japonicas* with okra under both saline and normal conditions cause significant (P<0.01) and considerable changes in studied parameters. Under saline medium, endophytic association causes slightly lower values of reduction, as -22% in chlorophyll a, -53% in chlorophyll b, and -33% in total chlorophyll, while promotion values noted as 22% in chlorophyll a/b ratio while 118% in total carotenoids as compare to non-saline medium.

Total Carbohydrate, proteins, lipids, and proline contents

According to the present investigation, when okra plants were subjected to salinity stress, plants showed significant (P<0.001) promotion in total carbohydrates (25%), total proteins (209%), total lipids (115%), and proline (169%), while comparing with control plants (Figure 2). Under the same project, an association of *Aspergillus japonicas* with okra under both saline and normal conditions cause significant (P<0.001) and considerable changes in studied parameters. Under saline medium, endophytic association causes slightly lower values of promotion, as 127% in total carbohydrates, 130% in total proteins, 235% in total lipids, and 291% in proline as compared to non-saline medium.

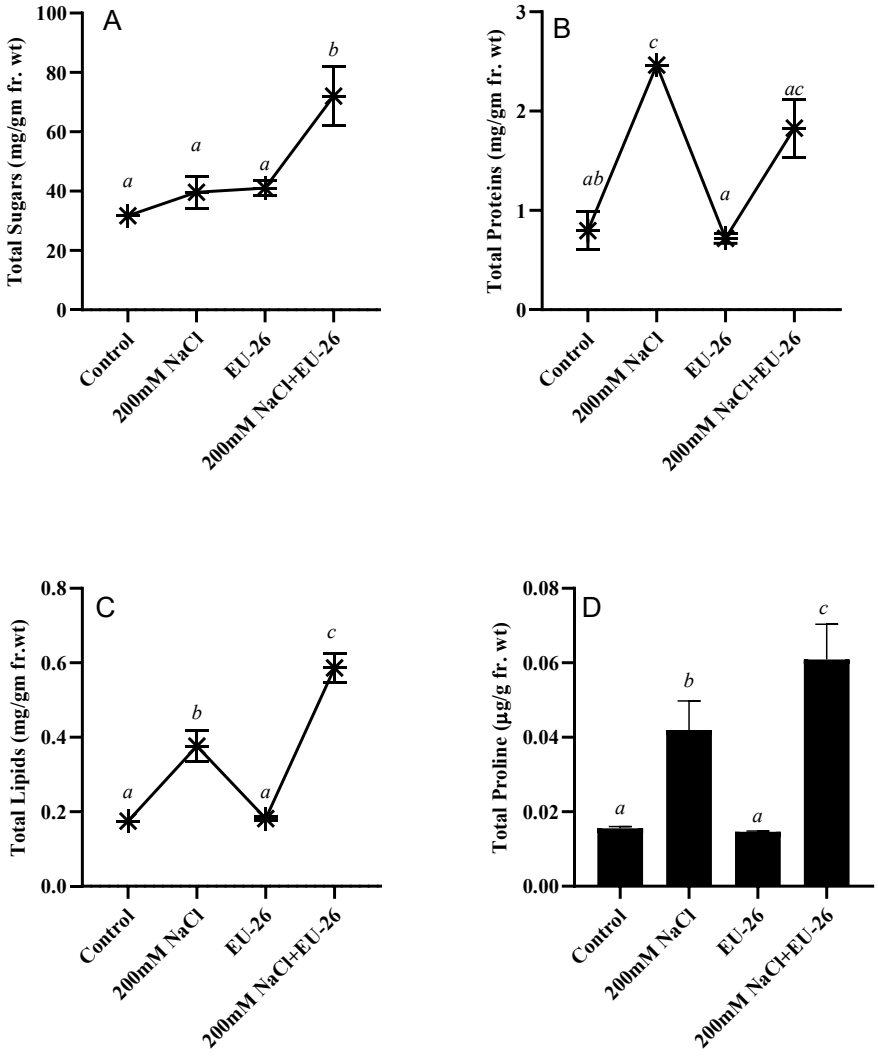

Figure 2. Beneficial effect of *Aspergillus japonicus* (EU-26) on Okra growth under normal and salt stress conditions. (**A**) total carbohydrates, (**B**) total proteins, (**C**) total lipids and (**D**) proline. Data bars are means ± SE (n = 3). Columns with italic letters denote the significant difference among treatments calculated by Duncan's multiple range test P<0.05.

Secondary metabolites

According to the present investigation, when okra plants were subjected to salinity stress, plants showed significant (P<0.01) promotion in total phenols (29%), total flavonoids (65%), and total tannins (144%) while compared with control plants (Figure 3). Under the same project, an association of *Aspergillus japonicas* with okra under both saline and normal conditions cause significant (P<0.01) and considerable changes in studied parameters. Under saline medium, endophytic association causes slightly lower values of promotion, as -32% in total phenols, 138% in total flavonoids, and 13% in total tannins, as compared to the non-saline medium.

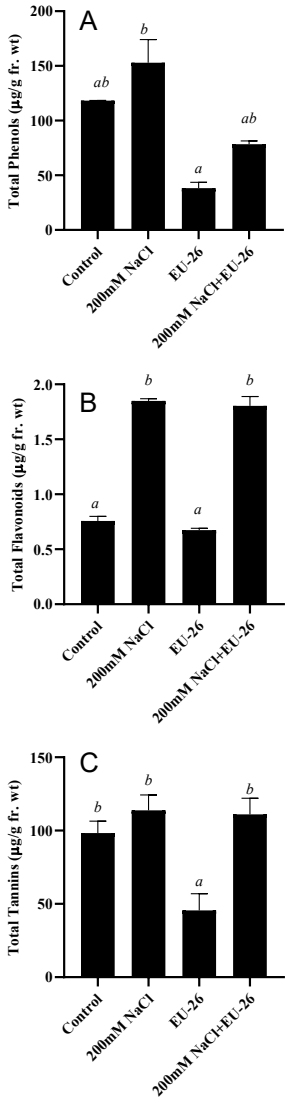

Figure 3. Beneficial effect of *Aspergillus japonicus* (EU-26) on Okra growth under normal and salt stress conditions. (**A**) total phenols, (**B**) flavonoids and (**C**) total tannins. Data bars are means ± SE (n = 3). Columns with italic letters denote the significant difference among treatments calculated by Duncan's multiple range test P<0.05.

Membrane Lipid peroxidation, Antioxidant enzymes, and IAA level

According to the present investigation, when okra plants were subjected to salinity stress, plants showed significant (P<0.01) promotion in lipid peroxidation (401%), catalase (44%), ascorbate peroxidase (44%), and reduction observed in endogenous IAA level (-45%) while comparing with control plants (Figure 4). Under the same project, an association of *Aspergillus japonicas* with okra under both saline and normal conditions cause significant (P<0.01) and considerable

changes in studied parameters. Under saline medium, endophytic association caused slightly lower values of promotion, as 310% in lipid peroxidation, 116% in catalase, and 6% in ascorbate peroxidase, while reduction was recorded as -67% in endogenous IAA level as compared to non-saline medium.

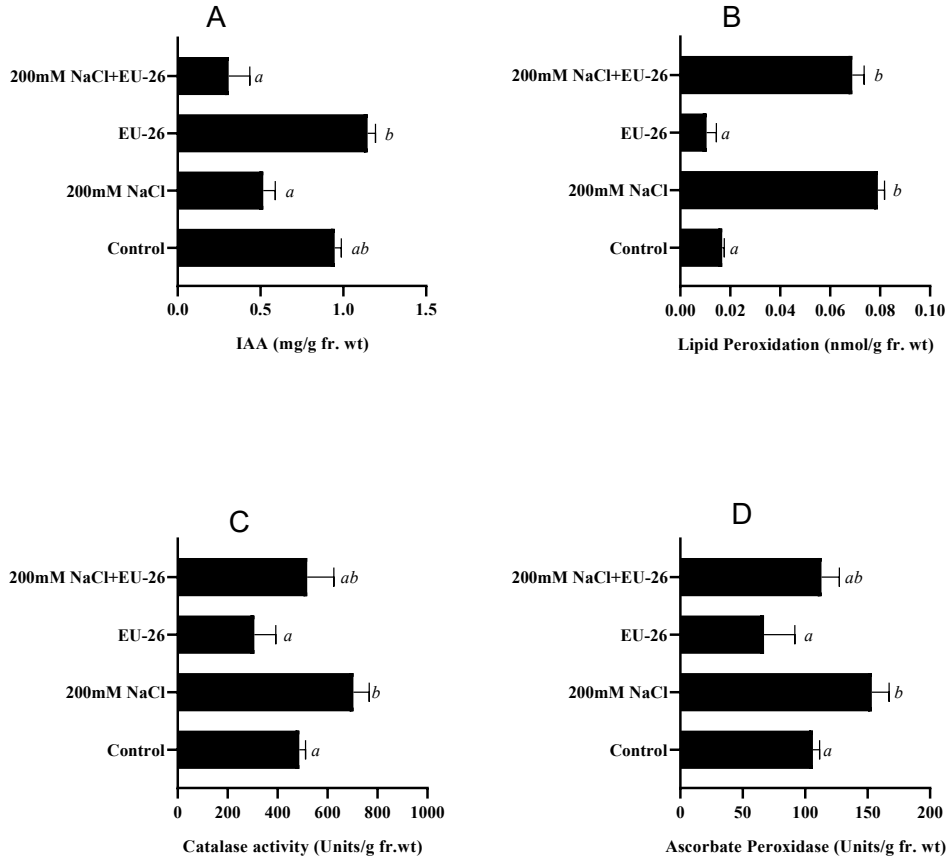

Figure 4. Beneficial effect of *Aspergillus japonicus* (EU-26) on Okra growth under normal and salt stress conditions. (**A**) IAA level, (**B**) MDA content, (**C**) Catalase and (**D**) Ascorbate peroxidase. Data bars are means ± SE (n = 3). Columns with italic letters denote the significant difference among treatments calculated by Duncan's multiple range test P<0.05.

Ions Concentration

According to the present investigation, when okra plants subjected to salinity stress, plants showed significant (P<0.001) promotion in sodium (545%), chloride (324%), Na/K (649%) and Na/Ca (1084%) while reduction observed in Potassium (-13%), Calcium (-45%), Magnesium (-25%), Phosphorus (-50%), Nitrogen (-59%) while comparing with control plants (Figure 5). Under the same project, an association of *Aspergillus japonicas* with okra under both saline and normal conditions cause significant (P<0.001) and considerable changes in studied parameters. Under saline medium, endophytic association causes slightly lower values of promotion, as 45% in sodium, 113% in chlorides, 56% in Na/K, and 69% in Na/Ca, while reduction recorded as -8% in potassium, -20% in calcium, -18% in magnesium, -40% in phosphorus, 3% in nitrogen as compare to non-saline medium.

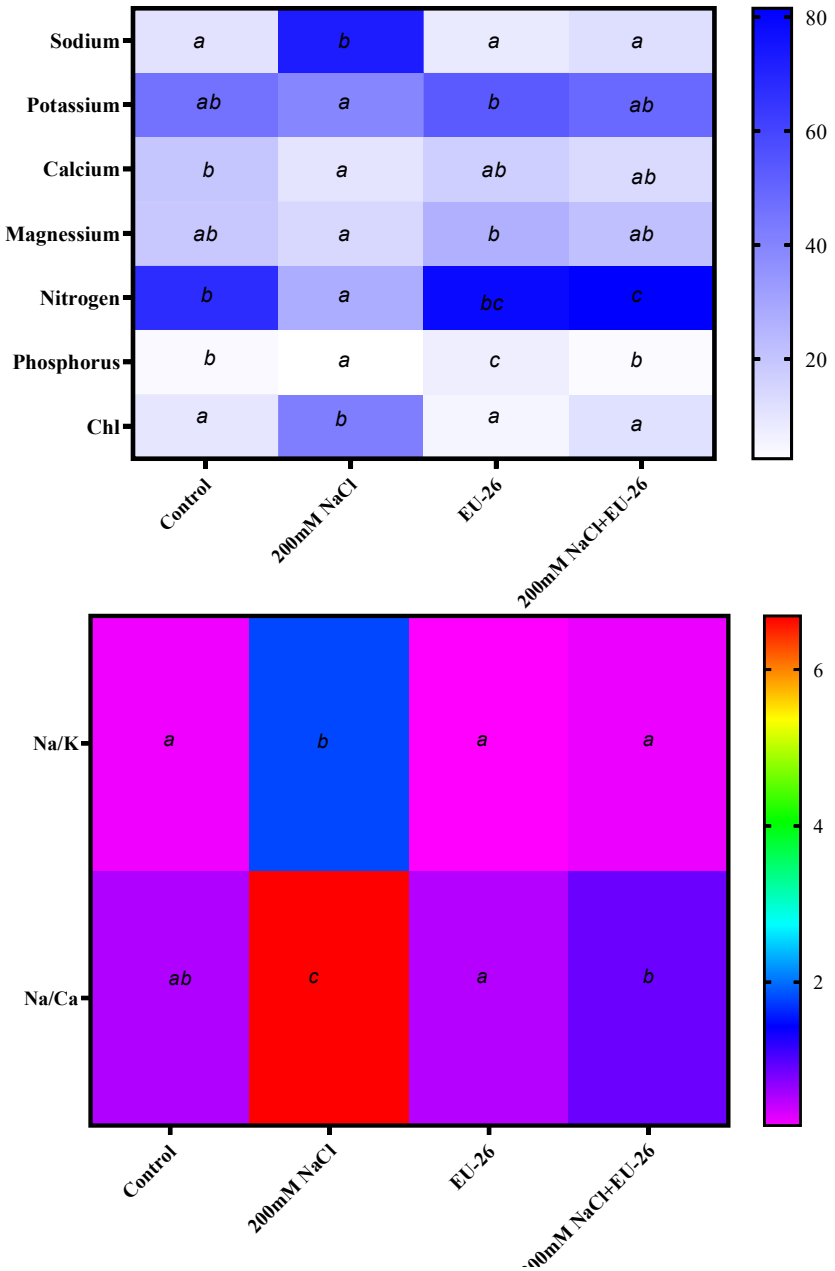

Figure 5. Beneficial effect of *Aspergillus japonicus* (EU-26) on Okra growth under normal and salt stress conditions with reference to different ions concentration. Data bars are means ± SE (n = 3). Columns with italic letters denote the significant difference among treatments calculated by Duncan's multiple range test $P<0.05$.

DISCUSSION

Effect of salt stress

The shortest distance between the top boundary (the highest point) of the primary photosynthetic tissues (excluding inflorescences) and the ground level is defined as plant height [Perez-Harguindeguy et al., 2013]. Plant roots are the primary organ for absorbing water and nutrients from a solution, whereas xylem vessels transfer nutrients to the aerial tissues. Furthermore, plant roots are the principal site for sensing saline levels, allowing the plant to respond quickly in order to retain functionality. By altering their morphology, roots can exclude and/or prevent potentially harmful chemicals. As okra plants were exposed to salinity stress, they showed significant reductions in shoot length, root length, number of leaves, and fresh

and dry weight when compared to non-saline control plants, according to the current study (Table 2). A drop in fresh or dry weight occurs when salt rises. Salt stress had the greatest impact on seedling growth, shoot height, and root length [Saddiq et al., 2019]. The elevated internal concentration of Na^+ ions decreases growth by reducing the water potential in the root zone, specific ion toxicity, and nutritional imbalance [Koyro, 2006]. A leaf (plural leaves) is the vascular plant stem's main lateral appendage, usually borne above ground and specialized for photosynthesis. Because the number of leaves during flowering is related to the plant's photosynthetic rate and reflects the potential yield, it is an important metric for plant growth (Nomura et al., 2017). The decrease in dry weight buildup could be related to increased cell wall stiffness caused by salinity-induced changes in cell wall structure. Osmotic stress is caused by salt stress in the root zone, and it affects cell ion homeostasis by inhibiting the uptake of important nutrients like K^+ and increasing the buildup of Na^+ and Cl^- [Paranchianakis and Chatzoulakis, 2005]. Increased Na^+ uptake competes with other nutritional ions, particularly K^+, resulting in K^+ shortage and a decreased K^+/Na^+ ratio in plants under salt stress [Kibria et al., 2017].

Table 2. Effect of *Aspergillus japonicus* (EU-26) on plant height, root length, number of leaves, fresh and dry biomass of Okra growing in 200 mM NaCl.

Treatment	Plant Height (cm)	Root Length (cm)	Number of Leaves	Fresh Biomass (g)	Dry Biomass (g)
Control	15.333±1.202^{b}	3.00±0.058^{b}	4.50±0.231^{b}	0.387±0.028^{b}	0.420±0.033^{b}
200mM NaCl	9.00±0.577^{a}	1.10±0.115^{a}	2.567±0.433^{a}	0.163±0.012^{a}	0.142±0.013^{a}
EU-26	16.333±0.882^{b}	3.233±0.145^{b}	4.167±0.186^{ab}	0.444±0.037^{b}	0.481±0.014^{b}
200mM NaCl+EU-26	8.333±1.453^{a}	1.233±0.273^{a}	3.367±0.291^{a}	0.195±0.033^{a}	0.198±0.044^{a}
Probability Level	***P<0.001***	***P<0.001***	***P<0.05***	***P<0.001***	***P<0.001***

Each data point is the mean of three replicates with ± standard errors. The columns represented with different letters are significantly different from each other as evaluated by DMRT analysis

Higher plants, algae, and cyanobacteria all contain chlorophyll, a photosynthetic pigment. They are essential for "light harvesting" in photosynthesis, which is an essential activity for the survival of both plants and animals (Humphrey, 2004). Photosynthetic pigment concentrations are easily quantified and are widely used to assess stress for regulatory purposes. When okra plants were exposed to salinity stress, they showed a large drop in chlorophyll a, chlorophyll b, and total chlorophyll while a significant increase in chlorophyll a/b ratio and carotenoids and total proline when compared to non-saline control plants (Figure 2). Salt-tolerant organisms have been less affected than salt-sensitive species in terms of photosynthetic rate [Iqbal et al., 2019]. Proline (Pro) is a crucial and versatile amino acid that can play a role in plant development and biotic and abiotic stress responses. Salinity produces many physiological and morphological changes, including the buildup of low molecular weight molecules known as compatible solutes like proline (Chen et al., 2007).

Carbohydrates are found in all living things and play a role in various critical processes. The carbohydrate ribose is an essential component of nucleic acids (RNA and DNA) in live cells. Still, carbohydrates also appear in various other compounds such as coenzymes, antibiotics, and poisons. Similarly, Plant lipids come in various forms and are necessary for cell function. They serve as a hydrophobic barrier for the membrane, ensuring the integrity of cells and organelles. Lipids are also stored in seeds in the form of chemical energy. They also serve as a signal molecule for cell metabolism regulation [Li-Beisson et al., 2013]. According

to the current study, when okra plants were exposed to salinity stress, they demonstrated a significant increase in total carbohydrates, proteins, and lipids compared to non-saline control plants (Figure 3). Salt stress has been found to raise the quantity of reducing sugars (sucrose and fructans) within the cells of a variety of plants from various species [Kerepesi and Galiba, 2000]. Trehalose buildup protects organisms against various physical and chemical challenges, including salt stress, and provides a carbohydrate reserve. In physiological reactions, they play an osmoprotective effect [Ahmad et al., 2013]. Stress proteins could be employed as crucial molecular indicators for improving salt tolerance utilizing genetic engineering approaches. Salt stress has been shown to increase total phospholipid content in both salt-sensitive and salt-tolerant species, including wheat root PM [Salama and Mansour, 2015], *C. roseus* cell suspensions [Elkahoui et al., 2004], and *Mesembryanthemum crystallinum* epidermal bladder cells [Barkla et al., 2018].

Plant phenolics play a role in protecting plants from ultraviolet radiation, viruses, parasites, and predators and contributing to their color. Flavonoids are a type of polyphenol secondary metabolite that can be found in a wide range of plants and diets. They're thought to contain antiviral, anti-inflammatory, cardioprotective, anti-diabetic, anti-cancer, anti-aging, and other bioactive properties. According to the current study, okra plants exposed to salinity stress revealed a considerable increase in total phenols, flavonoids, and tannins as compared to non-saline control plants (Figure 4). Phenolics impact various physiological processes in plants, including seed germination, cell division, and the manufacture of photosynthetic pigments [Tanase et al., 2019]. Bioremediation, allelochemicals, plant growth stimulation, and antioxidants as food additives have all been explored with phenolic compounds [Bujor et al., 2015]. Flavonoids are a class of low-molecular-weight compounds with a nucleus of 2-phenylchromone. The shikimic acid route is used to biosynthesize them from acetic acid/phenylalanine derivatives. A large number of PSMs, including terpenoids and steroids, phenolics and flavonoids, and alkaloids, have been reported to be produced, involved, or activated in cellular stress and defense response function when saline conditions influence plant growth physiology [Sytar et al., 2018]. For example, higher levels of production of aromatic compounds (e.g., alkaloids, isoprenoids, and phenols) and phenylpropanoids-derived compounds (e.g., tannins, flavonoids, and hydroxycinnamate esters) are thought to be mediated by salinity stress and free-radical scavengers, which represents an adaptation to the condition in secondary metabolite-producing plants [Sytar et al., 2018]. Tannins are phenolic secondary metabolites of plants with molar masses ranging from 300 to 30,000 Da. High molecular weight plant tannins can be found in connection with proteins or cell wall polysaccharides, whereas low molecular weight plant tannins are water-soluble molecules. They are a diverse category of (poly)phenolic compounds that are traditionally known for imparting astringent flavor to various plant parts (Zohra et al. 2016). Elfeel and Bakhashwain (2012) highlighted that when plants are exposed to salt, they produce a high amount of tannin, which has a detrimental impact on plant fiber and leaf quality.

The oxidative degradation of lipids having any number of carbon-carbon double bonds is known as lipid peroxidation. Lipid peroxidation is a well-known mechanism of cellular injury in both plants and animals, and it is used to detect oxidative stress in cells and tissues. Lipid peroxides degrade into a complicated succession of chemicals, including reactive carbonyl compounds, when exposed to air. Normal developmental processes, such as the synthesis of flavor and odor volatiles, the formation of molecules with growth-regulator-like functions, and senescence, all include lipid peroxidation. According to the current study, okra plants exposed to salinity stress revealed a considerable increase in lipid peroxidation, catalase, and ascorbate peroxidase activity as compared to non-saline control plants (Figure 5). Another study using the same experimental setup found that tobacco BY-2 cell cultures treated with proline reduced NaCl-induced cell mortality by reducing ROS generation and lipid

peroxidation while also improving membrane integrity [Banu et al., 2009]. Cavalcanti et al. (2004) found that under salt stress, cowpea plants accumulated greater amounts of malondialdehyde (MDA, a lipid peroxidation indicator) along with growth retardation. These findings matched those of those who found that salinity stress increased MDA, CAT, and POD activity. H_2O_2-scavenging enzyme ascorbate peroxidase (APX) is required for the protection of chloroplasts and other cell constituents against H_2O_2 and hydroxyl radicals (•OH) (Ali et al., 2019; Ibrahim et al., 2018). Ascorbate peroxidase employs ascorbate (AsA) as its particular electron donor to reduce H_2O_2 to water, producing monodehydroascorbate (MDHA), a univalent oxidant of ascorbate in the process (AA). AsA and dehydroascorbate are naturally disproportionate to monodehydroascorbate (DHA). The balance of the primary enzymatic H_2O_2 scavenging mechanisms in plants, ascorbate peroxidase (APX), glutathione peroxidase (GPX), and catalase (CAT), is critical for the suppression of harmful H_2O_2 levels in a cell [Apel and Hirt, 2004]. The ascorbate-glutathione cycle, in which ascorbate peroxidase (APX) isoenzymes play a key role in catalyzing the conversion of H_2O_2 into H_2O using ascorbate as a specific electron donor [Correa-Argunde et al., 2013], is a major hydrogen peroxide detoxifying system in plant cells under abiotic stressors.

The major auxin in higher plants, indole-3-acetic acid (IAA), significantly impacts plant growth and development. Plants and some plant diseases may both create IAA, which can be used to control plant development. Because of its potential to drive differential growth in response to gravity or light stimuli, auxin was identified as a plant growth hormone. According to the current study, when okra plants were exposed to salt stress, they demonstrated a considerable reduction in endogenous IAA compared to non-saline control plants (Figure 5). Auxin is a driver of various morpho-physiological processes [Zhao, 2010]. Auxin levels and transporter expression were reduced under salt stress conditions, preventing the plant from translocating auxins [Liu et al., 2015].

Plants do not require sodium, although it can be used in small amounts, similar to micronutrients, to aid in chlorophyll metabolism and synthesis. It facilitates the opening and closure of stomates, which helps manage internal water balance and can be used as a partial replacement for potassium in some plants. As okra plants were exposed to salinity stress, they showed considerable increases in sodium and chloride concentrations and Na/K and Na/Ca ratio when compared to non-saline control plants, according to the current study (Figure 6). Maintaining a balanced cytosolic Na^+/K^+ ratio has become a critical salt tolerance mechanism because Na^+ interacts with K^+ homeostasis, especially given its participation in multiple metabolic activities. Glycophytes and halophytes have roughly the same mechanism for Na^+ and K^+ absorption and translocation, although glycophytes are more vulnerable to ionic stress than halophytes. According to Shabala and Cuin (2008), both the Na^+/Ca^{+2} and the Na^+/K^+ ratios increased after applying salt, indicating that both are salt-sensitive indicators for plants. Shahzad et al. (2012) noticed high levels of different ratios (e.g., sodium/magnesium, sodium/potassium, and sodium/calcium) at the total plant level, as well as in the symplast and apoplast of developing tissues of seedlings when they grew mung bean seedlings in salt stress conditions.

For plant growth and development, potassium is a critical nutrient. Calcium also activates enzymes and conveys signals that help cells coordinate their activity. Potassium and calcium play an important role in the integrity and function of cell membranes [Pal et al., 2011]. Solute mobility, stomatal control, molecular communication for cell defense systems, and cell repair under stress are just a few of the physiological functions that calcium helps with. Phosphorus is a key plant macronutrient that accounts for around 0.2 percent of a plant's dry weight. Because it is a component of important compounds, including nucleic acids, phospholipids, and ATP, plants cannot thrive without a consistent supply of this vitamin. According to the current study, when okra plants were exposed to salinity stress, their

potassium, calcium, magnesium, phosphorus, and nitrogen concentrations dropped significantly compared to non-saline control plants (Figure 6). Crop tolerance to high NaCl concentrations is partly connected to inorganic phosphorus availability, according to Kalifa et al. (2000). Phosphorus absorption was reduced in various crop species due to high NaCl levels. Root uptake and phosphorus translocation to shoots were both reduced. Plants use nitrogen in a number of ways, including uptake, assimilation, translocation, recycling, and remobilization as the plant ages. Salt stress can affect a variety of physiological and biochemical processes, as well as inhibit carbon (C) and nitrogen (N) metabolism enzyme activity (Shakeel et al., 2017). According to Zhang et al., (2009), NaCl stress has a significant impact on carbon and nitrogen metabolism, resulting in a reduction in the amount of these nutrients within the plant. Carbon and nitrogen metabolism are considered necessary for plant growth and development. Magnesium is necessary for activating enzymes involved in respiration, photosynthesis, and the production of nucleic acids. It helps with phosphate metabolism by transporting phosphate molecules throughout the plant. Magnesium helps carbs (sugars and starches) move across the body and boosts the creation of oils and fats. According to Rahneshan et al., (2018), a decrease in magnesium ions content was detected during salt stress due to various enzymes essential for chlorophyll synthesis and catalysis. After salinity treatment, Amirjani (2010) found lower magnesium and calcium ion concentrations in soybean plants.

Response of Okra After Association with Endophyte

Salt stress reduces leaf size, internode length, and length, resulting in stunted growth of salt marsh and sand-dune plants [Koyro, 2006]. Physiological reactions such as changes in water status, mineral nutrition, ion balance, stomatal behavior, and photosynthetic effectiveness all contribute to the decline in growth [Cheng et al., 2020]. Biomass refers to the mass of living organisms such as plants, animals, and microorganisms, as well as cellulose, lignin, carbohydrates, lipids, and proteins from a biochemical standpoint. Under the same experiment, the connection of *Aspergillus japonicas* with okra under both salty and normal conditions resulted in significant changes in the shoot length, root length, number of leaves, and fresh and dry weight (Table 2). *Trichoderma spp.* has been shown to colonize plant roots, form symbiotic relationships with a wide range of host plants, and enhance plant growth and development, according to a number of prior studies (Harman, 2011). Furthermore, under salt stress circumstances, *Trichoderma parareesei* was found to promote tomato lateral root formation and growth promotion (Rubio et al., 2014). Plants infected with fungal strains and treated with NaCl showed significant increases in leaf area, fresh biomass, dry biomass, and total chlorophyll in both normal and stressed conditions (Richardson et al., 2009). When chickpea plants were grown in a saline environment, endophytes increased chlorophyll and total biomass. In contrast, lipid peroxidation and reactive oxygen species (ROS) were reduced under the same conditions (Abdallah et al., 2018). According to Chung et al. [2019], the decline in morphological parameters could be related to ion toxicity and osmotic stress caused by salinity exposure. In the case of endophytic fungus, Pandya et al. [2018] found that when *Aspergillus sp.* was applied to wheat and chickpea plants, the lengths of the roots and shoots of the plants increased. Furthermore, Asaf et al. [2018] found that when the endophytic fungus *Aspergillus flavus* CHS1 was applied to soybean plants under salinity stress or normal conditions, plant length and fresh and dry weight were dramatically increased.

Photosynthesis is directly related to stomatal conductance, chlorophyll concentrations, transpiration, and water potential, and photosynthesis is indirectly slowed by plant salinity. The loss of stomatal conductance due to water imbalance under salt stress can reduce leaf photosynthesis [Chandrasekaran et al., 2019]. Under the same experiment, the connection of *Aspergillus japonicas* with okra under both saline and normal conditions caused significant changes in chlorophyll a, chlorophyll b and total chlorophyll, chlorophyll a/b ratio, and

carotenoids (Figure 2). Applying plant-growth-promoting fungi increased chlorophyll concentrations in candy leaf (*Stevia rebaudiana*) plants (Vafadar et al., 2014). After being treated with the plant-growth-promoting fungus *Trichoderma longibrachiatum* T6, the contents of chlorophyll a, b, and total chlorophyll in wheat plants stressed with NaCl were not only boosted but also achieved a content similar to that of the control [Zhang et al., 2016]. Proline is an osmoprotectant, membrane stabilizer, and ROS scavenger in stressed cells (Dong et al., 2015). Under the same study, the connection of *Aspergillus japonicas* with okra under both saline and normal conditions caused a significant change in proline (Figure 3). Proline content acts as a dynamic component to measure salt repercussions, and AMF inoculation on plants demonstrates varied efficacies in treating salinity stress among mycorrhizal plants (Echeverria et al., 2013).

Carbohydrates are the most abundant organic compounds in nature. Carbohydrate storage sinks are typically modeled as passive reservoirs that are only filled when assimilate supply exceeds utilization sink needs. Under the same project, the connection of *Aspergillus japonicus* with okra under both saline and normal conditions resulted in a significant change in total carbohydrates, proteins, and lipids (Figure 3). Furthermore, wheat plants exposed to salt stress and colonized by an endophytic fungus (*Alternaria chlamydospora, Fusarium equiseti, Chaetomium coarctatum,* and *Fusarium graminearum*) produce more sugar than uninoculated plants [Bouzouina, 2020]. Li et al., 2017] confirmed that *Aspergillus aculeatus* could ameliorate the negative effects of specific environmental conditions, consequently boosting plant development by modifying sugar synthesis, degradation, and storage to improve salt resistance. Furthermore, Robert-Seilaniantz et al. [2007] suggested that plant-associated endophytic fungus can enhance sugar content as an osmoprotectant under salt-stress circumstances. Sugar levels in mycorrhizal plants are high, which suggests they play a role in salinity tolerance [Manchanda and Garg, 2011]. It's worth noting that inoculating soybean and sunflower plants with the endophytic fungus *Aspergillus japonicus* boosted soluble protein content as compared to untreated plants under normal and stress conditions [Hamayun et al., 2018]. Under abiotic stress circumstances, Ismail et al., (2018) found increased total lipids in *A. japonicus*-endophyte linked soybean. Baltruschat et al., (2008) found that leaves of *P. indica* injected with endophyte and grew under salt stress had a high proportion of fatty-acid lipids.

Compounds with one or more aromatic rings and one or more hydroxyl groups are known as phenolics. They are the most numerous secondary metabolites in plants and are widely spread across the plant kingdom. Under the same study, the connection of *Aspergillus japonicas* with okra under both saline and normal conditions resulted in a significant change in total phenols, flavonoids, and tannins (Figure 4). In addition, the presence of flavonoids/phenols allows the endophyte *M. caribbica* to scavenge ROS that may be produced under NaCl stress [Ismail et al., 2018]. The findings revealed that when the host plant was stressed, *M. caribbica* modified the host plant system to detoxify the harmful ROS rather than allowing the host plant to prevent salt intake. For example, in *Medicago sativa* [Catford et al., 2006], fungal symbiosis has been linked to quantitative changes in genistein or daidzein (or iso/flavonoids) concentration. According to Charlton et al. (2000), condensed tannin in plants has a high affinity for proteins and other nitrogen-based substances such as alkaloids. According to Paolocci et al. (2005), tannin is critical in plant tolerance formation when plants interact with microbes (mutualistic or pathogenic) under normal and stressful conditions.

In response to biotic and abiotic stressors, plants release reactive oxygen species, such as hydrogen peroxide. Hydrogen peroxide is engaged in numerous signal transduction pathways that lead to the proliferation of other defenses. Catalase activity was examined in several cob tissues during maize ear development because it contributes to maintaining reactive oxygen homeostasis during biotic and abiotic stress. Plant resistance mechanisms may have been activated, resulting in an increase in antioxidant enzyme activity (Ibrahim et al., 2018).

Under the same experiment, the association of *Aspergillus japonicas* with okra under both saline and normal conditions caused a significant change in lipid peroxidation, catalase, and ascorbate peroxidase activity (Figure 5). As a systemic resistance mechanism against salt stress, plants inoculated with AMF show decreased lipid peroxidation and increased antioxidant enzyme activities in common beans [Abd Allah et al., 2015]. PGPRs have also been shown to relieve salt stress in plants via a variety of methods, including fast activation of conserved salt stress-responsive signaling pathways [Adesemoye, 2009]. In comparison to control plants, endophyte and stress-treated plants had considerably decreased CAT, POD, and PPO activity. These enzymes assist plants in removing H2O2 from mitochondria and microbodies and regulate stress responses (Waqas et al., 2012). It has previously been hypothesised that an increase in enzymatic activities causes plant growth to slow [Al-Ghandi, 2009]. In mycorrhizal-stressed plants, the activities of enzymes involved in the detoxification of O_2 - radicals and H_2O_2, such as superoxide dismutase, catalase, and peroxidase, as well as enzymes that are important components of the ascorbate glutathione pathway responsible for the removal of H_2O_2, such as glutathione reductase and ascorbate peroxidase, increased significantly (Garg and Manchanda, 2008).

In plants under abiotic stress, IAA (exogenous or endogenous) can certainly control numerous developmental processes [Verma et al., 2016]. Under the same project, the connection of *Aspergillus japonicas* with okra under both saline and normal conditions resulted in a significant change in the endogenous IAA level (Figure 5). However, IAA is required for plant growth and development because of its role in root, axillary bud, and flower development. However, a recent study found that *S. indica* hyphae contain significant amounts of IAM and that both IAM and IAA levels rise during the colonization of *Brassica campestris* roots (Hua et al., 2017). In any case, it appears that IAA levels in host plants momentarily increase during the early stages of the interaction before quickly returning to non-colonized root levels (Hilbert et al., 2012).

High salt concentrations increase the amount of Na^+ and Cl^- in plants while decreasing the amount of other cations like K^+ and Ca^{2+}, resulting in mineral nutritional imbalance (Zhu and Gong, 2014). Maintaining ion homeostasis is one of the adaptation methods that tolerant plants utilize to cope with salt stress under salty circumstances. These techniques may aid the plant in avoiding the potentially hazardous effects of ions such as Na^+ and Cl^-, which cause various sorts of damage to lipids proteins, and nucleic acids (Rizwan et al., 2015). Plant metabolism, photosynthesis, osmosis (water transfer in and out of plant cells), and ionic balancing within the cell all require modest amounts of chloride. The activity of Na+ and K+ transporters and/or channels is required to achieve this homeostatic balance. The connection of *Aspergillus japonicas* with okra in both saline and normal settings caused a significant change in the sodium and chloride concentrations and Na/K and Na/Ca ratio under the same project (Figure 6). Increased phosphorus utilization efficiency, according to the authors, could have improved ion (Na^+ and K^+) uptake and allocation. Wheat plants' ability to adjust the reactive oxygen scavenging system when dealing with salinity stress has also been found to be improved by arbuscular mycorrhizal fungus (Talaat and Shawky, 2011). Song et al. (2015) also found that the lower Na^+: K^+ ratios in endophyte-colonized plants reduced harmful ion levels and the osmotic influence on plants under salt stress. Another study found that barley plants inoculated with endophytic fungus had a lower Na^+: K^+ ratio than uninoculated plants, suggesting that this ratio can be used to predict the severity of salt stress or to screen plant genotypes for high Na^+ tolerance (Ghabooli 2014). Chunthaburee et al. (2016) showed that the K/Na ratio in rice shoots was negatively correlated to the standard salinity evaluation score, and rice yield is expected to decrease with the increase in salinity evaluation score. These nutrients' concentrations and ratios (e.g., K/Na and Ca/Na) are widely used as screening parameters in ranking varieties for their tolerance to salt toxicity. These parameters are reliable

and useful in screening varieties for salt-stress tolerance. Increasing numbers of salt-tolerant transgenic plants have been generated with overexpression of vacuolar Na+ /H+ antiporter proteins mediating lower concentrations of Na and higher ratios of K/Na in cytosol (He et al., 2005)

Plant growth-promoting microorganisms, which include plant growth-promoting bacteria (PGPB), rhizobia, and arbuscular mycorrhizal fungi (AMF), are microbes that live in or around free-living soils, the rhizosphere/rhizoplane (e.g., rhizobacteria and ectomycorrhizal fungi), or the tissue interior (e.g., endophytic bacteria, end (Ma et al., 2011). The connection of *Aspergillus japonicas* with okra in both saline and normal settings caused significant changes in the potassium, calcium, magnesium, phosphorus, and nitrogen concentrations under the same project (Figure 6). Endophytic colonization improves plant development in saline circumstances by altering the expression levels of the main Na^+ and K^+ ion channels, which aids in establishing a balanced Na+ and K+ ion homeostasis under salt stress conditions (Abdelaziz et al. 2017). *Bacillus, Pseudomonas, Achromobacter, Alcaligenes, Brevibacterium, Serratia, Xanthomonas,* and *Rhizobium* (*Bacillus, Pseudomonas, Achromobacter, Alcaligenes, Brevibacterium, Serratia, Xanthomonas, and Rhizobium*) are phosphate-solubilizing halotolerant PGPRs (By secreting low molecular weight organic acids such as gluconic acid, citric acid, succinic acid, propionic acid, and lactic acid, these microbes can hydrolyze inaccessible phosphorus forms into absorbable forms by diverse methods such as chelation, ion exchange, and acidification (Saghafi *et al.,* 2018). In general, direct mechanisms include nutrient acquisition (e.g., nitrogen fixation, siderophore sequestration, and potassium and phosphate solubilization; Kohler et al., 2009), phytohormone synthesis (e.g., auxin, cytokinin, ABA, and GA; Ma et al., 2019a), exopolysaccharides (EPS; Naseem et al., 2018), volatile or non-volatile compounds, as well as induction of ACC deaminase (Forni et al., 2017). AMF endophyte association with different crops under salt stress was researched by different researchers [olive (*Olea europaea* L.) was studied by Porras-Soriano et al., (2009), and wheat (*Triticum aestivum* L.) was studied by Talaat and Shawky, (2014)]. According to the researchers, fungal associations promote plant nutrient uptake, tolerance, and growth.

CONCLUSION

In the present study, we analyze the growth of okra under salt stress, and the results negatively impact its growth, biochemistry, and different ion concentrations. The current investigation's results further revealed that the association of *Aspergillus japonicus* with the studied plant causes a positive effect, and improvement was observed in the plant's studied parameters under normal and salinity stress environments. Thus, *Aspergillus japonicus* is regarded as a promising candidate for developing a biofertilizer formulation for use in agricultural crop production under normal and saline conditions.

Data availability

All data is available in the manuscript.

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Competing interests

The authors declare no competing interests.

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