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Physiological responses of humic acid-treated rice (*Oryza sativa* L.) plants at germination under salinity and early seedling growth

Mohamed E. Abu-Ria[1*], Samy A. Abo-Hamed[1], Farag Ibraheem[1,2], and Wafaa M. Shukry[1]

[1]*Botany Department, Faculty of Science, Mansoura University, Egypt, E-mail: mohamedshokr@mans.edu.eg (corresponding author*), Sabohamed@mans.edu.eg, and wafaa_12@yahoo.com*

[2]*Biology and Chemistry Department, Al-Qunfodah University College, Umm Al-Qura University, Al Qunfodah 21912, Saudi Arabia, E-mail: faragKeyba@yahoo.com*

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Abstract: Salt sensitivity is a critical constraint for growth and productivity of rice, which is generally categorized as a typical glycophyte. In the current study, the effects of salt stress, induced by seawater at concentrations of 5%, 10%, and 12.5%, which are equivalent to EC values of (3.40, 6.77, and 8.00 mS/cm), respectively, on germination and seedling growth of two Egyptian rice cultivars (Giza 177 and Giza 179) were investigated. Also, potential of seed priming with humic acid as a stress-alleviating and growth-promoting agent at appropriate concentrations (40 mg/L for Giza 177 and 100 mg/L for Giza 179) in the two rice cultivars was tested. The results indicated that all seedling germination and growth-related traits such as plumule and radicle length and seedling weight were significantly decreased in response to increasing levels of salt stress. The tested cultivars exhibited interesting and differential responses to salt stress in absence and presence of humic acid. However, humic acid significantly improved the growth responses of the two cultivars via increasing α-amylase activity, photosynthetic pigments, total carbohydrates, and total soluble proteins. These results highlight the potential role of humic acid in improving rice germination and seedling growth in salt-affected lands.

Key words: *Oryza sativa*, seed priming, humic acid**.**

Introduction

Rice (*Oryza sativa* L.) is one of the most important cereal crops that is grown in many parts of the world and is used as the principal food for more than 50% of the world's population [1]. Moreover, rice provides 50–80% of daily calories with high nutritional value of carbohydrate, fat, protein, vitamins, and minerals. In addition to nutritional components, there are many bioactive compounds called phytochemicals have anticancer, antioxidant, antiinflammation, and antidiabetic activities [2],[3].

Salinity is a major constraint for rice production worldwide and 50% of irrigated lands are expected to suffer from salt stress by 2050 [4]. In fact, rice is generally categorized as a typical glycophyte, and its growth and productivity are highly sensitive to salinity stress. The magnitude of rice sensitivity to salinity depends on stages of rice's plant life cycle, rice cultivars, and experimental conditions under investigation [5]. Contrasting results about the sensitivity of various rice cultivars during different growth stages have been reported. For example, many reports have shown that rice is extremely sensitive to salinity during germination and young seedlings [6]. Recently, rice was very sensitive to salinity during seedling and reproductive stages although it showed a salt tolerance behavior during germination [7].

The physiological and biochemical processes that are regulated by various endogenous factors induced the embryo activation, a critical phase in the life cycle of rice plant and seedling establishment [8], [9]. During rice grain germination, the stored starch in the endosperm is hydrolyzed by α-amylase into small organic molecules and mobilized to the embryo to provide energy. During this stage, genes involved in the *de novo* synthesis of α-amylase and other hydrolases are induced by bioactive gibberellins (GAs) synthesis in the embryo [10]

Salinity stress reduces seed germination [11]**.** Salt stress creates ionic, osmotic, and secondary stresses especially oxidative stresses which induce significant metabolic alterations, decreases water and nutrient uptake, and subsequently reduces plant growth and development [12],[13]. During seed germination, the above salinity-induced stresses induce physiological and biochemical changes that significantly reduces percentage of germination, rate of germination, and various seedling parameters [14].

Seed priming is one of the important techniques that increases the resistance of seeds to future exposure to abiotic stresses like salinity. It induces pre-germination metabolic processes, increases seed vigor during germination and emergence, and consequently improves germination and growth under salinity conditions **[**15]. Humic acid is one of the promising seed priming agents. It positively modulates hormonal signaling pathways and specific abiotic functional and regulatory stress-responsive genes, thus alleviating the negative impact of stressor agents **[**16]. It also increases soil buffering and acts as a natural chelator for metal ions, so it increases nutrient uptake, and plant growth under salinity [17]. According to [18], humic acid enhances plant growth and yield when it is used directly on plants at low concentrations and thus constitutes a category of plant biostimulants. It alleviates the adverse effects of salinity on plants by increasing enzymatic and non-enzymatic antioxidant defense and increasing compatible solute production [19]. It also, stimulates synthesis of membrane proteins, especially proton pumps that increase the electrochemical proton gradient across the plasma membrane [20].

Rice breeders have identified many salinity-tolerant and salinity-susceptible cultivars. However, the physiological mechanisms behind the differential responses of these cultivars to salinity stress are not fully understood. In the current study, two rice cultivars, Giza 179, and Giza 177, that are commonly cultivated under the Egyptian agricultural conditions were selected. They are highly productive under non-saline conditions. However, they are described as salinity-tolerant and salinity-susceptible cultivars, respectively. Their differential physiological responses, if any, under salinity conditions are largely unknown. The aim of the current study is to assess the effects of the different levels of soil salinity on physiological responses of the two cultivars and identify physiological traits that may stand behind their differential responses to salinity stress. In addition, the potential use of humic acid as an alleviator of salinity stress in the cultivars was also tested. Herein, the hypothesis is that the differential responses of the two cultivars to salinity may be associated with particular physiological bioindicators. Therefore, performing a comparative analysis of the physiological responses of the two cultivars would be a useful approach to gain insights into their differential responses to salinity stress. The specific objectives of this study are to (i) evaluate the growth and development of the two cultivars under increased levels of salinity in absence and presence of humic acid, (ii) assess the impact of humic acid on the levels of critical physiological and biochemical indicators such as α-amylase and carbon metabolism during germination and early seedling growth.

Materials and methods

Plant Materials

Rice (*Oryza sativa* L.) grains var. Giza 177 (G7) and Giza 179 (G9) were purchased from the Rice Research and Training Center, Sakha, Kafr El-Sheik, Egypt.

Humic acid treatments

Test the effect of humic acid on grain germination in Giza 177 and Giza 179 cultivars

Four sets of homogenous grains of each rice cultivar, 50 grains each, were surface sterilized using sodium hypochlorite (3.6%) for 15 mins and washed thoroughly with distilled water. Grains were soaked either in different concentrations of humic acid (20, 40, 100 mg/l) as recommended by [16, 21] or in distilled water as control for 48 hours at 27 ± 2°C. The grains were then germinated in plastic boxes (22 x 17 x 9 cm) lined with filter papers and maintained in an incubator at optimum conditions for rice seedling growth. The grains were supplied with an adequate amount of distilled water twice a day. Each treatment was replicated five times. Seven days post germination (DPG), ten seedlings were harvested from each cultivar/treatment and used for determination of seedling growth parameters (see below). The rest of germinated seedlings were dried in an electric oven at 80°C until constant weights (48 hours) and used for dry weight determinations. The obtained results were used as a guide to determine the optimum concentration of humic acid for both Giza 177 and Giza 179 which was used in downstream experiments.

Evaluate the role of humic acid on rice germination under salinity stress and early seedling growth

Once the optimum concentration of humic acid for both Giza 177 and Giza 179 cultivars were determined, enough grains from both cultivars were surface sterilized and washed thoroughly with distilled water as described above. The sterilized grains from both cultivars were allocated into two subsets. The first subsets from both cultivars were soaked in distilled water as controls in the dark in an incubator at 27 ± 2 ^{o}C for 48 h whereas the other two subsets were soaked in the pre-determined optimum concentration of humic acid for Giza 177 (40 mg/l) and Giza 179 (100 mg/l). After soaking, each of the water- as well as the humic acid- treated grains from each rice cultivar were further allocated into two subsets and used for assessing the effects of humic acid on germination under controlled conditions as well as its effects on early seedling growth in pot experiments.

For the germination experiment, the water- and humic acid-treated grains of the first subset of each rice cultivar were transferred to plastic boxes (22 x 17 x 9 cm) lined with sterile Whatman No. 1 filter paper moistened with different seawater concentrations (0%, 5%, 10%, and 12.5%), which are equivalent to EC values of (0.55, 3.40, 6.77, and 8.00 mS/cm), respectively. The boxes were maintained under optimal conditions for rice seedling growth. Each treatment was replicated three times. The seedlings growth parameters were monitored. In addition, α-amylase activity in grains were determined at 0, 3, and 7 days after soaking (DAS).

For pot experiments, the water- and humic acid-treated grains of the second subset of each rice cultivar were grown in 20 pots (5 pot for each cultivar/treatment, 30 cm in diameter, each pot contained 7 kg of homogenous clay soil). The rice plants were maintained in greenhouse under natural growth conditions for 28 days then photosynthetic pigments, total carbohydrates, and total soluble proteins were monitored.

Seedling parameters

Plumule and radicle lengths (Cm) and seedling fresh and dry weights (gm) of ten 7 DAS seedlings were calculated according to [22].

Estimation of Alpha amylase activity

The alpha amylase activity was determined by measuring the amount of maltose and other reducing sugars released from starch hydrolysis using 3,5-dinitrosalysalic acid (DNSA) according to the method adopted by [23].

Estimation of photosynthetic pigment

Photosynthetic pigments were estimated using the spectrophotometric method as recommended by [24]. Briefly**,** 0.1 gm of fresh leaf was incubated with 7 ml of dimethyl sulfoxide (DMSO) in a water bath at 60 °C for 30 minutes, then cooled at room temperature. The contents were filtered, and the filtrate was made up to 10 mL using DMSO. A Shimadzu UV-160A spectrophotometer was used to measure the absorbance at 480, 644, and 663 nm. The concentrations of the pigment fractions were calculated according to [25] equations and expressed as mg g^{-1} FW.

Estimation of total carbohydrates

Total carbohydrates in control and humic acid-treated seedlings were extracted by mixing 0.1 g of dry tissue with 5 ml of HCl (2.5 N) and the mixture was boiled in a water bath for three hours. The mixture was cooled down and neutralized with Na_2CO_3. Total carbohydrates were determined by mixing 1 ml of the plant tissue extract with 4 ml of freshly prepared anthrone. The mixture was then boiled in a water bath for 8 minutes, cooled down, and the intensity of the produced color was recorded at

630 nm using a spectrophotometer (JENWAY ST15OSA-Model 7315, Bibby Scientific Ltd, Stone Staffs, UK) [26]. Total carbohydrates were determined using glucose standard curve and expressed as mg sugar/g DWT.

Estimation of total soluble proteins

Soluble protein was extracted in Tris-HCl buffer according to [27]. Soluble proteins in plant extracts were determined spectrophotometrically by mixing 980 µl of Bradford reagent with 20 µl of the extract and shaking vigorously. The absorbance of the developed color was measured at 595 nm using a Shimadzu spectrophotometer model UV-160A [28]. The concentration of total soluble protein was calculated using bovine serum albumin standard curve and expressed as mg protein/ g FWT.

Statistical analysis

The obtained data were statistically analyzed using one-way analysis of variance (ANOVA) and Pearson correlation coefficient was calculated using COSTAT Version 6.3 (Cohort software, Berkeley, California, USA). The data were presented as a mean ± SE. The Post Duncan's test was performed at $P \leq 0.05$. All graphs and heatmaps were conducted by Microsoft Office Excel software.

Results and discussion

Humic acid as a biostimulator

Humic acid has an auxin like effect on plants and this is the main reason for its beneficial effect on plants [29]. In the current study, increasing humic acid concentration increased all the growth parameters in both cultivars. The maximum humic acid-induced increase in growth was obtained at 40 mg/l for Giza 177 and 100 mg/l for Giza 179, respectively (Tables 1). These optimum concentrations of humic acid induced the best overall growth in Giza177 and Giza179, respectively, as they recorded significant increases in all growth parameters compared with other treatments. Our results are consistent with those of [21] who reported that rice grain priming with humic acid promotes grain germination and seedling growth. Humic acid is known to improve plant growth and yield by increasing nutrient availability, water holding capacity, and hormonal activity [30]. [31] reported that priming with potassium humate improved rice grain germination and seedling growth.

Table (1): Effects of different concentrations of humic acid on germination of Giza 177 and Giza 179 cultivars. Data are the means ± standard error of ten biological replicates. Records followed by similar letters indicate non-significant differences at 5% probability level.

parameters/ treatments	G7 Plumule Length (Cm)	G7 Radicle Length (Cm)	G7 Seedling Length (Cm)	G7 Seedling fresh mass (g)	G7 Seedling dry mass (g)	G9 Plume Length (Cm)	G9 Radicle Length (Cm)	G9 Seedlg Length (Cm)	G9 Seedling fresh mass (g)	G9 Seedling dry mass (g)
C	9.50^{b} ±0.04	13.57^{b} ±0.44	23.07^{b} ±0.45	0.067^{b} ±0.000	0.006^{b} ±0.000	9.14^{c} ±0.14	12.11^{c} ±0.43	21.25^{c} ±0.40	0.058^{c} ±0.001	0.007^{c} ±0.000
40mg/l	10.56^{a} ±0.05	15.49^{a} ±0.57	26.05^{a} ±0.58	0.077^{a} ±0.000	0.008^{a} ±0.000	9.40^{bc} ±0.10	14.04^{b} ±0.75	23.44^{b} ±0.77	0.064^{b} ±0.001	0.007^{bc} ±0.000
80mg/l	9.71^{b} ±0.16	13.98^{b} ±0.36	23.69^{b} ±0.36	0.065^{b} ±0.001	0.006^{b} ±0.000	9.65^{b} ±0.12	14.54^{b} ±0.42	24.19^{b} ±0.38	0.068^{b} ±0.001	0.008^{b} ±0.000
100mg/l	9.45^{b} ±0.08	11.84^{c} ±0.46	21.29^{c} ±0.48	0.055^{c} ±0.000	0.005^{c} ±0.000	10.37^{a} ±0.08	16.49^{a} ±0.35	26.86^{a} ±0.34	0.077^{a} ±0.001	0.009^{a} ±0.000

Humic acid alleviates salinity-induced inhibition of rice grain germination.

Plumule and radicle growth retardation are the most important responses for salt stress in rice [14]. The applied levels of seawater (5%, 10%, and 12.5% seawater) induced a reduction in germination and post-germination growth of both cultivars in a dose-dependent manner (Table 2). The severe salt stress (12.5%) showed a maximum reduction of 31.42%, 36.20%, and 32.14% in Giza 179 seedling length, fresh and dry weight, respectively, compared to 38.16%, 29.13%, and 36.06% in Giza 177 seedling. Our results agreed with [32] who reported that rice grain germination and seedling growth were decreased by salinity. [33] observed that shoot and root length, fresh and dry weight decreased with increasing salt stress. This confirms earlier report which suggested that salt stress reduced shoot and root fresh and dry weigh of thirty rice varieties [34]. The mechanism of inhibition of seed germination and seedling growth by salinity was due to insufficient water absorption, toxic ion effect on the embryo and limiting hydrolysis of seed storage food through water imbalance and enzyme activity reduction [35], [36].

Priming both cultivars at their corresponding optimum humic acid concentrations improved the germination and post germination growth under saline and non-saline conditions. These results provide strong evidence regarding the positive effects of humic acid on the growth performance of rice plants under saline and non-saline conditions. This is consistent with [37] who showed that soaking rice grain in humic acid improved grain germination and seedling growth under salinity stress. Also, [38] showed that humic acid reduced the negative impact of salinity on germination and growth of *Medicago sativa* L. Heatmap confirmed the progressive decrease in all growth parameters with increasing salinity stress and showed the beneficial effect of humic acid under saline and non-saline conditions (Figure 2).

Humic acid maintains α-amylase activity during germination under salinity

Our results demonstrated that α-amylase activity increased over the initial value (0 DAS) at 3 DAS and then declined at 7 DAS in humic acid-treated and untreated Giza 179 and Giza 177 rice cultivars under saline and non-saline conditions. Our results agreed with those of [39] who showed that α-amylase activity increased significantly in all tested cereals on germination then declined at 8 days of germination. Pearson

correlation analysis showed a significant positive correlation of α-amylase activity with seedling length, fresh and dry weight in both cultivars, reflecting the importance of α-amylase in seed germination and early seedling growth (Tables 3). Compared to saline untreated control in Giza 179 cultivar, increasing salinity stress levels caused a progressive decrease in α-amylase activity by 3.83%, 7.86%, and 9.78% at 3DAS and by 1.96%, 6.03%, and 10.70% at 7DAS. Similarly, in the Giza 177 cultivar, there was a progressive decrease by 12.54%, 15.53%, and 17.83% at 3DAS and by 5.72%, 10.07%, and 15.10% at 7DAS (Figure 1). [40] found that salinity inhibits rice grain germination by decreasing α-amylase activity. Furthermore, they showed a positive relationship between α-amylase activity and rice grain germination rate which is confirmed in the current study (Tables 3). This agrees with [41] who found that rice grain germination under saline-alkaline stress was greatly affected as a result of inhibition of α-amylase activity. Salinity stress, according to [42],[43] inhibits α-amylase activity, which is the primary cause of poor hydrolysis of stored food and its subsequent translocation to the embryo.

Grain pre-soaking with humic acid increased α-amylase activity in both cultivars under saline and non-saline conditions. These results are in harmony with those obtained by [44] who found that priming maize hybrid with humic acid improves the activity of α-amylase. [45] reported that the principle of various seed priming techniques is to minimize the period between seed sowing and seedling establishment. This was confirmed by our findings in Giza 179, where α-amylase activity was higher in humic acid-treated grains than in untreated ones after soaking and 3 days, but lower after 7 days (Figure 1). Such a response could imply that 3DAS stage represents the most active metabolic interconversion, requiring high GA3 content and subsequent active α-amylase efficient induction, and that such requirements fade at 7DAS. Thus, humic acid succeeded in decreasing the time between grain germination and seedling establishment. Pearson correlation analysis confirmed our results as there’s a significant positive correlation between α-amylase activity at 3DAS and seedling length, fresh and dry weight. However, there's a non-significant positive correlation in α-amylase activity at 7DAS with seedling parameters (Table 3). In contrast, the activity of α-amylase in Giza 177 were higher in treated grains than in untreated ones at all time-points (Figure 1). From the results in figure (2), we conclude that priming rice grains with an appropriate concentration of humic acid increases α-amylase activity and seedling growth parameters under saline and non-saline conditions.

Table (2): Effect of different levels of seawater (Control, 5%, 10%, 12.5%) on growth parameters of Giza 177 and Giza 179 rice cultivars in presence or absence of optimum concentration of humic acid (40 mg/l for Giza 177 and 100 mg/l for Giza 179). Data are the means ± standard error of six biological replicates. Records followed by similar letters indicate non-significant differences at the 5% probability level.

arameters Treatments	G7 Plumue Length (Cm)	G7 Radice Length (Cm)	G7 Seedling Length (Cm)	G7 Seedling fresh mass (g)	G7 Seedling dry mass (g)	G9 Plumule Length (Cm)	G9 Radicle Length (Cm)	G9 Seedling Length (Cm)	G9 Seedling fresh mass (g)	G9 Seedling dry mass (g)
Control	10.33^{bc} ±0.15	11.28^{b} ±0.31	21.62^{b} ±0.29	0.076^{b} ±0.002	0.010^{ab} ±0.000	10.00^{b} ±0.14	10.30^{b} ±0.61	20.30^{b} ±0.64	0.068^{b} ±0.001	0.009^{ab} ±0.000
5%	9.63^{de} ±0.08	7.55^{d} ±0.19	17.18^{e} ±0.16	0.069^{c} ±0.001	0.009^{bc} ±0.000	9.02^{c} ±0.24	8.93^{bc} ±0.13	17.95^{c} ±0.34	0.069^{b} ±0.002	0.009^{ab} ±0.000
10%	9.23^{ef} ±0.12	6.52^{e} ±0.42	15.75^{f} ±0.39	0.060^{d} ±0.001	0.008^{cd} ±0.000	8.20^{d} ±0.24	7.17^{cd} ±0.45	15.37^{de} ±0.67	0.056^{c} ±0.001	0.007^{c} ±0.000
12.5%	7.72^{g} ±0.28	5.65^{e} ±0.20	13.37^{g} ±0.36	0.054^{e} ±0.000	0.006^{e} ±0.000	7.62^{d} ±0.16	6.30^{d} ±0.61	13.92^{e} ±0.64	0.043^{d} ±0.002	0.006^{c} ±0.000
Humic	11.90^{a} ±0.20	13.32^{a} ±0.78	25.27^{a} ±0.86	0.087^{a} ±0.002	0.011^{a} ±0.000	10.80^{a} ±0.38	12.17^{a} ±0.76	22.97^{a} ±0.89	0.077^{a} ±0.001	0.010^{a} ±0.000
H+5%	10.50^{b} ±0.22	9.57^{c} ±0.07	20.07^{c} ±0.24	0.076^{b} ±0.001	0.010^{a} ±0.000	9.93^{b} ±0.21	10.60^{ab} ±0.74	20.53^{b} ±0.87	0.078^{a} ±0.004	0.010^{a} ±0.000
H+10%	9.85^{cd} ±0.17	8.91^{c} ±0.14	18.76^{d} ±0.24	0.071^{c} ±0.000	0.009^{bc} ±0.000	9.02^{c} ±0.45	8.43^{c} ±0.56	17.45^{c} ±0.56	0.067^{b} ±0.000	0.008^{b} ±0.000
H+12.5%	8.77^{f} ±0.10	6.20^{e} ±0.09	14.97^{f} ±0.14	0.062^{d} ±0.000	0.008^{d} ±0.000	8.02^{d} ±0.14	8.00^{cd} ±0.67	16.02^{cd} ±0.70	0.052^{c} ±0.002	0.006^{c} ±0.000

Table (3): Pearson correlation analysis of 4 measured traits for (A) Giza 177 and (B)Giza 179 cultivar. ** and *** indicates significant meanwhile, ns indicates non-significant differences at the 5% probability level.

Parameters / verities		Seedling length	Seedling fresh mass	Seedling dry mass	α-amylase 3DAS
Giza 177	Seedling fresh mass	0.983***			
	Seedling dry mass	0.953***	0.979***		
	α-amylase 3DAS	0.981***	0.949***	0.904**	
	α-amylase 7DAS	0.956***	0.966***	0.974***	0.919**
Giza 179	Seedling fresh mass	0.916**			
	Seedling dry mass	0.918**	0.980***		
	α-amylase 3DAS	0.984***	0.889**	0.870**	
	α-amylase 7DAS	0.190 ns	0.178 ns	0.320 ns	0.025 ns

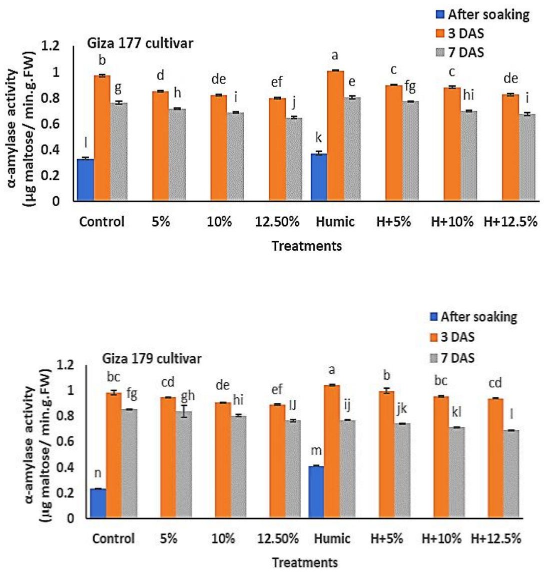
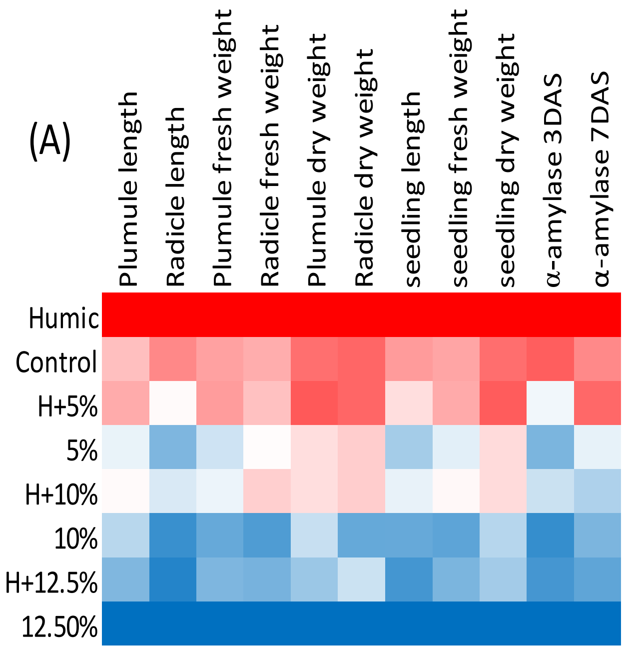


Fig. (1): Effect of salinity stress alone or in combination with the optimum concentrations of humic acid on α-amylase activity of Giza 177 and Giza 179 rice seedlings. α-amylase activity was detected at 0, 3, and 7 days after soaking (DAS) with three biological replicates. Vertical bars represent the standard error (±S.E.). Different bar letters show significant

Humic acid increases photosynthetic pigments, total carbohydrates, and total soluble proteins in 28-days-old seedling.

Careful examination of figure (3) showed that, as compared to control seedlings, priming both cultivars with their corresponding optimum concentration of humic acid induced accumulation of various photosynthetic pigments (Chl a, Chl b, Carotenoids, and total pigments) by 66.55%, 75.76%, 58.30%, and 65.19%, respectively in Giza 179 and by 58.33%, 58.68%, 70.42%, and 61.67%, respectively in Giza 177. Our results are supported by [46] who reported that humic acid increases the photosynthetic pigments (Chl a, Chl b, and carotenoids) in treated rice plants. [47] found that foliar application of potassium humate resulted in enhancements in chlorophyll content in rice. The enhancements in chlorophyll and carotenoid biosynthesis were attributed to the increase of cell membrane permeability and stimulating nutritional element absorption. [48].

The optimum concentrations of humic acid induced significant changes in total carbohydrates and soluble proteins in 28- days-old seedlings of both rice cultivars. Compared to control seedlings, humic acid induced increases of 10.80% and 8.17% in total carbohydrates and total soluble proteins in Giza 179 corresponding to 22.02% and 16.94% in Giza 177, respectively. These results are consistent with those of [21] who reported that starch content, total soluble sugars, and total soluble proteins in rice plant were significantly improved by seed priming with humic acid. [49] also, showed that humic acid significantly increased total carbohydrates and total soluble proteins in the leaves of wheat (*Triticum durum*) plants. The role of humic acid in increasing total carbohydrates and protein biosynthesis comes from its role in increasing photosynthesis, mineral nutrient uptake and transcriptional activation [29]. This agreed with [50] who showed that humic substances have greater involvement in RNA and protein synthesis.differences at the 5% probability level



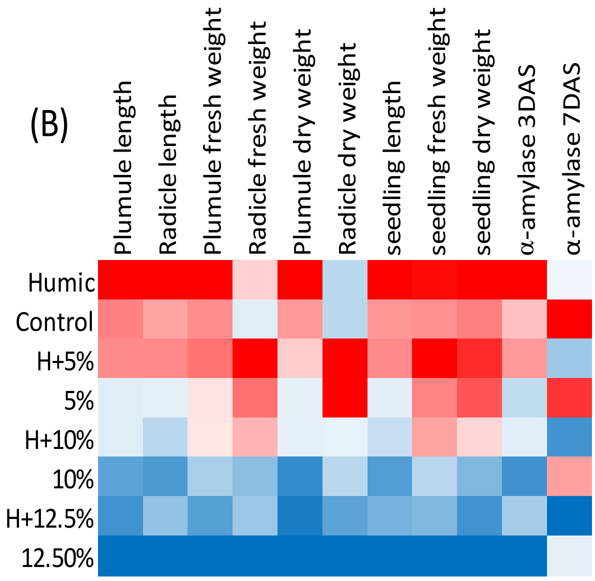
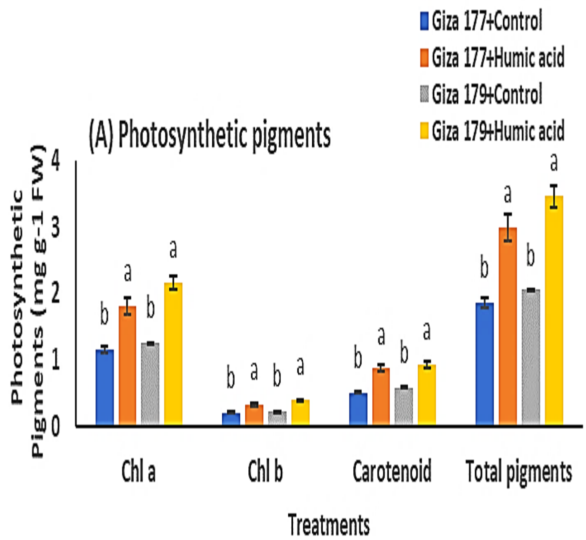
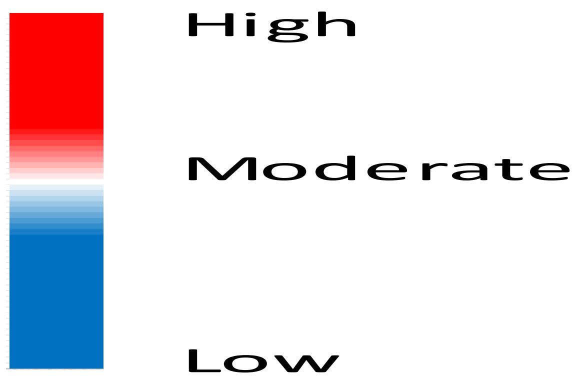
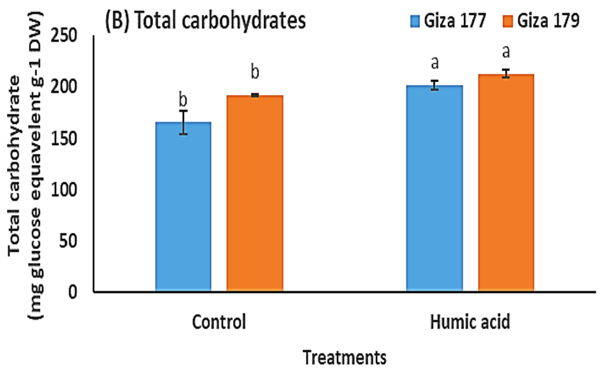


Fig. (2): Heatmap of the effect of salinity stress alone or in combination with the optimum concentrations of humic acid on 11 measured traits for (A) Giza 177 and (B) Giza 179 cultivar during germination. Color bars indicate high (red) and low (blue) levels of the investigated traits.







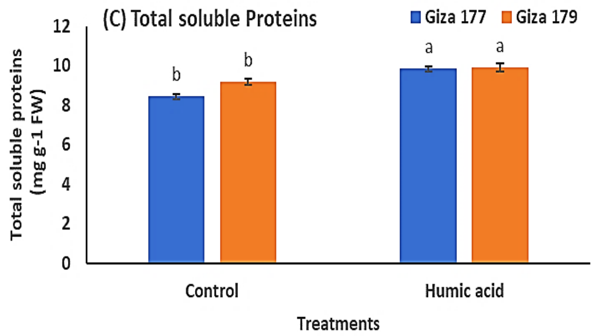


Fig. (3): Effect of the optimum concentrations of humic acid on (A) Photosynthetic pigments (B) Total carbohydrates (C) Total soluble proteins of Giza 179 and Giza 177 rice seedlings over a period of 28 days. Vertical bars represent the standard error (±S.E.). Different bar letters show significant differences at the 5% probability level among treatments separately.

Conclusions

The results of germination and seedling growth parameters of both Giza 177 and Giza 179 rice cultivars proved that 40 mg/L was the optimum concentration for growth of Giza 177, whereas 100 mg/L was the optimum concentration for growth of the Giza 179 cultivar. From the results of this investigation, it can be concluded that humic acid at lower concentrations has a stimulating effect on the germination process and seedling growth of rice plants. In general, soaking rice grains in the optimum concentrations of humic acid improves early seedling growth by increasing α-amylase activity, photosynthetic pigments, total carbohydrates, and total soluble proteins. The promotive effects of humic acid on growth under saline and non-saline condition, along with its availability and easiness of its application highlights the economic significance of humic acid in rice production in non-saline and salt-affected lands.

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