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The Integration Efficacy between Beneficial Bacteria and Compost Tea on Soil Biological Activities, Growth and Yield of Rice Under Drought Stress Conditions



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ABSTRACT

During the two growing seasons 2020 and 2021, A field experiment was carried out at Sakha Agricultural Research Station, Kafr El- Sheikh Governorate, Egypt to assess the impact of individual or combination treatments (T₁, T₂, T₃ and T₄) of Plant growth promoting rhizobacteria (PGPR) and foliar spray with compost tea (CT) for enhancing vegetative growth, physiological characteristics, nutrient content and soil biological activities as well as yield components of rice plant (Sakha 105) grown under various intervals of irrigation water regime (I₁=irrigation every 3 days, I₂=irrigation every 6 days, and I₃=irrigation every 9 days). Results showed that all different irrigation water intervals accompanied with PGPR + CT treatment gave the highest values of vegetative growth and physiological characteristics during the two seasons. At the flowering stage, leaves K⁺, K⁺/Na⁺ (%), Zn, Mn, Fe and Cu (mg Kg⁻¹) showed a significant increase when rice plants exposed to I₃ T₄ treatment, whereas there was a decrease in Na⁺ (%). Similar trend was observed in the microbial communities in the rhizosphere of rice plants. Soil enzymes were noted to increase with the combination treatment (PGPR + CT), at I₃ treatment compared to the other treatments, respectively. For yield parameters, irrigation treatments followed the order of I₁ > I₂ > I₃. However, it followed as T₄ > T₃ > T₂ > T₁ under soil and foliar spray treatments. Thus, combination treatment under different irrigation water intervals is an efficient way to partially get rid of the effects of drought on growth dynamics of rice.

Keywords: Drought stress; Rice; PGPR; Compost tea; soil biological activities

INTRODUCTION

Water shortage is one of the main threats to the agricultural economy in Egypt during the past decade due to the increase in population, horizontal expansion and limited resources for irrigation with fresh water, resulting in a gap emerged between demand and available water, reached to approximately 13.5 billion cubic meters/year (Omar and Moussa 2016). Drought is one of the most important environmental factors that reduce the growth and productivity of many crops which due to the low value of the precipitation and the irregular distribution (Osakabe *et al.* 2014; Emami Bistgani *et al.* 2017). Based on the global climate changes, scientists predict a rise in the temperature reached 1.4 in 2050, which leads to more transpiration and evaporation losses (Agrawala *et al.* 2004; Sadok *et al.* 2021), and these climate changes negatively affect the productivity of various crops and may endanger global food security (Kirby *et al.* 2016; Raza *et al.* 2019).

In Egypt, rice (*Oryza sativa* L.) is one of the main field crops for local consumption as well as is the most important crop for farmers, because it has a high-income source, where the area harvested reached 554205 hectares (ha) with a total production of 4.89 million tons (FAOSTST 2020). Also, rice plant is affected by water stress, especially during in vegetative stage *i.e.* root length, root moisture extraction, canopy size, leaf elongation rate, transpiration rate and relative water content (RWC), and yield stage *i.e.*

spikelet number, panicle development, and grain yield (Bernier *et al.* 2007; Prasad *et al.* 2008). Therefore, it needs for irrigation water is about two - three times higher than what is needed to produce other crops, *i.e.* maize or wheat (Wang *et al.* 2017), equivalent to 2.6 m³ per 1 kg of rice (Maraseni *et al.* 2017).

Recently several experimental solutions have been applied to treat the effects of water stress in rice. One effective solution was the use of plant growth promoting rhizobacteria (Abd El-Mageed *et al.* 2022), and foliar spray with organic nutrients *i.e.* compost tea (Moridi *et al.* 2019). Beneficial microorganisms or PGPR, can play an important role in minimizing the negative effects of drought stress on plants (Vurukonda *et al.* 2016), through several mechanisms 1) phytohormones production (gibberellic acid, abscisic acid, indole-3-acetic acid and cytokinins), 2) reduce the level of ethylene by ACC deaminase in the roots, 3) induced systemic tolerance by compounds produced by bacteria, and 4) exopolysaccharides production (Carlson *et al.* 2020; Getahun *et al.* 2020; Poudel *et al.* 2021). These mechanisms can lead to increase water saving and increase crop yield productivity under deficit water stress conditions.

Compost tea is rich in humic acids, growth hormones (auxin and cytokinin), amino acids, enzymes, vitamins, nutrients (N, K, Mg, Zn, Ca, Fe and Cu) as well as beneficial microorganisms, which can enhance the growth and the productivity of different crops and increase the

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resistance against diseases (Aghamohammadi *et al.* 2016; Ibrahim 2019; Osman *et al.* 2022). Compost tea can be applied as foliar spray or root drench which due to enhance root elongation and plant growth by produce both of cytokinins (Zhang *et al.* 2014) and gibberellic acids (Pant *et al.* 2012), as well as buffer soil pH by contains of organic acids and humic substances (Morales-Corts *et al.* 2018; Van Heerden and Hardie 2020).

Pang *et al.* (2020) reported that endophytic bacteria and fungi was applied to alleviate drought stress in rice plant by increasing the activity of antioxidant enzyme, soluble sugar content and rice seedling growth. Abd El-Mageed *et al.* (2022), showed that rice plants exposed to drought stress (deficit drip irrigation) and inoculated with PGPR (*Bacillus subtilis* and *B. megatherium*), could ameliorate the deleterious effects of water stress by improving photosynthetic pigments, antioxidant enzymes, plant growth and yield. Moridi *et al.* (2019), suggested that the application of liquid organic fertilizers can be increase of shoot dry weight, shoot nutrients uptake and reduce water consumption of maize plants grown under water deficit stress (Field Capacity (FC), 80% FC and 60% FC). Also,

positive effect was observed in sugar beet plant exposed to water stress by foliar spray with with compost tea, which improved the growth dynamics, physiological process and the productivity (Osman *et al.* 2022).

The present study was designed to evaluate the impact of individual and combination treatments of PGPR and foliar spray with compost tea on enhancing vegetative growth, physiological, nutrient content and soil biological activities as well as yield of rice plant grown under different irrigation water intervals during 2020 and 2021 seasons.

MATERIALS AND METHODS

To enhance the parameters of vegetative growth, physiological, nutrient content and soil biological activities as well as yield of rice plant (*Oryza sativa* L.) grown in salt-affected soil, plant growth promoting rhizobacteria (PGPR) and foliar application of compost tea treatments were applied. To achieve this goal, field experiments were conducted at Sakha Agricultural Research Station, Kafr El-Sheikh, Egypt during 2020 and 2021 seasons. The experimental design was split-plot as shown in Figure 1.

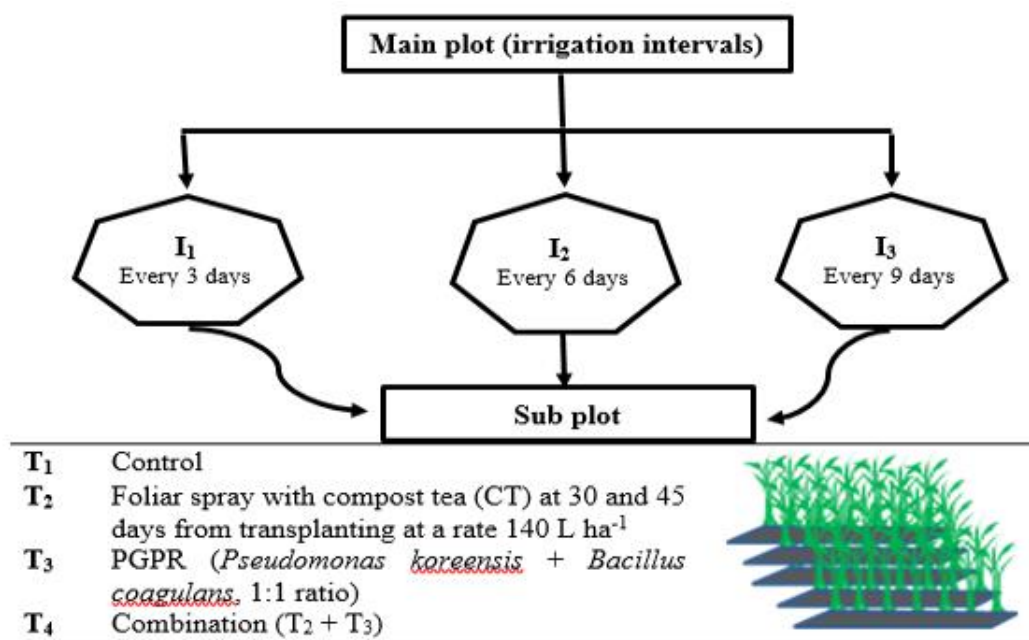


Figure 1. Experimental layout (3 x 4), 3 treatments for main plot (irrigation intervals) and 4 treatments for sub plot (foliar application and inoculation with PGPR) for field experiment during 2020 and 2021 seasons. Each treatment had 3 replicates.

Seeds of rice, *Oryza sativa* L., (Sakha 105, viability was 98%) were obtained from the Rice Research and Training Center, at Kafr El-Sheikh governorate, Egypt and then broadcast in the flooded nursery on April 15th in 2020 and April 20th in 2021 at the rate of 120 kg ha⁻¹ after soaking for one day in fresh water and incubated for another day. After 25 days, the seedlings were transferred from the nursery to transplanted at the rate 3-4 seedlings per hill in the experimental plots (5 x 9 m and 1 m apart), and to avoid lateral water movement there are 1.5 m alleys between the main-plots. Some physicochemical and biological properties of the soil used having the following characteristics: pH, 7.91 and 7.86; EC, 7.22 and 7.16 dSm⁻¹; organic matter, 1.58 and 1.67 %; particle size distribution clay, silt and sand (%), 47.50 and

47.10, 35.35 and 35.65, 17.15 and 17.35; available N, P and K (mg Kg⁻¹), 7.95 and 8.11, 6.10 and 6.36, 361.9 and 342.1, during the two growing seasons, respectively. Also, total count of microorganisms (CFU/g), were 217 x 10⁶ and 188 x 10⁶ for bacteria; 97 x 10⁴ and 106 x 10⁴ for fungi, 64 x 10⁵ and 88 x 10⁵ for actinobacteria in 2020 and 2021 season, respectively. The recommended fertilizer doses (125 kg P₂O₅ ha⁻¹ from calcium superphosphate, 15.5% was used before transplanting and 160 kg N ha⁻¹ from urea, 46.5% was used in three equal doses). Other rice agricultural practices i.e., soil preparation, weed management and pest management were followed according to the recommendations of the Ministry of Agriculture and Land Reclamation, Egypt.

Plant growth promoting rhizobacteria (PGPR) and compost tea properties

Pseudomonas koreensis MG209738 and *Bacillus coagulans* NCAIM B 1086 were prepared by inoculating King's B broth medium (King *et al.* 1954), and Nutrient Broth medium (Atlas 1997), respectively. From each culture (1 x 10⁹ CFU mL⁻¹), 300 ml was mixed with 300 g of the sterilized carrier and dispersed after 10 days from transplanting. Compost tea was used as a foliar spray at 30 and 45 days from transplanting at a rate 140 L ha⁻¹, and diluted to 460 L water ha⁻¹, which supply the plant and soil with microorganisms and mineral nutrients. Chemical and biological properties of compost tea used were pH, 6.8; EC (dS m⁻¹), 2.88; total N (ppm), 110.77; available P (ppm), 44.5; available K (ppm), 129.11; total count of bacteria (Log CFU ml⁻¹), 7.88; total count of actinobacteria (Log CFU ml⁻¹), 4.40 and total count of fungi (Log CFU ml⁻¹), 4.55. PGPRs and compost tea were obtained from Agricultural Microbiology department, Soils, Water and Environment Research Institute (SWERI), and Agricultural Research Centre (ARC), Egypt.

Morph-Physiological parameters

From each plot and during flowering stage, plants of four hills were collected randomly to estimate plant height (cm plant⁻¹), leaf area (cm²), and number of tillers plant⁻¹. For physiological characteristics, chlorophyll content (total) was determined in five leaves by using chlorophyll meter (Model-SPAD502) Minolta Camera Co. Ltd., Japan (Meier 2001).

Proline content

According to Bates *et al.* (1973), proline content (μ g-1 FW) was measured. Using a mortar and pestle, the leaf sample (0.5 g) was homogenized in sulfosalicylic acid at a rate of 5 ml (3%). Then, two ml of the extract was taken and was put them in a test tube containing 2 ml of glacial acetic acid and 2 ml the reagent (ninhydrin). In a water bath (100 °C for 1 h), the mixture was boiled, and after cooled, 4 mL of toluene was added and the absorbance was read at 520 nm in UV spectrophotometer (Jenway model 6705, UK).

Relative water content

For Relative water content (RWC), leaves were separated from plant, freshly weighed (FW), and moistened in distilled water for one day to obtain the turgid weight (TW), and at 60 °C, it was dried in the oven for 2 days to obtain the dry weight (DW). Through the following formula, the relative water content was measured (Meier 2001).

$$\text{RWC (\%)} = \frac{\text{FW} - \text{DW}}{\text{TW} - \text{DW}} \times 100$$

Determinations of K⁺, Na⁺, K⁺/Na⁺ ratio and micro-nutrients in rice Leaves

At the flowering stage, leaf sample (0.5 g) was digested in concentrated sulfuric acid and H₂O₂ (30%) on a hot plate according to the methods of (Jones *et al.* 1991). The percentage of each K⁺, Na⁺, and the ratio between them were determined by a Flame photometer (Cottenie *et al.* 1982), and by using an atomic adsorption spectrophotometer (Perkin Elmer 3300), Zn, Mn, Fe, and Cu were measured as mg Kg⁻¹ (Cottenie *et al.* 1982).

Microbial community and soil enzymes estimations

Ten g of soil samples (rhizosphere) were added to 90 ml of sterilized distilled water, thoroughly mixed, and shaken for a duration of 30 min at a speed of 150 rpm. Total

count of bacteria (log CFU 10 g⁻¹ dry soil) was estimated using soil extract agar media (Allen, 1959). After pasteurizing the soil dilutions (10⁻³), total count of *Bacillus* (log CFU 10 g⁻¹ dry soil) was evaluated by nutrient agar medium (Atlas 1997). Total count of *Pseudomonas* (log CFU 10 g⁻¹ dry soil) was estimated using King's B medium according to King *et al.* (1954).

For soil enzymes estimation, dehydrogenase activity (mg TPF g⁻¹ soil day⁻¹) of soil samples were measured spectrophotometrically by reduction of 2, 3, 5-triphenyltetrazolium chloride (TTC) to triphenyl formazon (TPF, red-color), according to Casida *et al.* (1964). Urease activity (mg NH₄⁺- N g⁻¹ soil day⁻¹) of soil samples were measured by determining the amount of ammonium produced by urea hydrolysis in soil (Pancholy and Rice 1973). Amylase activity (mg glucose g⁻¹ soil h⁻¹) of different soil treatments were determined by using starch as a substrate as described by Roberge (1978). Invertase activity (mg sucrose g⁻¹ soil h⁻¹) of soil samples were determined by using sucrose as a substrate according to the method described by Ross (1965).

Yield parameters

From the middle of each plot, 10 plants were randomly harvested to estimate 1000-grain weight, number of grains panicles⁻¹, number of panicles m² as well as grain and straw yields (t ha⁻¹).

Statistical Analysis

The results obtained were calculated using COSTAT-C Statistical Software package according to Gomez and Gomez (1984), at P < 0.05, followed by Duncan's multiple range test (Duncan 1955).

RESULTS AND DISCUSSION

Growth parameters

The results pertaining to plant height (cm plant⁻¹), leaf area (cm²), and number of tillers plant⁻¹ influenced by irrigation intervals (every 3, 6 and 9 days) are presented in Table 1, which found statistically significant (P < 0.05) at the flowering stages. Among all the treatments, significantly results of 79.63, 80.83 cm plant⁻¹, 34.61, 36.50 cm², and 21.75, 22.75 plant⁻¹ were recorded in the combination treatment (I₃T₄, irrigation every 9 days and combination of PGPR + CT) for plant height, leaf area and number of tillers during 2020 and 2021 seasons, respectively (Table 1). However, the lowest results of 63.54, 70.91 cm plant⁻¹, 22.84, 24.19 cm², and 16.43, 17.43 plant⁻¹ were recorded in the combination treatment (I₃T₁, irrigation every 9 days and control) for height plant, leaf area and number of tillers during 2020 and 2021 seasons, respectively (Table 1). The improvement in growth parameters of rice plant by PGPR and foliar spray with CT might be due to the mechanisms by which they promote plant growth and the ability to produce or change the concentration of plant hormones (Mordukhova *et al.* 1991), a symbiotic N₂ fixation (Boddey and Dobereiner 1995), solubilization of mineral phosphate and other nutrients (Goswami *et al.* 2013), and enzymes (Yuda *et al.* 2016). These results are confirmed in rice plant (Hafez *et al.* 2019; Amer *et al.* 2021; Devarajan *et al.* 2021 and Abd El-Mageed *et al.* 2022), Maize plant (Moridi, *et al.* 2019), Okra plant (Baliah and Muthulakshmi 2017), and sugar beet plant (Ghaffari *et al.* 2022).

Table 1. Effect of different irrigation intervals, soil and foliar treatments and their interactions on height plant (cm plant⁻¹), leaf area (cm²), and number of tillers plant⁻¹ of rice plant during 2020 and 2021 seasons.

Treatment	2020 season			2021 season		
	Height (cm plant ⁻¹)	Leaf area (cm ²)	No. Tillers (hill ⁻¹)	Height (cm plant ⁻¹)	Leaf area (cm ²)	No. Tillers (hill ⁻¹)
Irrigation intervals						
I ₁	75.74 ± 2.14 a	33.20 ± 0.36 a	19.62 ± 2.09 a	76.48 ± 2.34 a	34.83 ± 0.35 a	20.87 ± 2.20 a
I ₂	75.25 ± 3.51 a	31.27 ± 0.23 b	19.23 ± 2.05 ab	76.04 ± 3.65 a	32.79 ± 0.23 b	20.73 ± 2.08 a
I ₃	72.86 ± 6.74 b	27.90 ± 0.56 c	18.90 ± 2.00 b	73.68 ± 6.88 b	29.29 ± 0.55 c	20.32 ± 2.00 a
LSD 0.05	1.33	0.25	0.46	1.39	0.19	0.59
Soil and foliar treatments						
T ₁	71.38 ± 2.27 c	27.49 ± 0.64 d	16.64 ± 0.18 d	71.93 ± 2.27 d	29.00 ± 0.64 d	17.64 ± 0.18 d
T ₂	72.06 ± 6.57 c	30.97 ± 0.35 c	18.65 ± 0.39 c	72.71 ± 6.49 c	32.45 ± 0.37 c	20.32 ± 0.25 c
T ₃	76.39 ± 1.50 b	32.15 ± 0.38 b	19.63 ± 0.46 b	77.16 ± 1.50 b	33.58 ± 0.38 b	21.51 ± 0.65 b
T ₄	78.64 ± 1.21 a	32.54 ± 0.37 a	22.08 ± 0.28 a	79.79 ± 1.22 a	34.18 ± 0.37 a	23.08 ± 0.28 a
LSD 0.05	0.74	0.15	0.25	0.74	0.17	0.26
Interaction						
I ₁ T ₁	74.21 ± 0.91 e	30.10 ± 0.02 f	16.83 ± 0.05 e	74.76 ± 0.91 d	31.71 ± 0.02 g	17.83 ± 0.05 e
I ₁ T ₂	74.96 ± 1.06 e	33.29 ± 0.03 b	19.11 ± 0.09 c	75.51 ± 1.06 d	35.02 ± 0.03 c	20.11 ± 0.09 d
I ₁ T ₃	74.78 ± 1.06 e	30.14 ± 0.02 f	20.15 ± 0.13 b	75.55 ± 1.06 d	31.69 ± 0.02 g	22.15 ± 0.13 b
I ₁ T ₄	79.03 ± 0.29 ab	34.78 ± 0.03 a	22.38 ± 0.09 a	80.09 ± 0.51 a	36.50 ± 0.03 a	23.38 ± 0.09 a
I ₂ T ₁	69.58 ± 0.79 f	29.52 ± 0.02 h	16.68 ± 0.04 e	70.13 ± 0.79 e	31.10 ± 0.02 h	17.68 ± 0.04 f
I ₂ T ₂	77.70 ± 0.50 bcd	30.80 ± 0.02 e	18.53 ± 0.19 d	78.32 ± 0.58 bc	32.24 ± 0.05 f	20.53 ± 0.19 cd
I ₂ T ₃	76.48 ± 0.65 d	32.07 ± 0.03 d	19.61 ± 0.05 c	77.25 ± 0.65 c	33.46 ± 0.03 e	21.61 ± 0.05 b
I ₂ T ₄	77.25 ± 0.94 cd	32.73 ± 0.05 c	22.10 ± 0.09 a	78.45 ± 0.94 bc	34.37 ± 0.05 d	23.10 ± 0.09 a
I ₃ T ₁	63.54 ± 0.84 g	22.84 ± 0.02 j	16.43 ± 0.08 f	70.91 ± 0.84 e	24.19 ± 0.02 j	17.43 ± 0.08 f
I ₃ T ₂	70.36 ± 1.37 f	28.84 ± 0.04 i	18.32 ± 0.25 d	64.31 ± 1.37 f	30.12 ± 0.04 i	20.32 ± 0.25 cd
I ₃ T ₃	77.91 ± 0.08 bc	29.79 ± 0.05 g	19.12 ± 0.14 c	78.68 ± 0.08 b	31.18 ± 0.04 h	20.78 ± 0.51 c
I ₃ T ₄	79.63 ± 0.57 a	34.61 ± 0.07 a	21.75 ± 0.03 a	80.83 ± 0.57 a	36.50 ± 0.07 a	22.75 ± 0.03 a
LSD 0.05	1.33	0.27	0.45	1.34	0.32	0.48

I₁: Irrigation every 3 days; I₂: Irrigation every 6 days; I₃: Irrigation every 9 days; T₁: control; T₂: inoculation with PGPR (*P. koreensis* + *B. coagulans*); T₃: foliar spray with compost tea; T₄: combination (PGPR + compost tea). Means in the same column followed by the same letter are not significantly different according to Duncan's test at 0.05 level.

Physiological characteristics

The data illustrated in Table (2) show the effect of soil and foliar treatments (PGPR and foliar spray with CT) on chlorophyll, proline and relative water contents of rice plants at the flowering stage as influenced by irrigation intervals. Total chlorophyll and relative water contents were enhanced in I₁ treatment (irrigation every 3 days). However, it reduced in I₃ treatment (irrigation every 9 days), during the two growing

seasons. Among soil and foliar treatments (sub main), T₄ treatment (PGPR + CT) caused the greatest effect over all the other treatments (Table 2). The interaction effect between the different irrigation water treatments and soil and foliar treatments showed significant effects on physiological characteristics of rice plant (Table 2).

Table 2. Effect of different irrigation intervals, soil and foliar treatments and their interactions on chlorophyll, proline and relative water contents of rice plant during 2020 and 2021 seasons.

Treatment	2020 season			2021 season		
	Chlorophyll content (SPAD value)	Proline content (µ g ⁻¹ FW)	RWC (%)	Chlorophyll content (SPAD value)	Proline content (µ g ⁻¹ FW)	RWC (%)
Irrigation treatments						
I ₁	41.74 ± 2.84 a	6.93 ± 0.80 b	86.17 ± 13.54 a	43.11 ± 2.85 a	7.25 ± 0.51 b	86.48 ± 13.61 a
I ₂	41.53 ± 4.95 a	7.04 ± 0.98 b	80.69 ± 10.12 b	42.81 ± 4.85 a	7.42 ± 0.85 b	80.97 ± 10.21 b
I ₃	35.11 ± 3.62 b	8.00 ± 1.05 a	80.09 ± 5.21 b	36.36 ± 3.50 b	8.50 ± 1.09 a	80.39 ± 5.31 b
LSD 0.05	1.32	0.14	1.48	1.36	0.18	1.50
Inoculation treatments						
T ₁	35.61 ± 2.75 b	8.07 ± 1.12 a	67.57 ± 4.01 d	37.21 ± 2.75 b	8.18 ± 1.12 a	67.80 ± 4.01 d
T ₂	40.55 ± 7.09 a	7.41 ± 0.59 b	88.19 ± 4.42 b	41.92 ± 7.21 a	7.54 ± 0.58 b	88.38 ± 4.38 b
T ₃	40.58 ± 4.41 a	7.02 ± 0.93 c	82.97 ± 6.92 c	41.78 ± 4.41 a	7.49 ± 1.09 b	83.17 ± 6.99 c
T ₄	41.11 ± 2.39 a	6.80 ± 1.12 d	90.54 ± 4.54 a	42.12 ± 2.43 a	7.68 ± 1.12 b	91.10 ± 4.54 a
LSD 0.05	0.89	0.16	1.01	0.90	0.22	1.02
Interaction						
I ₁ T ₁	39.08 ± 0.19 c	6.90 ± 0.10 f	72.45 ± 2.11 i	40.68 ± 0.19 c	7.01 ± 0.10 fg	72.68 ± 2.11 i
I ₁ T ₂	44.81 ± 0.51 ab	7.53 ± 0.06 e	83.11 ± 0.21 f	46.41 ± 0.51 ab	7.65 ± 0.05 ef	83.34 ± 0.21 f
I ₁ T ₃	39.06 ± 0.24 c	7.60 ± 0.10 de	79.48 ± 0.74 g	40.26 ± 0.24 c	7.74 ± 0.10 ef	79.63 ± 0.74 g
I ₁ T ₄	44.02 ± 0.89 a	5.70 ± 0.20 h	85.33 ± 1.11 e	45.08 ± 1.00 b	6.58 ± 0.20 ij	85.89 ± 1.11 e
I ₂ T ₁	34.62 ± 0.80 e	7.87 ± 0.06 cd	66.42 ± 1.12 j	36.22 ± 0.80 de	7.98 ± 0.06 de	66.65 ± 1.12 j
I ₂ T ₂	39.60 ± 0.25 c	8.00 ± 0.20 bc	88.36 ± 1.59 d	40.58 ± 0.23 c	8.14 ± 0.20 d	88.54 ± 1.55 d
I ₂ T ₃	46.10 ± 0.12 a	5.80 ± 0.17 h	77.37 ± 0.79 h	47.30 ± 0.12 a	6.19 ± 0.60 j	77.52 ± 0.79 h
I ₂ T ₄	39.71 ± 0.36 c	6.50 ± 0.20 g	90.63 ± 0.90 c	40.69 ± 0.36 c	7.38 ± 0.20 fg	91.19 ± 0.90 c
I ₃ T ₁	33.13 ± 0.95 e	9.43 ± 0.38 a	63.85 ± 0.57 k	34.73 ± 0.95 e	9.54 ± 0.38 a	64.08 ± 0.57 k
I ₃ T ₂	31.13 ± 0.77 f	6.70 ± 0.17 fg	93.10 ± 0.79 b	32.33 ± 0.77 f	6.84 ± 0.17 hi	93.25 ± 0.79 b
I ₃ T ₃	36.59 ± 2.19 d	7.67 ± 0.21 de	92.07 ± 0.89 bc	37.79 ± 2.19 d	8.55 ± 0.21 c	92.36 ± 1.09 bc
I ₃ T ₄	46.60 ± 1.69 a	8.20 ± 0.10 b	95.65 ± 0.80 a	47.58 ± 1.69 a	9.08 ± 0.10 b	96.21 ± 0.80 a
LSD 0.05	1.60	0.29	1.82	1.61	0.39	1.84

I₁: Irrigation every 3 days; I₂: Irrigation every 6 days; I₃: Irrigation every 9 days; T₁: control; T₂: inoculation with PGPR (*P. koreensis* + *B. coagulans*); T₃: foliar spray with compost tea; T₄: combination (PGPR + compost tea). Means in the same column followed by the same letter are not significantly different according to Duncan's test at 0.05 level. Values are means ± standard deviation (SD) from three replicates.

Treatment I₃T₄ (irrigation every 9 days + PGPR + CT) recorded the highest values 46.60 (SPAD unit) and 95.65 % at the first growing season (2020), and 47.58 (SPAD unit) and 96.21 % at the second growing season (2021), for chlorophyll content and RWC compared to the other treatments, respectively. Regarding proline content, there was an increase with I₃T₁ treatment (irrigation every 9 days + control), which recorded the highest values 9.43 and 9.54 μg⁻¹ FW for the first and second growing seasons compared to the other treatments, respectively (Table 2). The application of PGPR and CT treatment for the plant has a positive effect on physiological characteristics (chlorophyll, proline and RWC), especially under stress conditions. This increase has been known as a tolerance mechanism to drought which probably due to the osmotic regulation in plants by organic solutes *i.e.* proline and soluble sugars (Salehi *et al.* 2016), and this is also reflected in an increase in absorb more sunlight and increase plant growth and yield (Ievinsh 2020; Ghaffari *et al.* 2022). Furthermore, foliar-applied with liquid organic fertilizers (Ji *et al.* 2017), and PGPR (Hafez *et al.* 2019), who showed a substantial potential for improving the biosynthesis of chlorophyll pigments, such as total chlorophyll, stomatal conductance, and relative water content under water stress.

K⁺, Na⁺ and K⁺/Na⁺ ratio in leaves of rice plant

Generally, there was a statistically significant (P < 0.05) increase in K⁺ % as well as K⁺/Na⁺ ratio in leaves of rice plant with increasing irrigation intervals with soil and foliar treatments, during both growing seasons, while Na⁺ % showed opposite results (Figure 2). Mostly, the highest K⁺ %, K⁺/Na⁺ ratio and the lowest Na⁺ % were obtained from plants that treated with PGPR + CT (T₄), as compared to individual application of PGPR (T₂) or CT (T₃) to rice plants under drought stress conditions (Figure 2). Under I₃ treatment (irrigation every 9 days), the best treatment that gave the highest percent of K was T₄ treatment (combination) attained 1.36 and 1.45 % followed by T₃ treatment (CT), attained 1.19 and 1.28 %, followed by T₂ treatment (PGPR) attained 1.06 and 1.09 %, compared to T₁ treatment (control), 1.00 and 1.07 % in seasons 2020 and 2021, respectively (Figure 2A). Similar trend was observed in K⁺/Na⁺ ratio (Figure 2C). On the contrary, Na⁺ percent declined with soil treatments and foliar application treatments. The highest reduction of Na⁺ percent was in rice leaves grown under irrigation water stress (irrigation every 9 days, I₃), which decreased from 2.23 % (control, T₁) to 2.11 % (PGPR, T₂), 1.88 % (CT, T₃) and 1.61 % (combination, T₄) in season 2020, whereas in season 2021 the same rate was attained from 2.30 % (control, T₁) to 2.15 % (PGPR, T₂), 1.90 % (CT, T₃) and 1.66 % (combination, T₄) significantly as shown in Figure (2B). The application of PGPR with CT to soil and plants can increase the availability of macroelements (N, P and K), and nutrient cycling in the soil, and their reflection to improve the growth and crop production (Al- Eazy *et al.* 2018). This can be clearly in our study, as the leaf K⁺ % increased and the Na⁺ % decreased which led to an increase in the K⁺/Na⁺ ratio under each irrigation interval. On the other hand, PGPR can be reduce plant uptake of Na⁺ through the secretion of IAA (indole acetic acid) and EPS (exopolysaccharides), where it can bind Na⁺ and prevent its absorption into plants (Mena *et al.* 2015; Hafez *et al.* 2019).

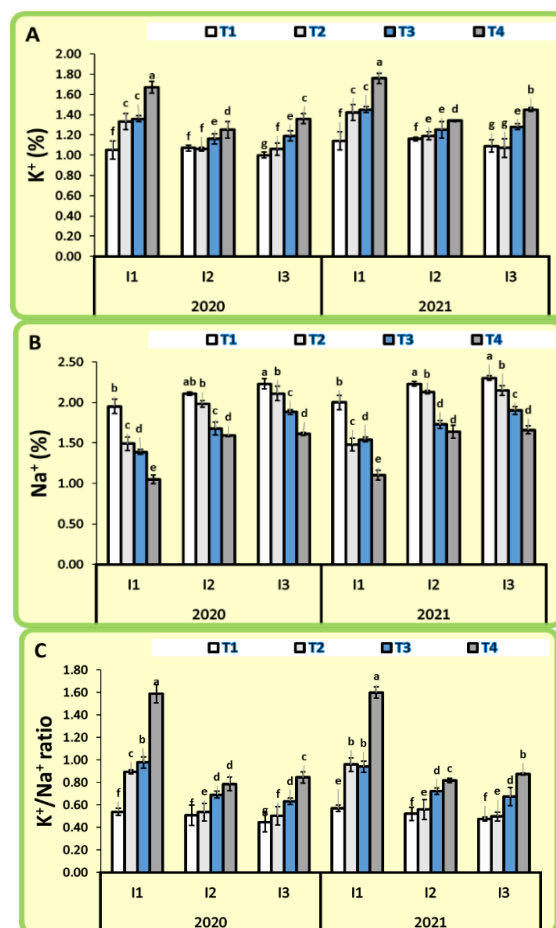


Figure 2. Effect of different irrigation intervals, soil and foliar treatments and their interactions on K⁺, Na⁺ and K⁺/Na⁺ ratio in leaves of rice plant during 2020 and 2021 seasons. I₁: Irrigation every 3 days; I₂: Irrigation every 6 days; I₃: Irrigation every 9 days; T₁: control; T₂: inoculation with PGPR (*P. koreensis* + *B. coagulans*); T₃: foliar spray with compost tea; T₄: combination (PGPR + compost tea). Column followed by the same letter are not significantly different according to Duncan’s test at 0.05 level. Values are means ± standard deviation (SD) from three replicates.

Microelements in rice leaves

Rice plant was exposed to different intervals of irrigation water (I1, I2 and I3) and combination treatment (PGPR + CT) showed significant (P < 0.05) higher microelements content (Zn, Mn, Fe and Cu), than single treatment (PGPR or CT) (Figure 3). At the flowering stage, T4 treatment (combination; PGPR +CT) leads to record 51.56, 37.38, 91.13 and 8.27 for Zn, Mn, Fe and Cu (mg Kg⁻¹) in rice leaves under I3 treatment (irrigation every 9 days) during 2020 season compared to control treatment, respectively. Similar trend was observed in 2021 season (Figure 3). Therefore, T4 treatment (combination; PGPR +CT) showed the maximum uptake of microelements in rice plants than other studied treatments and the descending order for irrigation treatment (main plots) were I1 > I2 > I3. However, the descending order for soil and foliar spray treatments (sub plots) were T4 (combination; PGPR +CT) > T3 (CT) > T2 (PGPR) > T1 (control) (Figure 3). These

elements are very important for the plant growth, as they are involved in the formation of enzymes, amino acids, proteins and DNA, as well as in the processes of photosynthesis. Therefore, soil microorganisms and foliar spray with compost

tea can increase plant nutrients by using traits that are appropriate and can be identified as growth promoters for plants (Khalifa *et al.* 2021; Omara *et al.* 2022).

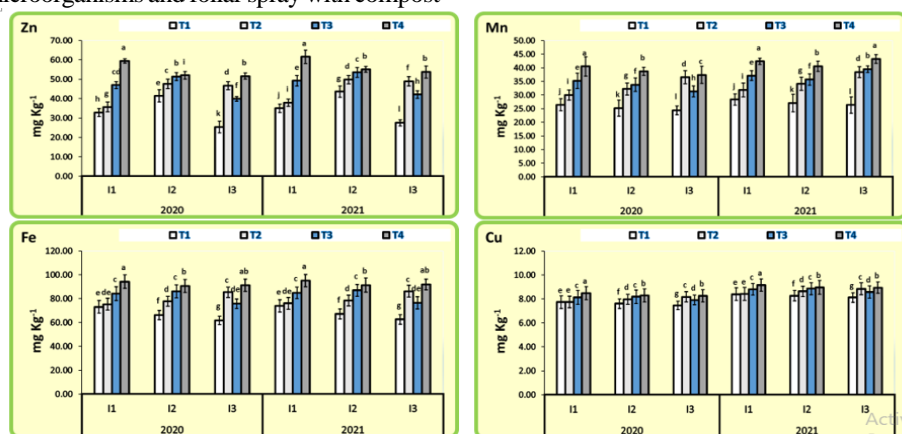


Figure 3. Effect of different irrigation intervals, soil and foliar treatments and their interactions on microelements (mg Kg⁻¹), in leaves of rice plant during 2020 and 2021 seasons. I1: Irrigation every 3 days; I2: Irrigation every 6 days; I3: Irrigation every 9 days; T1: control; T2: inoculation with PGPR (*P. koreensis* + *B. coagulans*); T3: foliar spray with compost tea; T4: combination (PGPR + compost tea). Column followed by the same letter are not significantly different according to Duncan’s test at 0.05 level. Values are means ± standard deviation (SD) from three replicates.

Microbial communities

At the flowering stage, microbial community i.e. total bacterial count, total count of *Bacillus* and total count of *Pseudomonas*, in the rhizosphere of rice plants grown under drought stress conditions (irrigation water intervals), were significantly varied with respect to the application of soil and foliar spray treatments ($P < 0.05$) in both seasons 2020 and 2021 (Fig. 4). In general, results illustrate that different soil and foliar spray treatments attained differences in microbial community. Where, T4 treatment (combination) showed the highest population of total count of bacteria 6.67, 6.69 and 6.17 CFU (log₁₀) g⁻¹ (Fig. 4A), total count of *Bacillus* 3.88, 3.55 and 3.37 CFU (log₁₀) g⁻¹ (Fig. 4B), and total count of *Pseudomonas* 1.93, 1.86 and 1.77 CFU (log₁₀) g⁻¹ (Fig. 4C), for the different irrigation water intervals I1, I2 and I3 during the first growing season (2020), compared to the other treatments, respectively. The same trend was noticed in the second growing season (2021). Some researchers support these results; Ji *et al.* (2017) suggested that the liquid organic fertilizers significantly increased the soil’s functional microbial community at the rhizospheric of *Chrysanthemum* compared with control treatment. Hafez *et al.* (2019) showed that the application of PGPR (*Azospirillum brasilense* and *Azotobacter chroococcum*) increased soil microbial activity which led to increase field capacity and available soil water in the rhizosphere of wheat plants. In addition, Khalifa *et al.* (2021) found that the soil amended with PGPR inoculation (*Azospirillum lipoferum*, *B. coagulans*, *B. circulance* and *B. subtilis*) improved significantly microbial growth in the rhizosphere of maize plants.

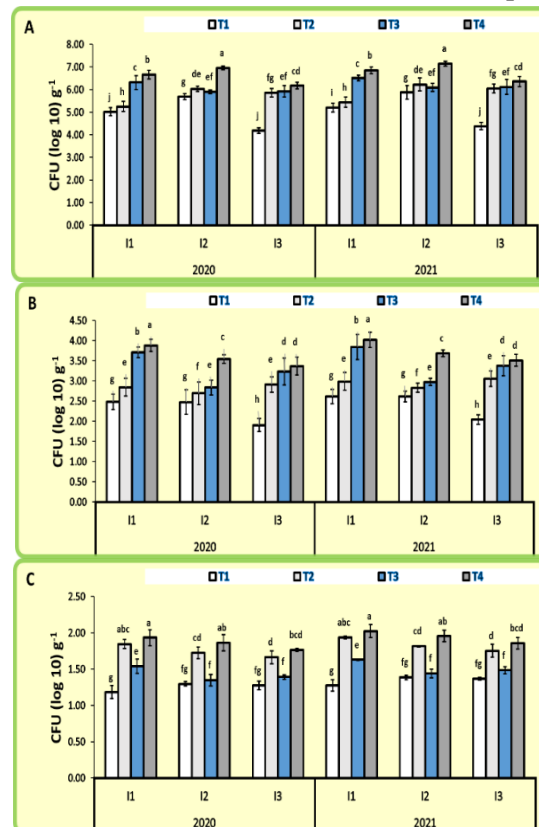


Figure 4. Effect of different irrigation intervals, soil and foliar treatments and their interactions on microbial communities (total count of bacteria (A), total count of *Bacillus* (B), total count of *Pseudomonas* (C)), in the rhizosphere of rice during 2020 and 2021 seasons. I1: Irrigation every 3 days; I2: Irrigation every 6 days; I3: Irrigation every 9 days; T1: control; T2: inoculation with PGPR (*P. koreensis* + *B. coagulans*); T3: foliar spray with compost tea; T4: combination (PGPR + compost tea). Column followed by the same letter are not significantly different according to Duncan’s test at 0.05 level. Values are means ± standard deviation (SD) from three replicates.

Activity of soil enzymes

The variation in soil enzyme activities (dehydrogenase, urease, amylase and invertase) were selected upon their different biological processes in the soil i.e. dehydrogenase was estimated to determine microbial activity (overall), urease was selected for their role of nitrogen cycle in soil. Besides, amylase and invertase were selected for their importance of carbon cycle in soil (Fig. 5). Generally, all soil enzymes were noted to increase with the combination treatment (PGPR + CT, T4) and recorded 125.63 and 143.63 mg TPF g⁻¹ soil day⁻¹ for dehydrogenase activity, 84.33 and 93.13 NH₄⁺-N g⁻¹ soil day⁻¹ for urease activity, 0.11 and 0.12 mg glucose g⁻¹ soil h⁻¹ for amylase activity and 0.059 and 0.070 mg sucrose g⁻¹ soil h⁻¹ for invertase activity under I3 treatment (irrigation every 9 days), during 2020 and 2021 seasons compared to the other treatments, respectively (Fig.5). Increasing soil enzyme activity by applying PGPR and foliar spraying with compost tea under drought stress in the present study may accelerate the metabolic processes of aerobic organisms that play an important role in controlling the release of bioavailable nutrients from organic compounds (Sinica *et al.* 2013; Sinsabaugh *et al.* 2014). The main reason is that soil enzyme activity is closely related to the class, counts, and abundance of soil microbes (Wei and Yan 2018), and the metabolism as well as reproduction of a soil microbial

community has been adopted to be influenced by temperature, precipitation and moisture over time (Zhang *et al.* 2015). These results are confirmed in rice plant (Hafez *et al.* 2019; Qu *et al.* 2020); Eucalyptus plant (Ren *et al.* 2020, 2021); maize plant (Khalifa *et al.* 2021); wheat plant (Omara *et al.* 2022).

To understand the impact of different studied treatments (main and sub main) on crop resilience and improvement, yield parameters, such as 1000-grain weight, number of grains panicles-1, number of panicles m² as well as grain and straw yields (t ha⁻¹) were measured during 2020 and 2021 seasons (Table 3). At different irrigation water treatments, treatment I1 (irrigation every 3 days) gave the highest values of rice yield compared to those stress treatments (I2 and I3). Therefore, the data of irrigation treatments followed the descending order of I1 > I2 > I3. While it followed as T4 > T3 > T2 > T1 under soil and foliar spray treatments. Regarding the interaction between the main plot (irrigation water intervals) and sub main plot (soil and foliar spray), data showed that I3T4 treatment (combination) attained 25.92, 140.53, 463.75, 8.42 and 14.49 compared to I3T1 treatment-t (control), which attained 23.60, 106.20, 350.46, 7.93, 10.95 in 2020 season for 1000-grain weight, number of grains panicles-1, number of panicles m², grain and straw yields (t ha⁻¹), respectively.

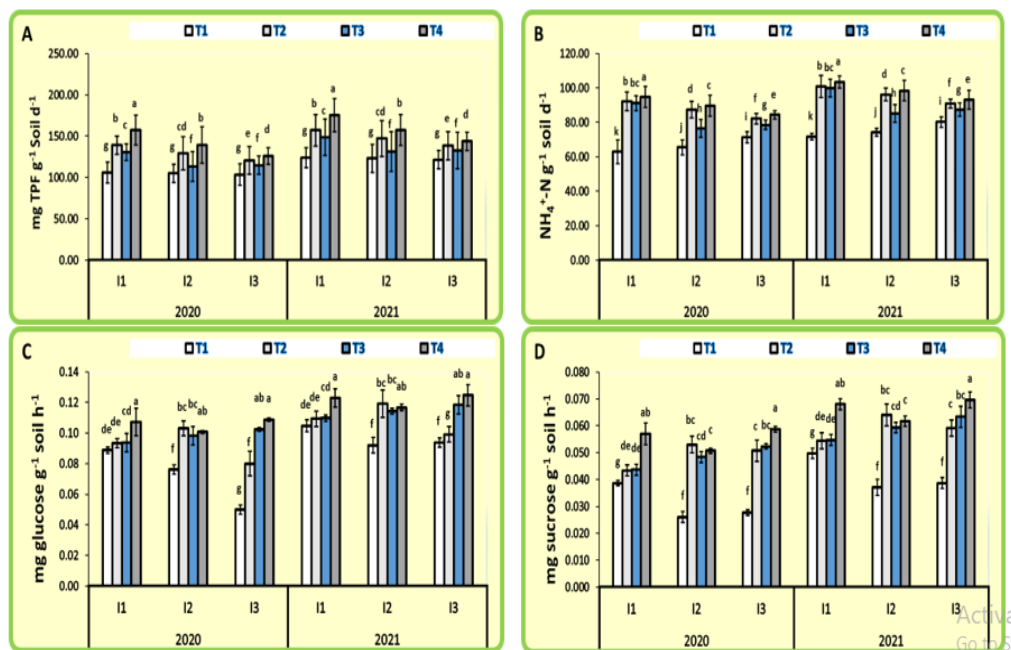


Figure 5. Effect of different irrigation intervals, soil and foliar treatments and their interactions on activity of soil enzymes (dehydrogenase (A), Urease (B), Amylase (C), Invertase (D)) in the rhizosphere of rice during 2020 and 2021 seasons. I1: Irrigation every 3 days; I2: Irrigation every 6 days; I3: Irrigation every 9 days; T1: control; T2: inoculation with PGPR (*P. koreensis* + *B. coagulans*); T3: foliar spray with compost tea; T4: combination (PGPR + compost tea). Column followed by the same letter are not significantly different according to Duncan's test at 0.05 level. Values are means ± standard deviation (SD) from three replicates. Yield parameters

Similar results were observed in the second growing season (Table 3). These results showed that PGPR and CT can increase yield parameters of rice under water stress conditions which due to increasing availability and amount of soil nutrients uptake. This finding was found to be in

harmony with several studies such as wheat (Hussain *et al.* 2014), soybean (Kang *et al.* 2014), maize (Cohen *et al.* 2009), and rice (Hafez *et al.* 2019; Abd El-Mageed *et al.* 2022).

Table 3. Effect of different irrigation intervals, soil and foliar treatments and their interactions on 1000-grain weight, number of grains panicles⁻¹, number of panicles m⁻², grain and straw yields (t ha⁻¹) of rice plant during 2020 and 2021 seasons.

Treatment	2020 season					2021 season				
	1000-grain weight(g)	No. grains Panicle ¹	No. Panicles (m ²)	Grain yield (tha ¹)	Straw yield (tha ¹)	1000-grain weight(g)	No. grains Panicle ¹	No. Panicles (m ²)	Grain yield (tha ¹)	Straw yield (tha ¹)
Irrigation treatments										
I ₁	27.23±1.10a	134.35±19.41a	443.36±64.06a	8.87±0.24a	13.85±2.00a	27.47±1.12a	138.93±20.53a	455.61±65.21a	8.98±0.25a	14.00±2.01a
I ₂	24.35±1.08b	122.81±14.11b	405.28±46.58b	8.36±0.27b	12.67±1.46b	24.60±1.06b	127.81±15.25b	416.78±48.08b	8.47±0.27b	12.82±1.47b
I ₃	20.81±1.92c	117.24±8.76c	386.90±28.91c	7.58±0.57c	12.09±0.90c	21.06±1.91c	122.24±10.00c	400.15±30.89c	7.70±0.60c	12.25±0.92c
LSD0.05	0.36	1.64	6.57	0.13	0.20	0.37	1.34	6.67	0.12	0.21
Inoculation treatments										
T ₁	22.86±3.81c	105.94±1.44d	349.60±4.74d	8.02±0.53c	10.93±0.15d	23.09±3.81c	108.94±1.44d	361.60±4.74d	8.13±0.53c	11.08±0.15d
T ₂	23.14±2.50c	120.64±8.36c	398.12±27.59c	8.04±0.99c	12.44±0.86c	23.41±2.47c	125.42±8.00c	408.12±29.08c	8.14±0.99c	12.57±0.87c
T ₃	24.93±2.63b	130.71±8.15b	431.34±26.88b	8.45±0.33b	13.48±0.84b	25.21±2.65b	136.38±7.71b	442.68±24.06b	8.57±0.31b	13.63±0.81b
T ₄	25.58±2.37a	141.92±14.12a	468.32±46.59a	8.56±0.51a	14.64±1.46a	25.80±2.37a	147.92±4.12a	484.32±46.59a	8.70±0.51a	14.82±1.46a
LSD0.05	0.39	2.30	7.78	0.09	0.24	0.38	2.29	7.78	0.08	0.24
Interaction										
I ₁ T ₁	26.82±0.50b	107.05±1.06g	353.28±3.50g	8.66±0.15bc	11.04±0.11g	27.05±0.50b	110.05±1.06f	365.28±3.50f	8.77±0.15bc	11.19±0.11g
I ₁ T ₂	25.80±0.26c	131.69±1.47c	434.57±4.84c	8.80±0.17b	13.58±0.15c	26.03±0.26c	136.02±0.42c	446.57±4.84c	8.90±0.16b	13.72±0.16c
I ₁ T ₃	28.18±0.71a	140.27±1.63b	462.90±5.38b	8.81±0.11b	14.47±0.17b	28.47±0.71a	145.27±1.63b	471.90±5.38b	8.90±0.11b	14.59±0.17b
I ₁ T ₄	28.10±0.26a	158.39±4.39a	522.68±14.50a	9.21±0.03a	16.33±0.45a	28.32±0.26a	164.39±4.39a	538.68±14.50a	9.35±0.03a	16.51±0.45a
I ₂ T ₁	18.17±0.46h	104.57±1.39g	345.07±4.60g	7.46±0.07g	10.78±0.14g	18.40±0.46h	107.57±1.39f	357.07±4.60f	7.57±0.07f	10.93±0.14g
I ₂ T ₂	20.10±0.34g	115.85±0.63f	382.32±2.09f	6.74±0.05h	11.95±0.07f	20.39±0.34g	120.85±0.63e	391.32±2.09e	6.83±0.05g	12.07±0.07f
I ₂ T ₃	22.26±0.30f	121.72±1.39e	401.68±4.58e	8.07±0.06f	12.55±0.14e	22.50±0.30f	127.72±1.39d	417.68±4.58d	8.21±0.09e	12.73±0.13e
I ₂ T ₄	22.73±0.64f	126.83±4.41d	418.54±14.57d	8.06±0.11f	13.08±0.46d	22.95±0.64f	132.83±4.41c	434.54±14.57c	8.20±0.11e	13.26±0.46d
I ₃ T ₁	23.60±0.42e	106.20±0.63g	350.46±2.06g	7.93±0.09f	10.95±0.06g	23.83±0.42e	109.20±0.63f	362.46±2.06f	8.04±0.09e	11.10±0.06g
I ₃ T ₂	23.52±0.24e	114.38±0.95f	377.47±3.12f	8.59±0.12cd	11.80±0.10f	23.81±0.24e	119.38±0.95e	386.47±3.12e	8.68±0.12cd	11.92±0.10f
I ₃ T ₃	24.35±0.04d	130.14±1.38cd	429.45±4.56cd	8.48±0.06de	13.42±0.14cd	24.64±0.02d	136.14±1.38c	438.45±4.56c	8.60±0.06cd	13.58±0.14cd
I ₃ T ₄	25.92±0.21c	140.53±2.68b	463.75±8.84b	8.42±0.05e	14.49±0.28b	26.14±0.21c	146.53±2.68b	479.75±8.84b	8.56±0.05d	14.67±0.28b
LSD0.05	0.70	4.14	13.97	0.16	0.43	0.69	4.11	13.99	0.17	0.44

I₁: Irrigation every 3 days; I₂: Irrigation every 6 days; I₃: Irrigation every 9 days; T₁: control; T₂: inoculation with PGPR (*P. koreensis* + *B. coagulans*); T₃: foliar spray with compost tea; T₄: combination (PGPR + compost tea). Means in the same column followed by the same letter are not significantly different according to Duncan's test at 0.05 level. Values are means ± standard deviation (SD) from three replicates.

CONCLUSION

Based on the results obtained here, plants treated with PGPR (*P. koreensis* + *B. coagulans*) + compost tea (CT) showed higher vegetative growth, physiological characteristics compared to control plants under irrigation water stress conditions. These results had a positive impact on nutrient content and soil biological activities as well as yield of rice plants. So, PGPR and CT can be a great achievement to reduce the drought stress in rice plants.

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التأثير المشترك للبكتيريا النافعة وشاي الكمبوست على نمو وإنتاجية محصول الأرز وخصائص التربة البيولوجية تحت ظروف إجهاد الجفاف

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خلال موسمي الزراعة الصيفيين 2020 و 2021، تم إجراء تجربتين حقليتين بتصميم القطعة المنشقة في محطة البحوث الزراعية بسخا، بمحافظة كفر الشيخ بمصر، لتقييم تأثير المعاملات الفردية والمختلطة (ثلاث مكررات لكل معاملة) لبكتيريا الجذر المشجعة لنمو النبات والرش الورقي لشاي الكمبوست على تحسين النمو الخضري والخصائص الفسيولوجية والمحتوى الغذائي والأنشطة البيولوجية للتربة بالإضافة إلى إنتاج محصول الأرز عند الري علي فترات مختلفة. أظهرت النتائج أن جميع فترات الري المختلفة المصحوبة بالمعاملة المختلطة أعطت أعلى قيم للنمو الخضري (ارتفاع النبات ومساحة الورقة وعدد الأشطاء)، الخصائص الفسيولوجية (الكوروفيل الكلي والبرولين والمحتوى المائي النسبي) خلال موسمي الزراعة. أيضاً، في مرحلة الإزهار، أظهر محتوى الأوراق من البوتاسيوم ونسبة البوتاسيوم إلي الصوديوم والزنك والمنجنيز والحديد والنحاس زيادة معنوية عند تعرض نباتات الأرز لمعاملة (الري كل 9 أيام + المعاملة المختلطة)، بينما كان هناك انخفاض في نسبة الصوديوم خلال موسمي 2020 و 2021. ولوحظ اتجاه مماثل في التعداد الميكروبي في تربة جذور نبات الأرز. ومن ناحية أخرى، لوحظ أن جميع إنزيمات التربة تزداد مع المعاملة المختلطة (الديهيدروجينيز واليوريز والأميليز والإنفرتيز) تحت تأثير معاملة (الري كل 9 أيام)، خلال موسمي 2020 و 2021 مقارنة بالمعاملات الأخرى. بالنسبة لمعايير الإنتاج، فقد تأثرت سلباً كلما ازدادت فترات الري. بينما تحسنت النتائج علي الترتيب ابتداء من المعاملة الأولى إلي المعاملة الرابعة. وعماً، يمكن استخدام المعاملة المختلطة تحت فترات الري المختلفة بكفاءة للتخلص جزئياً من آثار الإجهاد المائي على نمو وإنتاجية الأرز.