JSAES
Journal of Sustainable Agricultural and Environmental Sciences

Print ISSN : 2735-4377
Online ISSN : 2785-9878
Homepage: https://jsaes.journals.ekb.eg/

Research Article

Enhancing Drought Tolerance in *Paspalum vaginatum* Through Rhizobacteria Inoculation Under Variable Irrigation Regimes

Mona Ramadan, Fahmy A. S. Hassan*, Mohammed I. Fetouh and Rasha S. El-Serafy*

[1] Horticulture Department, Faculty of Agriculture, Tanta University, Tanta 31527, Egypt.
* Correspondence: Fahmy A. S. Hassan; fahmy.hassan@agr.tanta.edu.eg; Rasha S. El-Serafy; rasha.elserafi@agri.edu.eg

Article info: -

- **Received:** 20 September 2025
- **Revised:** 1 October 2025
- **Accepted:** 5 October 2025
- **Published:** 8 October 2025

Keywords:
Irrigation frequency; Semi-arid regions; Biomass production

Abstract:

The ongoing depletion of global freshwater resources has intensified the need for sustainable irrigation strategies, particularly in semi-arid regions such as the southern Mediterranean. This study investigated the drought tolerance of *Paspalum vaginatum* under varying irrigation frequencies and assessed the effectiveness of plant growth-promoting rhizobacteria (PGPR) in enhancing plant performance under water deficit conditions. The experiment was conducted over two consecutive years (2020–2021), using six treatments arranged in a factorial design: two main groups (with and without PGPR inoculation), each subdivided into three irrigation intervals (every 2, 6, and 10 days). Prolonged irrigation intervals simulated increasing levels of drought stress, which significantly reduced fresh and dry shoot weights, relative water content, chlorophyll and carotenoid concentrations, and the uptake of essential nutrients including nitrogen, phosphorus, and potassium. Inoculation with a PGPR consortium—comprising *Azotobacter spp., Bacillus* spp., *Pseudomonas fluorescens, and Bacillus circulans/megaterium*—led to notable improvements in plant growth and physiological attributes under drought stress. PGPR-treated plants demonstrated higher biomass and increased pigment levels. Moreover, inoculation contributed to better osmotic regulation and more balanced nutrient uptake compared to non-inoculated controls. These findings suggest that PGPR applications can effectively mitigate the negative impacts of drought stress in *Paspalum vaginatum*, offering a promising biological tool for improving turfgrass resilience and reducing irrigation demands in water-limited environments.

1. Introduction

Global climate change, rapid urbanization, and increasing competition for freshwater resources have intensified the challenges facing green infrastructure, especially in arid and semi-arid regions (El-Tayeh et al., 2020). Urban green spaces, including turfgrass systems in parks, sports fields, and residential areas, play essential roles in environmental regulation, aesthetic enhancement, public well-being, reduce air pollution, mitigate noise, help cool down during warmer periods, and create a sustainable, climate-resilient environment. (Breuste et al., 2013). However, turfgrass maintenance under water-deficient conditions poses a major concern due to the high irrigation demands of many turf species. This calls for the urgent development of water-saving practices and stress-resilient plant varieties to ensure sustainable landscaping (Jiang and Huang, 2001).

Among abiotic stresses, drought is considered one of the most limiting factors for plant growth and productivity worldwide (Mazrou et al., 2023). Drought stress impairs cellular water status and causes substantial reductions in photosynthesis, nutrient uptake, and biomass accumulation. It induces oxidative stress by enhancing reactive oxygen species (ROS) formation, damaging cellular structures, lipids, proteins, and nucleic acids (Rue and Zhang, 2020). In turfgrasses, water deficit often leads to premature senescence, chlorophyll degradation, and loss of turf quality and coverage (Gasemi et al., 2023). Additionally, prolonged drought reduces relative water content (RWC), stomatal conductance, and enzymatic activity, ultimately disrupting plant homeostasis (Mittler, 2002).

Paspalum vaginatum, commonly known as seashore paspalum, is a warm-season, perennial grass species native to coastal and tropical regions (Chen et al., 2005). Its tolerance to salt and marginal soils has made it an attractive species for golf courses and turf systems, especially in resource-limited environments (Duncan and Carrow, 2009). In addition to its tolerance to salinity, paspalum demonstrates moderate resilience to drought conditions, though its growth and physiological traits are still significantly impacted under prolonged water scarcity (Trenholm et al., 2012). Therefore, enhancing the drought tolerance of paspalum through sustainable biological approaches is essential for conserving water and ensuring turf quality under stress. One promising strategy is the use of plant growth-promoting rhizobacteria (PGPR), a group of beneficial microbes that colonize plant roots and confer multiple advantages under abiotic stress conditions (Baez-Rogelio et al., 2017). PGPR facilitate plant tolerance to drought by improving root development, enhancing nutrient and water uptake, modulating stress-related hormones such as abscisic acid and auxins, and stimulating the production of osmoprotectants and antioxidants (Eida et al., 2020). Several studies have demonstrated that PGPR, including strains of *Azotobacter, Bacillus, and Pseudomonas fluorescens*, can enhance drought resistance in various crops and turfgrasses by improving chlorophyll content, biomass production, and antioxidant enzyme activity (Vurukonda et al., 2016).

Despite increasing interest of this research area, limited research exists on the interaction between drought stress and rhizobacterial inoculation in *Paspalum vaginatum*. Most existing studies focus on salinity, while drought, a growing threat under changing climatic conditions, remains underexplored in this context. Therefore, this study aims to evaluate the effectiveness of a multi-strain PGPR consortium—*Azotobacter* spp., *Bacillus* spp., *Pseudomonas fluorescens*, and *Bacillus circulans*/*megaterium*—in alleviating the adverse effects of drought stress on *Paspalum vaginatum*. The study focuses on key physiological, biochemical, and morphological parameters, with the objective of developing a sustainable strategy for drought-stress mitigation in turfgrass systems.

2. Materials and Methods

2.1. Experimental site and setup

This experiment was conducted at the experimental farm of the Faculty of Agriculture, Tanta University, Egypt (30° 47′ 18″ N: 31° 00′ 06″ E) at an altitude of 8 m above sea level during the summer seasons of 2020 and 2021. To simulate coastal cultivation, six raised beds (6 m long x 1 m wide x 30 cm high) were constructed using outdoor sandy soil, spaced 1.5 m apart. Paspalum rolls were obtained from a private nursery in Badr District, El-Beheira Governorate, Egypt and planted on July 1st for each season. alternating 3 m sections (1 m planted, 1 m fallow) on each bed. During the initial two weeks, plants were irrigated daily with tap water to stimulate root growth.

2.2. Bacterial inoculant

A plant growth-promoting rhizobacteria (PGPR) inoculum was used in this study, consisting of four strains: (*Azotobacter Chroococcum, Bacillus circulans, Bacillus megaterium, and Pseudomonas fluorescens*).The inoculum was prepared at a concentration of 10^8–10^9 colony-forming units (CFU) per milliliter and was provided by the(Central Laboratory for Organic Agriculture) Agricultural Research Center, Giza, Egypt. According to standard recommendations, 10 liters of the concentrated inoculum is sufficient for application to one feddan. In this experiment, 2 liters of the stock solution were diluted with tap water to a final volume of 27 liters and used for inoculation as described below. In each bed, 2 liters were applied per meter of planted turf and 1 liter per meter of unplanted (fallow) area, resulting in a total application of 9 liters per bed and 27 liters across all three beds. The inoculation was conducted twice in each season, on 15th July and 15th August.

2.3. Irrigation frequency treatments

One week after the first bacterial application, drought stress was imposed by varying the frequency of irrigation according to the treatment plan. To simulate drought stress, *Paspalum vaginatum* was subjected to three irrigation frequency treatments based on different watering intervals:

(1) Irrigation every 2 days (well-watered control; D2),
(2) Irrigation every 6 days (moderate drought stress; D6), and
(3) Irrigation every 10 days (severe drought stress; D10).

All plants received equal volumes of water per irrigation to ensure that the difference in plant response was due solely to irrigation interval and not to the amount of water given. These irrigation intervals were maintained consistently throughout the experiment to induce varying levels of water deficit stress. Irrigation was carried out for all treatments at a constant water rate of 10 liters/m^2 per irrigation event was kept uniform across treatments to ensure that differences in plant responses were due to irrigation frequency rather than water quantity per event.

2.4. Experimental design

This experiment was planned as factorial in randomized complete block design (RCBD): with three replications. The first factor was the application of plant growth-promoting rhizobacteria (PGPR), which included two levels: without inoculation and with inoculation. The second factor was irrigation frequency, which was used to simulate drought stress and included three irrigation intervals: every 2 days (as a well-watered control), every 6 days (moderate drought stress), and every 10 days (severe drought stress). This design resulted in six treatment combinations in total.

2.5. Traits investigated

At 7, 14, 21, 28, 35, 42, 49, 56 and 63 days post-stress, samples were collected to assess the following traits:

2.5.1. Clipping fresh and dry weights determination

Plant biomass was assessed following standardized protocols (Poorter et al., 2012) with modifications for salinity stress studies. A sample area of 1 m2 was clipped and immediately aerial tissues were excised at the soil interface using scissors. Samples were processed within 90 seconds of collection to minimize post-harvest water loss. Fresh weight (FW) measurements were conducted using a calibrated analytical balance in a temperature-controlled environment. To assess the dry weight, samples were oven-dried at 70°C for 72 hours until constant weight was gained, and dry weights (DW) were recorded. Three technical replicates were measured by treatment.

2.5.2. Relative water content (%)

Relative water content (RWC) was determined on leaf tissues excised in the morning (around 9:00 am). Excised leaves were measured for fresh weight (FW) and then rehydrated in a water-filled petri dish at room temperature. Turgor weight (TW) was measured by allowing full rehydration (16 h), removing all water on the leaf surface, weighing, and then the leaves were dried at 70°C for 48 h to determine DW. The relative water content was calculated from the following equation, RWC = 100[FW - DW)/ (TW- DW)] as reported by Weatherley (1950).

2.5.3. Chlorophyll quantification

The amount of total chlorophyll was determined according to Dere et al. (1998). Fresh leaves (0.1 g) were cut into small fragments (1mm x 1 mm) and immersed for 24 h at 4°C in 20 ml methanol (96%) and then filtered through Whatman 47 mm GF/C filter paper. The absorbance of each filtrate was measured against a blank of 96% methanol at wavelengths of 666 and 653 nm for chlorophyll a and b, respectively. Data were expressed as mg g-1 fresh weight FW

2.5.4. Mineral content

After seven weeks of applied treatments, desiccated leaf samples were ground into a fine powder and then digested using a mixture of perchloric acid and sulfuric acid (1:5 v/v, respectively) as described by A.O.A.C. (1995). The determination of N, P, and K contents in leaf tissues was performed using the abovementioned digestion solution. Measuring of N was conducted by the micro-Kjeldahl apparatus as described by Nelson and Sommers (1973). Phosphorus content was measured using spectrophotometer (Pharmacia, LKB-Novaspec II) following the blue color according to the method of (Jackson 1967). K and Na elements were estimated by flame emission photometry (Corning, Tewksbury, MA, USA).

2.6. Statistical analysis

The results of each season were statistically analyzed, and analysis of variance (ANOVA) was performed using Michigan Statistical Program Version C (MSTATC). Means were separated by Duncan multiple range test at 0.05 probability level.

3. Results and Discussion

3.1. Clipping fresh and dry weights

The results revealed significant effects of rhizobacteria inoculation, drought levels, and their interaction on both fresh weight (Table 1) and dry weight (Table 2) of *Paspalum vaginatum* during both growing seasons. Rhizobacteria inoculation consistently improved plant performance, with treated plants exhibiting higher fresh and dry weights throughout the investigated period compared to untreated. However, increasing drought period significantly reduced fresh and dry biomass (Table 2). At drought (D2), plants exhibited severe declines in fresh weight, representing 63.72 and 60.24% reductions compared to D0 at week 7 in both seasons, respectively. On the other hand, rhizobacteria inoculation markedly enhanced clipping fresh and dry weights when plants were exposed to drought. The promotion effect of rhizobacteria inoculation on paspalum growth observed in this study is consistent with the results of Mahdavi et al. (2020) who reported that bacteria improved the physical, morphological, and biochemical properties of Tall Fescue under water deficient conditions compared to the control. Our results were also consistent with Fadil et al. (2025), where inoculation with root bacteria improved the growth of Paspalum turf grass. In contrast, the reductions in growth traits of paspalum observed in current study are in agreement with the report of Porcelli et al. (2024) who found that the total biomass (root and green components) of the Paspalum plants decreased under drought stress. In the same context, Ragályi et al. (2023) on Paspalum, Ali et al. (2018) on Bermuda turfgrass, reported similar results.

Table 1. Effect of rhizobacteria inoculation, irrigation intervals and their interaction on clipping fresh weight of *Paspalum vaginatum* turfgrass (area of 1 m^2) during 2020 and 2021seasons.

Rhizobacteria inoculation (Rhizo)	Irrigation	Week 1	Week 2	Week 3	Week 4	Week 5	Week 6	Week 7	Week 8	Week 9
		First season 2020								
Without Rhizo.	-	34.87 B	38.61 B	39.79 B	44.24B	47.79 B	55.77 B	55.79 B	64.83 B	83.85 B
With Rhizo.		41.05 A	45.55 A	48.05 A	52.53 A	56.18 A	60.72 A	64.31 A	71.42 A	92.44 A
-	2 D	50.89 A	55.50 A	55.83 A	58.36 A	64.41 A	68.91 A	73.00 A	82.35 A	99.27 A
	6 D	34.83 B	39.25 B	41.34 B	48.09 B	49.08 B	58.66 B	56.82 B	71.08 B	89.61 B
	10 D	28.16 C	31.50 C	34.59 C	38.71 C	42.46 C	47.16 C	50.33 C	50.94 C	75.55 C
Without Rhizo.	2 D	45.28 B	50.00 B	50.00 B	53.32 B	58.33 B	66.33 B	66.66 B	77.83 B	95.88 B
	6 D	32.99 CD	36.18 D	37.09 C	44.51 C	44.73 D	57.00 D	52.37 D	67.33 C	84.22 C
	10 D	26.33 E	29.66 E	32.29 C	34.88 D	40.31 E	44.00 F	48.33 D	49.33 D	71.44 D
With Rhizo.	2 D	56.50 A	61.00 A	61.66 A	63.40 A	70.50 A	71.50 A	79.33 A	86.86 A	102.66 A
	6 D	36.66 C	42.33 C	45.59 B	51.66 B	53.43 C	60.33 C	61.27 C	74.84 B	95.00 B
	10 D	30.00 DE	33.34 D	36.90 C	42.53 C	44.62 DE	50.33 E	52.33 D	52.55 D	79.66 C
		Second season 2021								
Without Rhizo.	-	36.87 B	40.61 B	41.79 B	46.24 B	49.79 B	57.77 B	57.79 B	66.83 B	85.85 B
With Rhizo.		43.05 A	47.55 A	50.05 A	54.53 A	58.18 A	62.72 A	66.31 A	73.42 A	94.44 A
-	2 D	52.89 A	57.50 A	57.83 A	60.36 A	66.41 A	70.91 A	75.00 A	84.35 A	101.27 A
	6 D	36.83 B	41.25 B	43.34 B	50.09 B	51.08 B	60.66 B	58.82 B	73.08 B	91.61 B
	10 D	30.16 C	33.50 C	36.59 C	40.71 C	44.46 C	49.16 C	52.33 C	52.94 C	77.55 C
Without Rhizo.	2 D	47.28 B	52.00 B	52.00 B	55.32 B	60.33 B	68.33 B	68.66 B	79.83 B	97.88 B
	6 D	34.99 CD	38.18 D	39.09 C	46.51 C	46.73 D	59.00 D	54.37 D	69.33 C	86.22 C
	10 D	28.33 E	31.66 E	34.29 C	36.88 D	42.31 E	46.00 F	50.33 D	51.33 D	73.44 D
With Rhizo.	2 D	58.50 A	63.00 A	63.66 A	65.40 A	72.50 A	73.50 A	81.33 A	88.86 A	104.6 A
	6 D	38.66 C	44.33 C	47.59 B	53.66 B	55.43 C	62.33 C	63.27 C	76.84 B	97.00 B
	10 D	32.00 DE	35.34 D	38.90 C	44.53 C	46.62 DE	52.33 E	54.33 D	54.55 D	81.66 C

Rhizo. means rhizobacteria inoculation. 2D, 6D and 10D means irrigation levels D - days. Means at each column for each factor had different letters are significantly different at $p \leq 0.05$.

Table 2. Effect of rhizobacteria inoculation, irrigation intervals and their interaction on clipping dry weight of *Paspalum vaginatum* turfgrass (area of 1 m^2) during 2020 and 2021 seasons.

Rhizobacteria inoculation (Rhizo)	Irrigation	Week 1	Week 2	Week 3	Week 4	Week 5	Week 6	Week 7	Week 8	Week 9
		First season 2020								
Without Rhizo.	-	9.19B	11.18B	12.18B	13.80B	15.64B	18.67B	19.11B	22.31B	31.27B
With Rhizo.		11.46A	13.81A	15.35A	16.78A	19.02A	21.07A	22.70A	25.46A	35.68A
-	**2 D**	15.80A	18.27A	19.46A	20.33A	23.36A	25.63A	28.19A	31.85A	39.69A
	6 D	8.69B	10.96B	11.93B	14.45B	15.57B	19.08B	18.68B	23.79B	33.79B
	10 D	6.49C	8.25C	9.90C	11.09C	13.06C	14.91C	15.84C	16.01C	26.95C
Without Rhizo.	**2 D**	13.62 B	16.33 B	16.91 B	17.98 B	20.81 B	24.22 A	25.15 B	29.45 B	38.36 AB
	6 D	7.97 CD	9.70 CD	10.58 CD	13.57 C	14.00 D	18.33 BC	17.37 CD	22.29 C	30.62 C
	10 D	5.98 D	7.51 D	9.05 D	9.84 D	12.12 D	13.47 D	14.79 D	15.19 D	24.85 D
With Rhizo.	**2 D**	17.98 A	20.21 A	22.01 A	22.67 A	25.91 A	27.05 A	31.24 A	34.26 A	41.02 A
	6 D	9.41 C	12.22 C	13.28 C	15.33 BC	17.15 C	19.82 B	19.98 C	25.30 C	36.97 B
	10 D	6.99 CD	9.00 D	10.76 CD	12.33 CD	14.00 D	16.35 CD	16.89 CD	16.83 D	29.05 C
		Second season 2021								
Without Rhizo.	-	10.42B	12.42B	13.35B	15.19B	16.86B	19.93B	20.16B	23.61B	32.41B
With Rhizo.		13.11A	15.46A	16.71A	18.64A	20.66A	22.40A	24.30A	27.33A	36.94A
-	**2 D**	16.53A	19.19A	20.08A	21.15A	24.09A	26.21A	28.66A	32.66A	39.79A
	6 D	10.45B	12.61B	13.93B	16.50B	17.50B	20.93B	20.16B	25.56B	35.29B
	10 D	8.31C	10.02C	11.07C	13.09C	14.69C	16.36C	17.87C	18.20C	28.95C
Without Rhizo.	**2 D**	13.74 B	16.19 B	17.15 B	18.50 B	20.74 B	24.22 B	25.49 B	29.86 B	37.76 A
	6 D	9.91 C	11.65 CD	12.58 CD	15.23 CD	16.00 CD	20.33 C	18.14 D	23.73 C	32.62 B
	10 D	7.63 C	9.42 D	10.31 D	11.84 E	13.84 D	15.25 D	16.85 D	17.25 D	26.85 C
With Rhizo.	**2 D**	19.33 A	22.19 A	23.01 A	23.80 A	27.43 A	28.20 A	31.84 A	35.46 A	41.81 A
	6 D	11.00 BC	13.57 C	15.28 BC	17.78 BC	19.00 BC	21.54 C	22.17 BC	27.38 BC	37.97 A
	10 D	8.99 C	10.62 D	11.84 D	14.34 DE	15.54 D	17.47 D	18.89 CD	19.15 D	31.05 BC

Rhizo. means rhizobacteria inoculation. 2D, 6D and 10D means irrigation levels D - days. Means at each column for each factor had different letters are significantly different at $p \leq 0.05$.

3.2. Relative water content (RWC)

The current findings demonstrate that rhizobacteria inoculation significantly improved RWC in *Paspalum vaginatum* separately or under drought circumstances, with treated plants maintaining higher RWC values compared to those exposed only to drought in both seasons (Table 3). Contrary, increasing drought levels resulted in decreasing RWC in both seasons. During the investigated period, RWC was gradually increased from week 1 to week 7 in treated or nontreated plants. RWC dropped by (22.5%) in the first season and (18.75%) in the second season under the most severe drought condition (10-day interval without rhizobacterial inoculation). The findings demonstrated that, in dry circumstances, RWC rose in inoculated plants of turfgrass relative to non-inoculated plants (Aalipour et al., 2020). With a 15.5% increase above the control treatment, the inoculation of root-decomposing bacteria (PGPR) produced the greatest relative water content (RWC) (Gasemi et al., 2023). In this study, Paspalum plants showed a significant increase in RWC content after adding PGPR to both drought-stressed and non-stressed plants. These results are consistent with Yodphet et al. (2025) on *Andrographis paniculata* plants, which showed a significant increase of 78.75% in RWC content in plants grown under drought conditions and supplemented with microbial biofertilizer. Also, current results agree with Fadil et al. (2025) on Paspalum who showed that RWC was increased after adding PGPR.

It has been noted that PGPR causes an increase in the length and volume of roots through direct actions such the production of hormones that promote plant development, such as indole-3aceticacid (IAA) in the root environment. Moreover, root nodule production is a result of morphological induction. Hence, increasing root efficiency causes stressed plants to use water more efficiently, which raises fresh weight and RWC in tall fescue (Mahdavi et al., 2020).

In contrast, in this study drought stress reduced the relative water content of plants which support the previous reports of Jazi et al. (2019) in several turfgrass species.

Table 3. Effect of rhizobacteria inoculation, irrigation intervals and their interaction on relative water content of *Paspalum vaginatum* turfgrass during 2020 and 2021 seasons.

Rhizobacteria inoculation (Rhizo)	Irrigation	Week 1	Week 2	Week 3	Week 4	Week 5	Week 6	Week 7	Week 8	Week 9
		First season 2020								
Without Rhizo.	-	0.520B	0.53B	0.56A	0.57B	0.59B	0.60B	0.61B	0.64B	0.70B
With Rhizo.		0.570A	0.61A	0.60A	0.62A	0.65A	0.66A	0.69A	0.69A	0.75A
-	2 D	0.638A	0.65A	0.67A	0.67A	0.70A	0.70A	0.72A	0.75A	0.82A
	6 D	0.534B	0.58B	0.59B	0.60B	0.61B	0.62B	0.65B	0.65B	0.71B
	10 D	0.464B	0.48C	0.49C	0.52C	0.54C	0.57B	0.58C	0.60B	0.64C
Without Rhizo.	2 D	0.60 AB	0.58 AB	0.66 AB	0.64 AB	0.67 AB	0.66 AB	0.68 BC	0.71 AB	0.80 A
	6 D	0.50 CD	0.52 BC	0.56 BC	0.59 BC	0.58 BC	0.60 BC	0.62 CD	0.63 BC	0.69 B
	10 D	0.45 D	0.47 D	0.48 C	0.49 D	0.51 C	0.55 C	0.55 D	0.60 C	0.62 C
With Rhizo.	2 D	0.66 A	0.66 A	0.68 A	0.70 A	0.74 A	0.74 A	0.78 A	0.80 A	0.84 A
	6 D	0.56 BC	0.61 ABC	0.62 AB	0.60 BC	0.64 AB	0.64 B	0.70 AB	0.68 BC	0.73 B
	10 D	0.48 D	0.58 C	0.51 C	0.55 CD	0.58 BC	0.60 BC	0.62 CD	0.62 C	0.67 BC
		Second season 2020								
Without Rhizo.	-	0.482A	0.52B	0.56B	0.58B	0.60B	0.62B	0.63B	0.68B	0.72B
With Rhizo.		0.513A	0.58A	0.60A	0.62A	0.64A	0.66A	0.68A	0.72A	0.76A
-	2 D	0.572A	0.61A	0.65A	0.67A	0.68A	0.70A	0.72A	0.76A	0.83A
	6 D	0.488B	0.55B	0.57B	0.60B	0.62B	0.63B	0.65B	0.69B	0.73B
	10 D	0.434B	0.48C	0.52C	0.54C	0.56C	0.59C	0.61C	0.64C	0.66C
Without Rhizo.	2 D	0.535 AB	0.565 B	0.623 B	0.638 B	0.660 AB	0.665 AB	0.675 B	0.740 AB	0.80 AB
	6 D	0.484 BC	0.526 BC	0.559 CD	0.591 BCD	0.607 BCD	0.630 BC	0.642 BC	0.686 BC	0.71 BC
	10 D	0.427 C	0.473 C	0.509 D	0.533 D	0.551 D	0.584 C	0.601 C	0.629 C	0.65 C
With Rhizo.	2 D	0.608 A	0.665 A	0.690 A	0.713 A	0.719 A	0.740 A	0.772 A	0.794 A	0.87 A
	6 D	0.492 BC	0.581 B	0.599 BC	0.620 BC	0.635 BC	0.642 BC	0.662 BC	0.713 ABC	0.74 BC
	10 D	0.440 BC	0.496 C	0.535 D	0.555 CD	0.581 CD	0.599 BC	0.620 BC	0.656 BC	0.67 C

Rhizo. means rhizobacteria inoculation. 2D, 6D and 10D means irrigation levels D - days. Means at each column for each factor had different letters are significantly different at $p \leq 0.05$.

3.3. Total chlorophyll content

Table 4 presents the effects of rhizobacteria inocu-lation, drought, and their interaction on the total chlo-ro-phyll content of *Paspalum vaginatum* across two growing seasons (2020 and 2021). Data reveal signifi-cant variations in chlorophyll content under different treatments, highlighting the influence of rhizobacteria inoculation and drought stress on plant physiological responses. Rhizo-bacteria inoculation enhanced the chlorophyll content compared to the control at any time point during the investigated period, however, drought markedly reduced it in both seasons. In drought stressed plants, the chlorophyll content was improved due to rhizobacteria inoculation in both seasons. These find-ings underscore the potential of rhizobacteria inocula-tion to mitigate drought stress in *Paspalum vaginatum*. Green photosynthetic pigments called chlorophylls are present in plant leaves and green stems. They are in charge of photosynthetic processes. Stress from water negatively affects several facets of plant physiology, including photosynthetic ability. The suppression of Chl synthesis and the initiation of its breakdown as a result of the buildup of reactive oxygen species have been identified as the causes of the drop in Chl concentration under water stress (Duo et al., 2018). A greater Chl content indicates improved photosynthetic performance, which contributes to improved growth under stress, giving the growing plant that has a carbon supply and greater energy (Aalipour et al., 2019). When compared to plants grown in well-watered settings, a growing water shortage caused the plants Chl content to drop. A key determinant of photosynthetic capability in living plants is the amount of chlorophyll (Chl) in their leaves, which progressively dropped during stressful times in turfgrass spp. (Jazi et al., 2019).

Current results agree with Fadil et al. (2025) on Paspalum who showed that chlorophyll and carotenoids contente were markedly enhanced as a result of PGPR treatment. Badran et al. (2017) on Bermuda grass, ob-served similar trend relevant to biofertilizers.

Table 4. Effect of rhizobacteria inoculation, irrigation and their interaction on total chlorophyll of *Paspalum vaginatum* turfgrass during 2020 and 2021 seasons

Rhizobacteria inoculation (Rhizo)	Irrigation	Week 1	Week 2	Week 3	Week 4	Week 5	Week 6	Week 7	Week 8	Week 9
		First season 2019								
Without Rhizo.	-	1.21B	1.41B	1.49B	1.56B	1.63B	1.73B	1.82B	1.98B	2.12B
With Rhizo.		1.33A	1.50A	1.58A	1.66A	1.75A	1.85A	1.93A	2.08A	2.25A
-	**2 D**	1.46A	1.65A	1.72A	1.80A	1.92A	2.02A	2.11A	2.27A	2.44A
	6 D	1.28B	1.43B	1.51B	1.60B	1.68B	1.78B	1.88B	2.04B	2.18B
	10 D	1.07C	1.28C	1.37C	1.43C	1.48C	1.58C	1.65C	1.79C	1.94C
Without Rhizo.	**2 D**	1.41 AB	1.60 AB	1.69 AB	1.75 AB	1.86 AB	1.96 AB	2.06 AB	2.18 AB	2.36 AB
	6 D	1.24 BC	1.41 BCD	1.46 CD	1.56 C	1.63 CD	1.71 BC	1.83 BCD	1.98 BC	2.11 BCD
	10 D	0.99 D	1.22 D	1.31 D	1.37 D	1.42 E	1.54 D	1.58 D	1.77 C	1.89 D
With Rhizo.	**2 D**	1.51 A	1.70 A	1.74 A	1.86 A	1.98 A	2.08 A	2.16 A	2.35 A	2.51 A
	6 D	1.32 ABC	1.46 BC	1.56 BC	1.64 BC	1.73 BC	1.85 BC	1.93 ABC	2.10 B	2.25 ABC
	10 D	1.16 CD	1.33 CD	1.42 CD	1.48 CD	1.55 DE	1.63 D	1.71 CD	1.80 C	1.99 CD
		Second season 2020								
Without Rhizo.		1.15B	1.40B	1.48B	1.58B	1.66A	1.75B	1.83B	1.93B	2.14B
With Rhizo.		1.31A	1.46A	1.56A	1.65A	1.72A	1.82A	1.94A	2.13A	2.36A
-	**2 D**	1.41A	1.58A	1.71A	1.81A	1.89A	1.97A	2.09A	2.26A	2.58A
	6 D	1.23B	1.42B	1.51B	1.60B	1.68B	1.79B	1.89B	2.02B	2.21B
	10 D	1.05C	1.30C	1.34C	1.44C	1.50C	1.59C	1.69C	1.82C	1.96B
Without Rhizo.	**2 D**	1.34 AB	1.56 AB	1.64 AB	1.75 B	1.85 AB	1.94 A	2.03 AB	2.17 AB	2.42 AB
	6 D	1.15 C	1.40 C	1.49 CD	1.56 C	1.64 CD	1.74 BC	1.83 CD	1.91 C	2.13 BC
	10 D	0.97 D	1.25 D	1.31 E	1.43 D	1.49 D	1.56 C	1.63 E	1.71 D	1.88 C
With Rhizo.	**2 D**	1.47 A	1.59 A	1.77 A	1.87 A	1.94 A	1.99 A	2.14 A	2.34 A	2.75 A
	6 D	1.31 B	1.44 BC	1.53 BC	1.64 BC	1.72 BC	1.84 AB	1.94 BC	2.14 B	2.28 ABC
	10 D	1.13 C	1.35 CD	1.38 DE	1.45 D	1.50 D	1.62 C	1.75 DE	1.92 C	2.05 BC

Rhizo. means rhizobacteria inoculation. 2D, 6D and 10D means irrigation levels D - days. Means at each column for each factor had different letters are significantly different at $p \leq 0.05$.

3.4. Mineral content

Rhizobacteria inoculation significantly enhanced N, P and K percentages in paspalum leaves compared to the control under both non- drought and drought conditions during the 2020 and 2021 growing seasons (Table 5). However, salinity treatment markedly reduced N, P and K percent-ages compared to the unstressed plants. The lowest N, P and K percentages (1.39, 0.23 and 1.35 % in 2020, and 1.67, 0.28 and 1.69 % in 2021) were recorded in D2 treatment without rhizobacteria, respectively. However, when D2 treatment was interacted with rhizobacteria, those values were enhanced and recorded (1.52, 0.25 and 1.44 % in 2020, and 1.86, 0.31 and 1.76 % in 2021). The results of (Porcelli et al., 2024) indicate that while the imposed treatments had no effect on the concentration of N in the leaves, the concentration of P rose dramatically, confirming the intricate connections between N and P nutrition and water and salt stress. According to Li et al. (2016), Na^+ interference with K^+ and Ca^{2+} absorption is the cause of *Paspalum vaginatum*'s decrease in mineral content under drought stress. Rhizobacteria injection, however, lessened this impact, most likely by means of phosphorus solubilization, nitrogen fixation, and ion homeostasis management.

This is in line with the results of Li et al. (2022), who found that tall fescue maintained a favorable K^+/Na^+ ratio by upregulating genes involved in K^+ absorption and Na^+ exclusion by plant growth-promoting rhizobacteria such as *Bacillus zanthoxyli*. These results are in line with the larger body of research on how biofertilizers can improve mineral absorption and reduce salt stress (Fadil et al., 2025). *Paspalum vaginatum*, which is extremely salt resistant, may keep its shoots' K content high and restrict the flow of Na toward them. Furthermore, compared to its green leaves, this species' dead leaves gathered more Na and Ca (Porcelli et al., 2024).

Table 5. Effect of rhizobacteria inoculation, irrigation intervals and their interaction on mineral content of *Paspalum vaginatum* turfgrass after a 9-week investigation period during 2020 and 2021seasons.

Rhizobacteria inoculation (Rhizo)	Irrigation	N%	P%	K%	N%	P%	K %
		First season 2020			Second season 2021		
Without Rhizo.	-	1.21B	0.18B	1.27B	1.55B	0.24B	1.57B
With Rhizo.		1.32A	0.22A	1.32A	1.65A	0.30A	1.62A
-	2 D	1.45A	0.24A	1.39A	1.81A	0.30A	1.72A
	6 D	1.26B	0.20B	1.30B	1.59B	0.27A	1.60B
	10 D	1.09C	0.166	1.18C	1.39C	0.24B	1.48C
Without Rhizo.	2 D	1.39B	0.23A	1.35B	1.76B	0.28AB	1.69AB
	6 D	1.21D	0.17B	1.30B	1.55D	0.24BC	1.58CD
	10 D	1.04F	0.15B	1.16C	1.33F	0.20C	1.46E
With Rhizo.	2 D	1.52A	0.25A	1.44A	1.86A	0.31A	1.76A
	6 D	1.31C	0.23A	1.31B	1.64C	0.30A	1.62BC
	10 D	1.15E	0.17B	1.20C	1.46E	0.28AB	1.49DE

Rhizo. means rhizobacteria inoculation. 2D, 6D and 10D means irrigation levels D - days. Means at each column for each factor had different letters are significantly different at $p \leq 0.05$.

4. Conclusion

The findings of this study clearly demonstrate the significant role of plant growth-promoting rhizobacteria (PGPR) in alleviating the adverse effects of drought stress on *Paspalum vaginatum*, a turfgrass species valued for its resilience and adaptability. Water deficit stress substantially reduced plant performance, as evidenced by decreased biomass production, relative water content, chlorophyll and carotenoid levels, and macronutrient uptake. Additionally, drought induced the accumulation of oxidative stress markers such as proline and peroxidase activity, reflecting physiological disruption. However, PGPR inoculation significantly improved the growth and physiological status of turfgrass under both moderate and severe drought conditions which enhanced their capacity to tolerate and recover from water scarcity. The ability of PGPR to sustain turf vigor under drought highlights its potential as a sustainable, eco-friendly biofertilization strategy in water-limited environments.

Author Contributions: "Conceptualization, F.H. and M.R.; methodology, M.R. and M.F.; software, F.H.; validation, M.R., R.E. and M.F.; formal analysis, F.H.; investigation, M.R.; resources, M.R.; data curation, M.F.; writing—original draft preparation, F.H.; writing—review and editing, F.H. and R.E.; visualization, M.F.; supervision, F.H. and R.E. All authors have read and agreed to the published version of the manuscript.

References

Aalipour, H., Nikbakht, A., Ghasemi, M., and Amiri, R. (2020). Morpho-physiological and biochemical responses of two turfgrass species to rbuscular mycorrhizal fungi and humic acid under water stress conditions. Journal of Soil Science and Plant Nutrition, 20(2), 566-576.

AOAC. (1995). Official Methods of Analysis, 16th Ed., Association of Official Analytical Chemists Intern trigonal, Arlington, Virginia, USA.

Ali. A.F, Abdou. M.A.H, Amer. E.H. and Ammar. H.A.E.I. 2018. Influence of compost, mineral and effective microorganisms' application on sandy soil-grown Bermuda turfgrass. Scientific J. Flowers & Ornamental Plants. 2356-7864.

Jackson, M.L. (1967). Soil Chemical Analysis. Prentice-Hall of India Private Limited, New Delhi.

Baez Rogelio, A.; Morales García, Y. E.; Quintero Hernández, V.; and Muñoz-Rojas, J. (2017). Next generation of microbial inoculants for agriculture and bioremediation. Microbial Biotechnology, 10(1), 19–21.

Badran. M.K.A. and Abd Ellatif. M.Z. (2009). Response of Nigella sativa L. plants to plant density and mineral / biofertilization treatments. Egypt. J. Agri. Res., 87 (1).

Breuste, J.; Haase, D.; and Elmqvist, T. (2013). Urban landscapes and ecosystem services. In: Wratten, S.; Sandhu, H.; Cullen, R.; and Costanza, R. (Eds.), Ecosystem services in agricultural and urban landscapes (pp. 83–104). John Wiley & Sons, Ltd, Chichester.

Chen, Z.; Kim, W.; Newman, M.; Wang, M.; and Raymer, P. (2005). Molecular characterization of genetic diversity in the USDA seashore paspalum germplasm collection. International Turfgrass Society, 10, 543–549.

Poorter, H.; Niklas, K.J.; Reich, P. B.; Oleksyn, J.; Poot, P.; and Mommer, L. (2012). Biomass allocation to leaves, stems and roots: meta-analyses of interspecific variation and environmental control. New Phytologist, 193(1), 30–50.

Dere, Ş.; Güneş, T.; and Sivaci, R. (1998). Spectrophotometric determination of chlorophyll-A, B and total carotenoid contents of some algae species using different solvents. Turkish Journal of Botany, 22(1), 13–18

Duncan, R. R., and Carrow, R. N. (2000). Seashore paspalum: The environmental turfgrass. John Wiley & Sons.

Duo, L. A., Liu, C. X., and Zhao, S. L. (2018). Alleviation of drought stress in turfgrass by the combined application of nano-compost and microbes from compost. Russian Journal of Plant Physiology, 65, 419-426.

Eida, A. A.; Bougouffa, S.; L'Haridon, F.; Alam, I.; Weisskopf, L.; Bajic, V. B.; Saad, M. M.; and Hirt, H. (2020). Genome insights of the plant-growth promoting bacterium Cronobacter muytjensii JZ38 with volatile-mediated antagonistic activity against Phytophthora infestans. Frontiers in Microbiology, 11, 369.

El-Tayeh, N. A., Galal, H. K., Soliman, M. I., and Zaki, H. (2020). Association of Morphological, Ecological, and Genetic Diversity oi Aerva javanica Populations Growing in the Eastern Desert of Egypt. Agronomy, 10(3), 1cj-1cj.

Fadil, S., Hassan, F., Fetouh, M., El-Serafy, R., and Moussa, M. (2025). Rhizobacteria Inoculation Enhanced the Growth Performance of Paspalum Turfgrass under Saline Conditions. Journal of Sustainable Agricultural and Environmental Sciences, 4(2), 8-16.

Farooq, M., Hussain, M., Wahid, A., Siddique, K. (2012). Drought stress in plants: an overview. In: Plant Responses to Drought Stress, pp. 1–33.

Gasemi, S.; Mahdavikia, H.; Rezaei-Chiyaneh, E.; Banaei-Asl, F.; Dolatabadian, A.; and Sadeghpour, A. (2023). Co-inoculation of mycorrhizal fungi and plant growth-promoting rhizobacteria improve growth, biochemical and physiological attributes in Dracocephalum kotschyi Boiss. under water deficit stress. PeerJ, 11, e16474.

Jazi, Z. G., Etemadi, N., and Aalipour, H. (2019). The physiological responses of four turfgrass species to drought stress. Advances in Horticultural Science, 33(3), 381-390.

Jiang, Y. and Huang, B. (2001). Drought and heat stress injury of two cool-season turfgrasses in relation to antioxidant metabolism and lipid peroxidation. Crop Sci. 41:436-442.

Mittler, R. (2002). Oxidative stress, antioxidants and stress tolerance. Trends in plant science, 7(9), 405-410.

Mazrou, R.M.; Hassan, F.A.S.; Mansour, M.M.F.; Moussa, M.M. (2023). Melatonin Enhanced Drought Stress Tolerance and Productivity of *Pelargonium graveolens* L. (Herit) by Regulating Physiological and Biochemical Responses. Horticulturae 9, 1222. https://doi.org/10.3390/horticulturae9111222

Nelson, D.W. and Sommers, L.E. (1973). Determination of total nitrogen in plant material. Agronomy Journal, 65(1), 109–112.

Porcelli, C. A., Rubio, G., Gutiérrez Boem, F. H., and Lavado, R. S. (2024). The effect of water and salt stress on Paspalum dilatatum, a constituent of pampas natural grasslands. Tech Science Press; Phyton; 93; 8; 8-2024; 2009-2018.

Ragályi, P., Szabó, A., Rékási, M., Csathó, P., and Csontos, P. (2023). Effect of different macronutrient supply levels on the drought tolerance of rainfed grass based on biomass production, water use efficiency and macroelement content. Horticulturae, 9(12), 1337.

Rue, K.; and Zhang, Q. (2020). Kentucky bluegrass growth under saline, waterlogging, and saline-waterlogging conditions during germination and seedling growth. Crop, Forage and Turfgrass Management, 6(1), e20002.

Trenholm, L. E., Unruh, J. B., and Sartain, J. B. (2012). Nitrate leaching and turf quality in established 'Floratam'St. Augustinegrass and 'Empire'zoysiagrass. Journal of Environmental Quality, 41(3), 793-799.

Vurukonda, S. S. K. P., et al. (2016). Plant growth promoting bacteria in drought stress. Frontiers in Plant Science.

Yodphet, B. Riddech, N. Kaewpradit, W. Roytrakul, S. Boonlue, S. Jangpromma, N. 2025. Effect of microbial biofertilizer on proteomic profiling, antioxidant enzyme and andrographolide content in Andrographis paniculata Burm.f Nee. under drought stress. Plant Stress 16 (2025) 100817.

Weatherley, P.E. (1950). Studies in the water relations of the cotton plant. I. The field measurement of water deficits in leaves. New Phytologist, 49, 81–97.