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Performance of soybeans inoculated with multifunctional microorganisms under water-deficit stress

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Abstract

Soybeans (*Glycine max* (L.) Merr.) are mainly grown in Brazil during the rainy season. However, there are typically periods of rainfall deficiency, which causes water-deficit stress to the crop. Plant growth-promoting rhizobacteria (PGPR) can help alleviate these stresses by inducing water deficit tolerance. The objective of this study was to evaluate the role of PGPR in enhancing soybean tolerance to water-deficit stress. Six PGPR isolates, two for induction of water-deficit tolerance (ESA 441, BRM 034008), two AIA-producing (Ab-V5, BRM 063574), and two phosphate solubilizing (BRM 063573, BRM 67205), and their combination were evaluated, for a total of 16 treatments. The experiment was conducted in a greenhouse using a randomized block design with three replicates. Effects were measured on gas exchange parameters (stomatal conductance, transpiration, internal CO_2 concentration, and photosynthetic rate), growth parameters (shoot dry weight, root dry weight, root length, root surface area, root diameter, and root volume), and yield components (pod weight, number of pods, number of grains, and grain weight). Co-inoculation significantly reduces the effects of water stress on gas exchange, plant growth, and productivity compared to single inoculation. Notable combinations, such as BRM 063574 + BRM 67205 + BRM 034008 and BRM 063574 + BRM 063573 + ESA 441, improved root and shoot growth under stress conditions. Yield components also improved with co-inoculations, with combinations such as BRM 063574 + BRM 67205 + ESA 441 showing the highest efficacy. These results suggest that specific PGPR co-inoculations can improve soybean resilience to water deficit stress and promote better growth and yield.

Abbreviations: BNF, biological nitrogen fixation; DAE, days after emergence; GW, grain weight; NG, number of grains; NP, number of pods; PGPR, plant growth-promoting rhizobacteria; PW, pod weight; RD, root diameter; RDW, root dry weight; RL, root length; RSF, root surface area; RV, root volume; SDW, shoot dry weight.

Plain Language Summary

Soybean is an important crop grown worldwide, especially in Brazil, the United States, and Argentina. In Brazil, soybeans are often grown in dry areas and need special bacteria to help them get nutrients like nitrogen and phosphorus. These bacteria can also help soybeans grow better during droughts, which are periods without enough rain. The main goal of this research was to see if using multiple types of helpful bacteria together can improve soybean plants' growth and grain yield under water shortage conditions. The study found that using a mix of bacteria (co-inoculation) helped soybean plants keep their gas exchange (related to photosynthesis), grow stronger roots and shoots, and produce more, even during water shortages. Co-inoculation treatments improved plant health more than using just one type of bacteria. This approach is low-cost and easy for farmers to adopt, making soybean farming more sustainable in areas with water limitations.

1 | INTRODUCTION

Soybean (*Glycine max* (L.) Merr.) is one of the most important agricultural crops and a commodity of significant global economic importance, generating employment and income. Among the world's largest exporters are Brazil, the United States, and Argentina, accounting for 52.52%, 29.88%, and 6.43% of total exports, respectively (CONAB, 2023; Proque, 2019). In Brazil, soybean is grown in the whole country, particularly in the Central-West region, where soils are characterized by low fertility and high acidity, requiring fertilization and liming. In the Cerrado region, soybean cultivation takes place mainly from October to January, commonly referred to as the "water harvest," concentrating about 54% of the national production (CONAB, 2023).

Soybeans obtain >80% of the nitrogen (N) required for their development through biological nitrogen fixation (BNF) and do not require nitrogen fertilization in Brazil (Ciampitti & Salvagiotti, 2018). The use of inoculants with efficient bacteria has been propagated in Brazil since the 1950s (Siqueira et al., 2014), resulting in one of the most consolidated markets in the world for *Bradyrhizobium* inoculants recommended for this crop. Besides BNF, advances in studies of plant growth-promoting rhizobacteria (PGPR) have shown great success in their use for production of phytohormones, phosphorus (P) mineralization, and tolerance to salinity and drought, resulting in better plant development and increased productivity by mitigating adverse effects caused by biotic and abiotic factors (Barbosa et al., 2014).

Among farmers, the co-inoculation of *Bradyrhizobium* with PGPR, such as the *Azospirillum brasilense* strains Ab-V5 and Ab-V6, is already well established in soybean cultivation. The use of co-inoculation on farms has increased every year, as co-inoculated soybean plants show more abundant nodulation and increased yields compared to inoculation with *Bradyrhizobium japonicum* alone (Prando et al., 2024). This is because *Azospirillum* spp. are efficient producers of phytohormones, such as auxins (Bulegon et al., 2016; Cassán et al., 2014; Masciarelli et al., 2013) and gibberellins (Lenin & Jayanthi, 2012). These phytohormones contribute to improved plant development by establishing a more vigorous root system, providing greater nutrient uptake, and increasing tolerance to water stress (Grover et al., 2021; Jan et al., 2024; Kumar & Verma, 2018).

Regarding P, low soil P availability affects soybean production because P tends to be adsorbed on soil colloid surfaces or forms phosphate precipitates (Hong et al., 2021; Kamran et al., 2022). Thus, plants need to extract phosphorus from the soil solution to obtain nutrients, although phosphorus is usually retained in the solid phase (Almeida et al., 2019). According to Tian et al. (2022), there is a need to develop studies relating the phosphorus nutrient present in the soil with bacterial communities in the rhizosphere to identify the possibility of reducing its use.

In Brazil, soybean is grown primarily during the rainy season, when there are typically periods of 15–20 days without rain, causing water-deficit stress to the crop, the main cause of reduced productivity (Battisti & Sentelhas, 2014). Recent research on rhizobacteria has shown that inoculated plants exhibit increased drought tolerance through morphological and biochemical changes that result in greater root-soil contact, improving water and nutrient uptake (Ahluwalia et al., 2021; Ahmad et al., 2022; Jabborova et al., 2021).

Although there are reports of studies on the effect of PGPR inoculation on soybean crops, information on the use of multiple microorganisms (co-inoculation) combining more than two growth-promoting mechanisms is rather scarce, especially under water limitation. Therefore, this work aimed to evaluate the effect of co-inoculation with PGPR on the growth and productivity of soybean plants under water-deficit stress.

2 | MATERIALS AND METHODS

2.1 | Location of the study and soil used in the experiment

The study was conducted under greenhouse conditions at Embrapa Rice and Beans, Santo Antônio de Goiás, GO (16°30′06.40″ S; 49°16′54.56″ W). Throughout the experiment, temperature and relative humidity averaged approximately 25°C and 65%, respectively. The soil used in the experiment was subjected to chemical analysis according to Teixeira et al. (2017), and the results are presented in Table 1.

2.2 | Microorganisms used, treatments, and experimental design

The microorganisms evaluated were 1- *Azospirillum brasilense*, strain Ab-V5 (Hungria et al., 2018), 2- *Stenotrophomonas maltophilia*, strain BRM 063574 (Asobia et al., 2025), 3- *Rhizobium sp.*, strain BRM 034008 (Fernandes-Júnior et al., 2015), 4- ESA 441 (Silva, 2017), 5- *Bacillus pumilus*, strain BRM 063573 (Asobia et al., 2025), and 6- *Paenibacillus pabuli*, strain BRM 67205 (Asobia et al., 2025). These microorganisms have the following mechanisms of plant growth promotion: (1) production of indole-3-acetic acid (Ab-V5 and BRM 063574), (2) induction of tolerance to water-deficit stress (BRM 034008 and ESA 441), and (3) phosphate solubilization (BRM 063573 and BRM 67205).

Treatments included individual inoculation of each bacterium, combinations among them, and two control treatments, totaling 16 treatments, described as follows: T1 = BRM 063574+BRM 67205+ESA 441, T2 = BRM 063574+BRM 67205+BRM 034008, T3 = BRM 063574+BRM 063573+ESA 441, T4 = BRM 063574+BRM 063573+BRM 034008, T5 = Ab-V5+BRM 67205+ESA 441, T6 = Ab-V5+BRM 67205+BRM 034008, T7 = Ab-V5+BRM 063573+ESA 441, T8 = Ab-V5+BRM 063573+BRM 034008, T9 = BRM 063574, T10 = Ab-V5, T11 = BRM 67205, T12 = BRM 063573, T13 = ESA 441, T14 = BRM 034008, T15 = absolute control (without both fertilizer and microorganisms), and T16 = fertilized control (according to the soil analysis and technical recommendation for the crop). The combinations of microorganisms were selected based on previous results (Asobia et al., 2025). The experiment was conducted in a completely randomized block design with three replications, using the SITIS platform (Sistema Integrado para Tratamento Induzido de Seca), a Platform of Plant Phenotyping for Drought Tolerance (Pereira et al., 2017).

Core Ideas

- Co-inoculation improves drought resilience more than single inoculation in soybean.
- Specific microbial combinations optimize growth and yield of soybean under stress.
- Gas exchanges are stabilized by targeted co-inoculant mixtures.

2.3 | Preparation, implementation, and conduction of the experiment

At the sowing time, a fertilization of 625 mg kg^{-1} of soil of the formulation 5-30-15 (NPK) was performed, defined according to the soil chemical analysis (Table 1) and technical recommendations for soybean cultivation. In treatments using phosphate-solubilizing bacterial isolates (T1, T2, T3, T4, T5, T6, T7, T8, T11, and T12), half the phosphorus dose used in the fertilized control (T16) was applied, adding 312.5 mg kg^{-1} of soil from the 5-30-15 formulation. In previous studies involving maize (de Oliveira-Paiva et al., 2024) and common bean (Bittencourt et al., 2024), using half the recommended dose of phosphorus fertilizer and phosphate-solubilizing microorganisms has resulted in an equal or superior grain yield to using the full recommended dose. Thus, in our study, we attempted to replicate these results. The soil was used to fill the columns of the SITIS platform, with two columns prepared for each treatment; one was collected when the plants reached flowering, and the other at the end of the crop cycle.

The BRSMG 534 variety was used, which is a conventional early-cycle soybean cultivar with an indeterminate growth habit. The seeds were disinfected using 70% alcohol for 30 s, 2% sodium hypochlorite for 2 min, followed by 10 washes with autoclaved distilled water. After superficial disinfection, the seeds were inoculated with a commercial inoculant for BNF. Subsequently, six seeds were placed per column with a distance of 2 cm from the central wall of the transparent acrylic tube used for imaging with the scanner.

Thinning was performed 8 days after emergence (DAE), leaving three plants per pot. The roots were carefully exposed using a spatula to avoid damaging them, followed by inoculation with 0.8 mL of each isolate at a concentration of 1×10^9 cells mL^{-1}. The suspension was applied directly to the exposed roots, and the soil was then replaced to cover them.

From inoculation onward, the columns were maintained at 80% of field capacity, with the soil water content being adjusted gravimetrically by weighing the columns daily on a precision scale. At stage R3, when pod growth begins, most sensitive period for water scarcity (Poudel et al., 2023),

TABLE 1 Chemical characteristics of the soil used in the experiment.

pH	Ca ($cmol_c\ dm^{-3}$)	Mg $cmol_c\ dm^{-3}$	Al $cmol_c\ dm^{-3}$	H + Al $cmol_c\ dm^{-3}$	P ($mg\ dm^{-3}$)	K ($mg\ dm^{-3}$)	SB ($cmol_c\ dm^{-3}$)	CEC ($cmol_c\ dm^{-3}$)	BS (%)	OM ($g\ kg^{-1}$)
5.4	5.0	1.1	0.0	2.6	39.0	260	6.78	9.38	72.3	20.0

Note: BS bases saturation = ((K + Ca + Mg)/Tcec) × 100, where Tcec = K + Ca + Mg + total acidity at pH 7.0 (H + Al). Granulometric properties in gram per kilogram: 420 (clay), 70 (silt), and 510 (sand).

Abbreviations: CEC, cation exchange capacity; OM organic matter; SB, sum of bases.

the irrigation was interrupted for 15 days before resuming the water supply. Seven days after returning irrigation, the plants of one of the two columns from each treatment were analyzed and collected. Plant of the other column was maintained until physiological maturity (R7).

During crop management, weeds were manually removed. Two applications of triflumizole (1 g L^{-1}) were made at 10-day intervals up to 40 DAE to control powdery mildew, and one application of imidacloprid (2.5 mL L^{-1}) and beta-cyfluthrin (2.5 mL L^{-1}) was made at 45 DAE to control whiteflies.

2.4 | Determination of physiological parameters, growth, and yield components

The effect of water-deficit stress application on net photosynthesis rate (A), transpiration rate (E), and internal CO_2 concentration (Ci) was determined using the infrared gas-analyzer (LCpro-SD Inc.), with a leaf chamber of 6.25 cm^2, and a light pulse of 1200 µmol m^{-2} s^{-1} for 1 s. Three determinations were made: the first immediately before the onset of water-deficit stress (when the plant was turgid); the second 7 days after the onset of stress; and the third 15 days after the onset of stress.

At stage R3, the plants were cut close to the soil surface; then the CI - 600 Cano Scan root scanner (CID Bio-Science Version 3.1.19) was used, which was inserted into the acrylic tube at a depth of 0–20 cm to obtain images of the roots in 360°. The images were analyzed using WinRHIZO software to determine root length (RL), root surface area (RSA), root diameter (RD), and root volume (RV).

After obtaining the images, the roots were carefully removed from the columns and washed. The roots and shoots were placed in paper bags and dried in an air circulation oven (65°C; 72 h). After drying, roots and shoots were weighed to determine, respectively, root dry weight (RDW) and shoot dry weight (SDW).

Upon physiological maturity (R7 stage), the pods were harvested, dried in a circulating air oven (72°C; 72 h), weighed, and counted to determine pod weight (PW) and number of pods (NP). The pods were threshed, and the seeds were counted and weighed to determine the number of grains (NG) and grain weight (GW).

2.5 | Statistical analysis

Data were first checked for normality using the Shapiro–Wilk test and for homogeneity of variances using Levene's test, followed by analysis of variance (ANOVA), considering the inoculation, different times of stress, and combinations as fixed variables. When statistical differences between treatments were confirmed by the *F*-test ($p \leq 0.05$), means were compared by the Skott–Knott test ($p < 0.05$) using SISVAR software (Ferreira, 2019). Principal components analysis (PCA) was performed using the statistical packages Paleontological Statistics version 4.10.

3 | RESULTS

3.1 | Effect of inoculation on gas exchanges of soybean subjected to water-deficit stress

The gas exchanges of soybean inoculated with multifunctional microorganisms and subjected to water-deficit stress were affected by inoculation, different stress durations, and the interaction between these two factors (Table 2).

Stomatal conductance (gs) was different between treatments only 14 days from the onset of stress and remained higher in plants from treatments T2 (BRM 063574+BRM 67205+BRM 034008), T3 (BRM 063574+BRM 063573+ESA 441), T4 (BRM 063574+BRM 063573+BRM 034008), and T7 (Ab-V5+BRM 063573+ESA 441), which differed from other treatments ($p < 0.05$) (Table 3).

However, the decrease in E occurred in all treatment plants as early as day 7 and was even more intense by day 14 (Table 3). The treatments with the smallest reductions were the co-inoculated T1 (BRM 063574+BRM 67205+ESA 441), T2 (BRM 063574+BRM 67205+BRM 034008), T3 (BRM 063574+BRM 063573+ESA 441), T4 (BRM 063574+BRM 063573+BRM 034008), T5 (Ab-V5+BRM 67205+ESA 441), T6 (Ab-V5+BRM 67205+BRM 034008), T7 (Ab-V5+BRM 063573+ESA 441), and T15 (Absolute Control), demonstrating greater tolerance ($p < 0.05$) compared to other treatments. While the Absolute Control (T15) exhibited statistically similar results to the best treatments, water-deficit stress led to approximately a 20% reduction in

TABLE 2 Variance analysis for stomatal conductance (gs = mol m^{-2} s^{-1}), transpiration rate (E = mmol m^{-2} s^{-1}), internal CO$_2$ concentration (Ci = μmol mol^{-1}), and net photosynthetic rate (A = μmol m^{-2} s^{-1}) in inoculated soybean subjected to water-deficit stress.

Source of variation	*df*	gs	E	Ci	A
Treatment (T)	15	0.01**	3.31**	7613.57**	30.06**
Stress time (ST)	2	0.15**	208.8**	147090.53**	937.96**
T × ST	30	0.00**	1.31**	8797.88**	21.79**
CV (%)		14.22	12.81	7.51	25.83

Abbreviations: CV, coefficient of variation; DF, degrees of freedom.

* and ** denote significant by the *F* test at 5% and 1% of significance, respectively.

E for that treatment compared to the average of the other best treatments.

As the stress time increased from 7 to 14 days, the internal concentration of CO$_2$ within the substomatal chamber (Ci) increased or remained for all treatments (Table 3). The treatments with the lowest Ci at 14 days of stress differ significantly from the others, such as T1 (BRM 063574+BRM 67205+ESA 441), T2 (BRM 063574+BRM 67205+BRM 034008), T3 (BRM 063574+BRM 063573+ESA 441), T4 (BRM 063574+BRM 063573+BRM 034008), T5 (Ab-V5+BRM 67205+ESA 441), T6 (Ab-V5+BRM 67205+BRM 034008), T7 (Ab-V5+BRM 063573+ESA 441), T9 (BRM 063574), and T11 (BRM 67205). All these, except T9, exhibited at 14 days of stress the highest net photosynthetic rates (A), differing significantly from the others.

3.2 | Effect of inoculation on the growth of soybean subjected to water-deficit stress

The parameters related to the growth of soybean (SDW, RDW, RL, RSA, RD, and RV) subjected to water-deficit stress were affected by inoculation with multifunctional microorganisms (Table 4).

The highest values of SDW were observed in T1 (BRM 063574+BRM 67205+ESA 441), T3 (BRM 063574+BRM 063573+ESA 441), and T14 (BRM 034008), which also showed high values of RDW (Figure 1). High RDW was also detected in T4 (BRM 063574+BRM 063573+BRM 034008), T6 (Ab-V5+BRM 67205+BRM 034008), T7 (Ab-V5+BRM 063573+ESA 441), T10 (Ab-V5), and T12 (BRM 063573).

For RL parameter, treatments that showed better values were T10 (Ab-V5) and T12 (BRM 063573). Root surface area (RSA) and RD showed significant differences only for treatments T10 (Ab-V5) and T1 (BRM 063574+BRM 67205+ESA 441), respectively. In RV, treatments T5 (Ab-V5+BRM 67205+ESA 441), T6 (Ab-V5+BRM 67205+BRM 034008), T10 (Ab-V5), T12 (BRM 063573), T15 (Absolute Control), and T16 (Fertilized Control) had significant differences compared to other treatments. Among the parameters of root system development (RL, RSA, RD, and RV), strain Ab-V5 of *A. brasilense* was present in the best results of three parameters (RL, RSA, and RV).

3.3 | Effect of inoculation on yield components and grain production of soybean subjected to water-deficit stress

The parameters related to the productivity of soybean (PW, NP, NG, and GW) subjected to water-deficit stress were affected by inoculation with multifunctional microorganisms (Table 5).

In terms of PW, treatments T3 (BRM 063574+BRM 063573+ESA 441) and T14 (BRM 034008) produced the highest values. Treatments T3 (BRM 063574+BRM 063573+ESA 441), T11 (BRM 67205), and T14 (BRM 034008) showed significant results for NP and NG parameters (Figure 2).

For GW, significant differences were observed for the treatments T1 (BRM 063574+BRM 67205+ESA 441), T3 (BRM 063574+BRM 063573+ESA 441), T5 (Ab-V5+BRM 67205+ESA 441), T6 (Ab-V5+BRM 67205+BRM 034008), T8 (Ab-V5+BRM 063573+BRM 034008), T9 (BRM 063574), T10 (Ab-V5), T11 (BRM 67205), T12 (BRM 063573), T13 (ESA 441), T14 (BRM 034008), and T16 (Fertilized Control). It can be observed that for the four evaluated parameters of yield components, the best results were obtained from treatments T3 (BRM 063574+BRM 063573+ESA 441) and T14 (BRM 034008), showing a positive response in the production of soybean under water-deficit stress conditions (Figure 2).

3.4 | Principal component analysis

The principal component analysis (Figure 3) shows a clear effect between single inoculation and co-inoculation, as they are on opposite sides of the *X* axis. The co-inoculation treatments had a greater effect on the parameters A and gs, determined after 7 and 14 days of water-deficit stress, and on E, determined after 14 days of water-deficit stress, as well as on the parameters NG, NP, and SDW. On the other hand, the single inoculation treatments had a large effect on the other parameters evaluated, but they were grouped together with the absolute control and fertilized treatments.

Overall, single inoculation stood out in yield components related to production and plant development; however,

TABLE 3 Analysis of the interaction between treatments and stress times (days) for stomatal conductance (gs = mol m^{-2} s^{-1}), transpiration rate (E = mmol m^{-2} s^{-1}), internal CO_2 concentration (Ci = µmol mol^{-1}), and net photosynthetic rate (A = µmol m^{-2} s^{-1}) in inoculated soybean subjected to water-deficit stress.

	gs			E			Ci			A		
	Days of stress			Days of stress			Days of stress			Days of stress		
Treatments	0	7	14	0	7	14	0	7	14	0	7	14
T1	0.20bA	0.21bA	0.14bB	6.55bA	2.93aB	2.43aB	216.33aA	221.00bA	237.67dA	13.87aA	14.18aA	8.48aB
T2	0.23aA	0.22bA	0.20aA	7.11aA	3.04aB	3.44aB	203.67aB	220.67bB	273.67dA	16.06aA	15.30aA	9.73aB
T3	0.25aA	0.28aA	0.20aB	7.38aA	3.25aB	3.13aB	203.00aB	255.00aA	250.00dA	10.33aA	15.00aA	11.58aA
T4	0.20bA	0.22bA	0.20aA	6.44bA	3.04aB	2.78aB	209.33aB	278.33aA	258.00dA	12.55aA	11.44aA	10.82aA
T5	0.11dC	0.23aA	0.16bB	4.36dA	3.16aB	2.70aB	218.67aB	260.67aA	277.33dA	7.59aB	13.79aA	7.78aB
T6	0.22aA	0.22bA	0.15bB	6.97aA	3.43aB	2.64aB	208.00aB	269.33aA	264.00dA	15.87aA	13.86aA	7.32aB
T7	0.16cB	0.25aA	0.18aB	5.65cA	3.40aB	2.95aB	220.00aB	268.00aA	244.33dA	11.71aA	15.09aA	11.19aA
T8	0.24aA	0.19bA	0.06dB	7.11aA	2.99aB	1.39bC	228.33aB	240.67bB	334.67cA	12.80aA	14.66aA	3.43bB
T9	0.15cB	0.22bA	0.07dC	6.28bA	3.17aB	1.61bC	232.67aB	272.67aA	273.67dA	11.29aA	13.32aA	4.68bB
T10	0.20bA	0.23bA	0.06dB	6.60bA	3.01aB	1.37bC	226.67aB	204.00bB	371.00bA	13.05aA	13.80aA	0.25bB
T11	0.16cB	0.21bA	0.09cC	6.11bA	2.92aB	1.93bC	180.00aB	266.00aA	242.67dA	14.12aA	12.35aA	6.57aB
T12	0.12dB	0.26aA	0.02eC	4.71dA	3.63aB	0.46cC	213.00aB	247.67aB	402.00bA	11.39aA	14.37aA	0.34bB
T13	0.11dA	0.11cA	0.02eB	4.28dA	2.03aB	0.51cC	205.33aC	255.67aB	390.00bA	11.55aA	11.04aA	0.40bB
T14	0.17cA	0.19bA	0.04eB	5.39cA	3.08aB	1.06bC	189.00aB	223.67bB	370.33bA	14.47aA	14.57aA	1.40bB
T15	0.19bB	0.25aA	0.11cC	6.53bA	3.29aB	2.30aC	200.67aC	264.00aB	567.67aA	13.24aA	12.14aA	2.31bB
T16	0.13dB	0.19bA	0.05dC	5.17cA	3.28aB	1.38bC	197.67aC	263.00aB	348.67cA	10.82aA	11.67aA	0.62bB
Average	0.18	0.22	0.11	6.04	3.10	2.01	209.52	250.65	319.11	12.54	13.54	5.43

Note: Means followed by the same lowercase letter in the column or uppercase letter in the row, do not differ from each other by the Scott–Knott test. T1 = BRM 063574+BRM 67205+ESA 441; T2 = BRM 063574+BRM 67205+BRM 034008; T3 = BRM 063574+BRM 063573+ESA 441; T4 = BRM 063574+BRM 063573+BRM 034008; T5 = Ab-V5+BRM 67205+ESA 441; T6 = Ab-V5+BRM 67205+BRM 034008; T7 = Ab-V5+BRM 063573+ESA 441; T8 = Ab-V5+BRM 063573+BRM 034008; T9 = BRM 063574; T10 = Ab-V5; T11 = BRM 67205; T12 = BRM 063573; T13 = ESA 441; T14 = BRM 034008; T15 = absolute control; and T16 = fertilized control.

Abbreviation: CV, coefficient of variation.

TABLE 4 Variance analysis for shoot dry weight (SDW = g plant^{-1}), root dry weight (RDW = g plant^{-1}), root length (RL = cm plant^{-1}), root surface area (RSA = cm^2 plant^{-1}), root diameter (RD = mm plant^{-1}), and root volume (RV = mm^3 plant^{-1}) of soybean co-inoculated with plant growth-promoting rhizobacteria (PGPRs) under water-deficit stress.

Source of variation	*df*	SDW	RDW	RL	RSA	RD	RV
Treatment	25	2.15*	0.97**	21.24 **	18.51**	10.35**	9.70**
CV (%)		25.09	3.57	14.93	11.22	17.06	24.89

Abbreviations: CV, coefficient of variation; DF, degrees of freedom.
* and * denote significant by the *F* test at 5% and 1%, respectively.

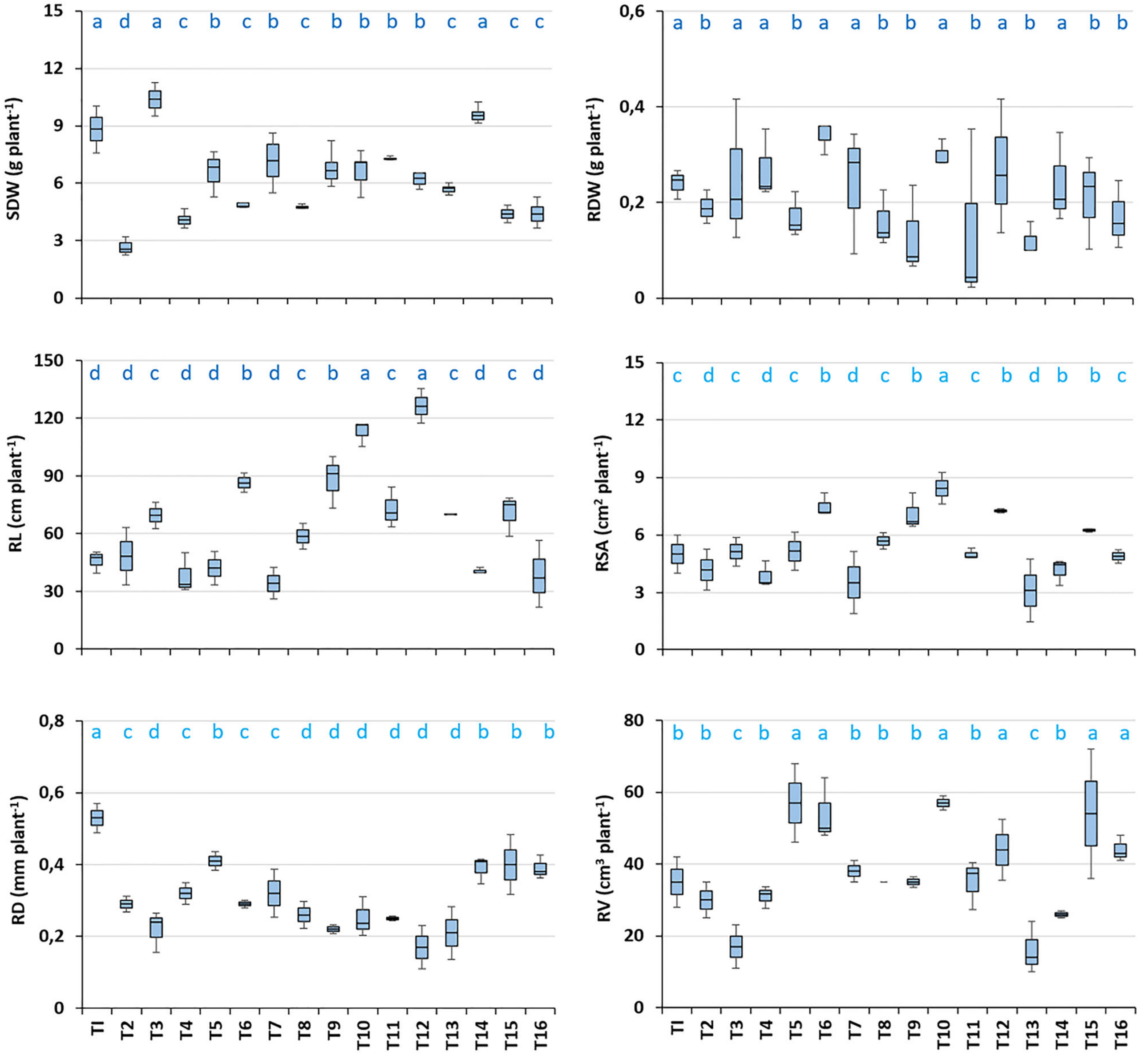

FIGURE 1 Boxplot and Scott–Knott test for shoot dry weight (SDW, g plant^{-1}), root dry weight (RDW – g plant^{-1}), root length (RL – cm plant^{-1}), root surface area (RSA – cm^2 plant^{-1}), root diameter (RD – mm plant^{-1}), and root volume (RV – mm^3 plant^{-1}) of soybean co-inoculated with plant growth-promoting rhizobacteria (PGPRs) under water-deficit stress. Treatments: T1 = BRM 063574+BRM 67205+ESA 441; T2 = BRM 063574+BRM 67205+BRM 034008; T3 = BRM 063574+BRM 063573+ESA 441; T4 = BRM 063574+BRM 063573+BRM 034008; T5 = Ab-V5+BRM 67205+ESA 441; T6 = Ab-V5+BRM 67205+BRM 034008; T7 = Ab-V5+BRM 063573+ESA 441; T8 = Ab-V5+BRM 063573+BRM 034008; T9 = BRM 063574; T10 = Ab-V5; T11 = BRM 67205; T12 = BRM 063573; T13 = ESA 441; T14 = BRM 034008; T15 = absolute control; and T16 = fertilized control. Groups labeled by the same letter did not differ significantly by the Scott–Knott test ($p < 0.05$).

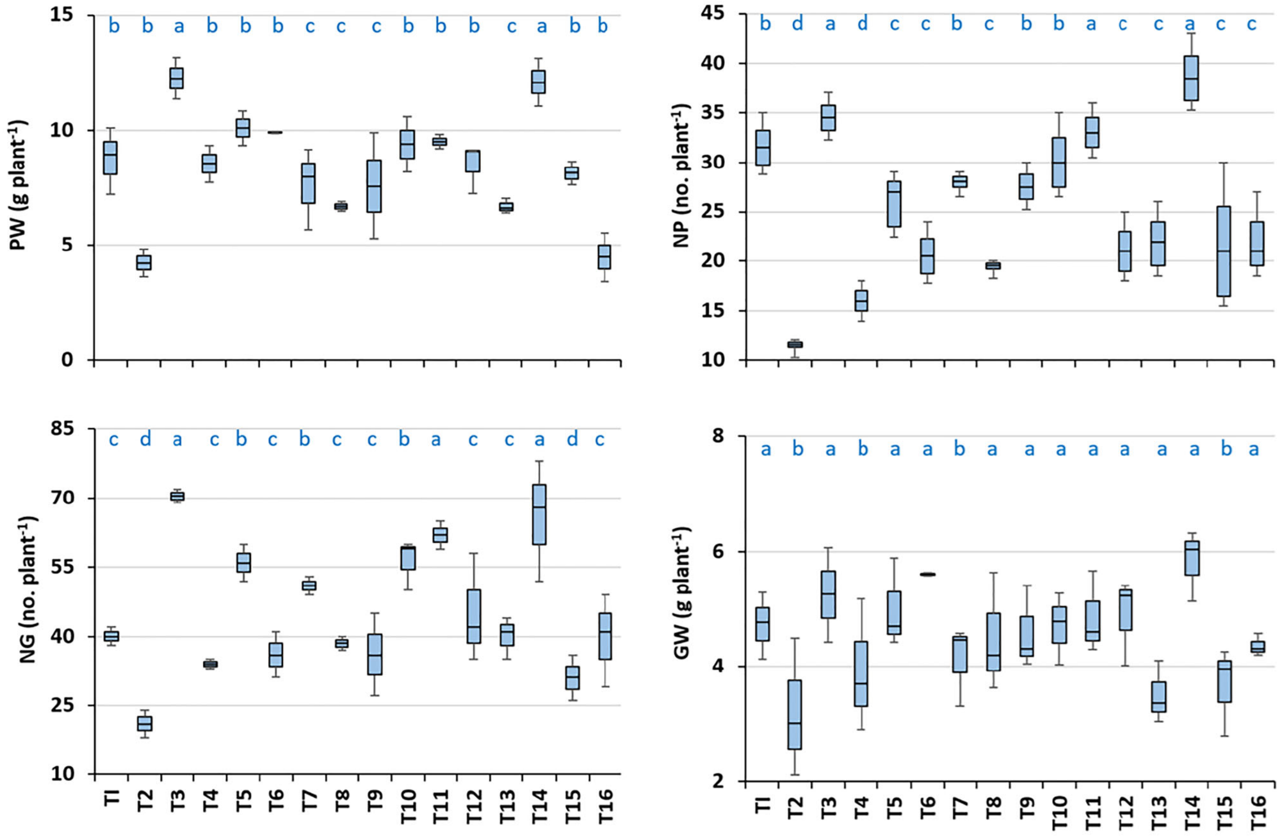

FIGURE 2 Boxplot and Scott–Knott test for pod weight (PW – g plant^{-1}), number of pods (NP – no. plant^{-1}), number of grains (NG – no. plant^{-1}), and grain weight (GW – g plant^{-1}) of soybean co-inoculated with plant growth-promoting rhizobacteria (PGPRs) under water-deficit stress. Treatments: T1 = BRM 063574+BRM 67205+ESA 441; T2 = BRM 063574+BRM 67205+BRM 034008; T3 = BRM 063574+BRM 063573+ESA 441; T4 = BRM 063574+BRM 063573+BRM 034008; T5 = Ab-V5+BRM 67205+ESA 441; T6 = Ab-V5+BRM 67205+BRM 034008; T7 = Ab-V5+BRM 063573+ESA 441; T8 = Ab-V5+BRM 063573+BRM 034008; T9 = BRM 063574; T10 = Ab-V5; T11 = BRM 67205; T12 = BRM 063573; T13 = ESA 441; T14 = BRM 034008; T15 = Absolute Control and T16 = Fertilized Control. Groups labeled by the same letter did not differ significantly by the Scott–Knott test ($p < 0.05$).

TABLE 5 Variance analysis for pod weight (PW = g plant^{-1}), number of pods (NP = unit plant^{-1}), number of grains (NG = unit plant^{-1}), and grain weight (GW = g plant^{-1}) of soybean co-inoculated with plant growth-promoting rhizobacteria (PGPRs) under water-deficit stress.

Source of variation	*df*	PW	NP	NG	GW
Treatment	15	2.08*	1.35*	0.74*	1.62*
CV (%)		13.97	18.93	15.17	10.21

Abbreviations: CV, coefficient of variation; DF, degrees of freedom.
* denote significant by the *F* test at 5% of significance.

co-inoculation showed better responses in gas exchange parameters. PCA results indicate that the interaction of PGPR in soybean under water-deficit stress conditions is positive, allowing for the resilience of soybean plants.

4 | DISCUSSION

With the exception of treatments T2 and T4, mean gs values were lower after 14 days than after 7 days of water limitation. This indicates stomatal closure resulting from reduced leaf tissue turgor and decreased leaf growth (Tombesi et al., 2015). However, this effect was notably less pronounced in treatments inoculated with PGPRs. This is probably due to rhizobacteria producing plant hormones such as auxins, gibberellins, and cytokinins. These plant hormones help to maintain plant growth by counteracting the negative effects of abscisic acid and ethylene, which induce leaf abscission (Bulegon, Almeida, et al., 2019; Bulegon, Guimarães, et al., 2019).

There was also a reduction in the E due to the induced water-deficit stress; however, in treatments inoculated with PGPRs, this reduction in E was smaller. Similar E responses to water-deficit stress in inoculated plants have been reported by Silva, Nascente, et al. (2020). Stomatal closure was slower than transpiration. This was evidenced by significant reductions in gs and E, respectively, after 7 and 14 days of stress. Stomatal closure limits water loss, allowing the plant to make better use of the available water in the soil, resulting in a greater fixation of CO_2 per unit of water transpired (Bulegon,

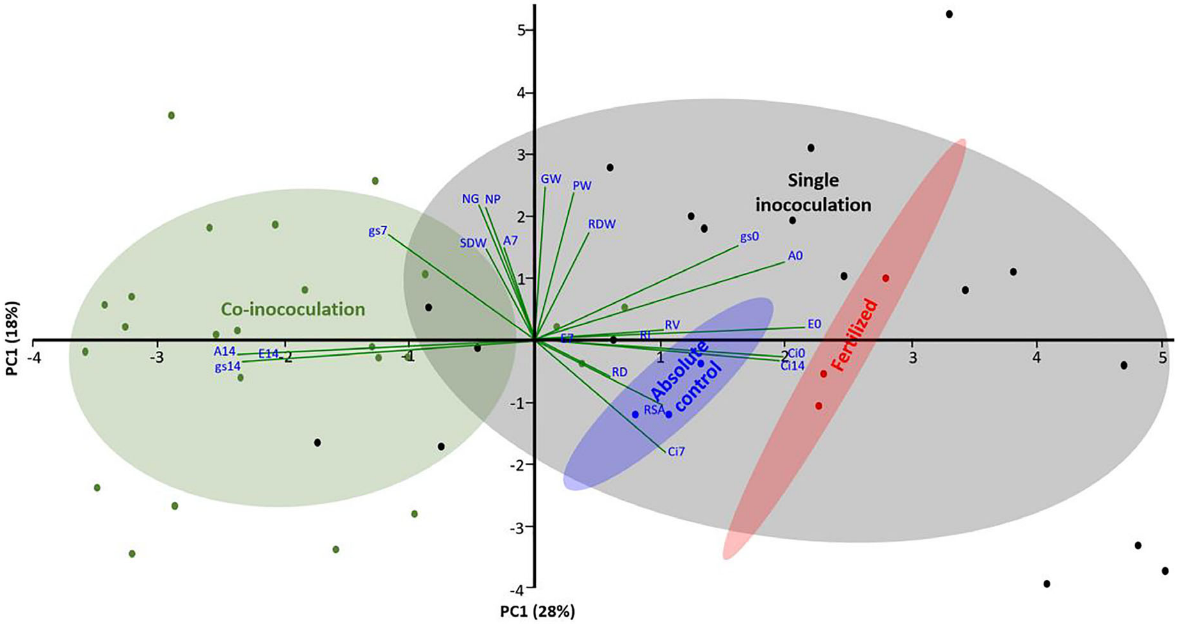

FIGURE 3 Principal component analysis among the parameters stomatal conductance (gs), transpiration rate (E), internal CO_2 concentration (Ci), net photosynthesis rate (A), shoot dry weight (SDW), root dry weight (RDW), root length (RL), root surface area (RSA), root diameter (RD), root volume (RV), pod weight (PW), number of pods (NP), number of grains (NG), and grain weight (GW) in inoculated soybean subjected to water-deficit stress.

Guimarães, et al., 2019), thus increasing the plant's water use efficiency.

The degree of stomatal closure also influences Ci, which remained stable or increased after 14 days of stress in all treatments (Table 3). This is due to low CO_2 consumption in photosynthesis and greater release in respiration and photorespiration, conditions that hinder the plant's ability to gain C. However, treatments with lower Ci (T1, T2, T3, T4, T5, T7, and T11) had a less negative impact and also exhibited higher A. Therefore, they accumulated more C under moderate water limitation due to the benefits of inoculations, except in T9. Most treatments with better metabolic performance in this evaluation had the benefit of co-inoculations, except T11.

The inoculation of *A. brasilense* promotes better root growth, allowing for improved exploration of soil nutrients and water, keeping cells more turgid, and indirectly influencing stomatal opening, thus enhancing light absorption, CO_2 assimilation, and transpiration (E) (Bulegon, Almeida, et al., 2019; Buso et al., 2021). However, parameters related to photosynthesis, such as A, E, and gs, decrease with the increase of the water tension in the soil (Silva, Santos, et al., 2020), corroborating the results found in the present study.

SDW was clearly higher at T1, T3, and T14 compared to the other treatments, while RDW was higher at T1, T3, T4, T6, T7, T10, T12, and T14. Only T1, T3, T4, T6, and T7 can be associated with higher photosynthetic taxa (A); therefore, higher gain of C at the end of stress (Figure 1; Table 3). Lower SDW can be determined by leaf abscission to reduce water loss during stress or lower leaf emission in the recovery period. RDW varied less, mainly for the plant's need to maintain root growth to increase water absorption alone. There are variations in the root architecture (RL, RSA, RD, and RV) between treatments that reflect the effect of the inoculations (Figure 1).

Water-deficit stress negatively affects the growth of soybean plants, compromising their productivity. In response to environmental conditions, plants react through morphological, physiological, biochemical, and molecular changes (Natali et al., 2018; Qaseem et al., 2018). Reductions in both length and dry mass of shoot and root, as well as in photosynthetic pigments and relative water content in soybean genotypes exposed to water-deficit stress, are reported (Mohamed & Latif, 2017).

The inoculation of PGPR has become one of the strategies for promoting plant growth under water-deficit stress conditions (Ilyas et al., 2020) through improved water and nutrient absorption. Better water and nutrient absorption promotes changes in root system architecture, such as increased root growth and/or the formation of lateral roots and root hairs (Grover et al., 2021). A wide variety of PGPR produce exopolysaccharides (EPS), assisting plants in better colonization of their root systems (Morcillo & Manzanera, 2021), as well as improving seed germination and tolerance to stress (Astorga-Eló et al., 2021; Kumar & Verma, 2018).

Changes in root system architecture are driven by the synthesis of phytohormones in plants, such as auxins, gibberellins, cytokinins, ethylene, and abscisic acid (Jan et al., 2024), resulting in significant differences in growth parameters, such as root and SDW, as observed by co-inoculating soybean with *B. japonicum* and *A. brasilense* (Puente et al., 2018).

The decline in A at the end of the stress period, which occurred at the reproductive structure formation—R3 stage (Table 3), led to lower availability of photoassimilates. This justifies the significant reduction in the NP and seeds in some treatments (Figure 2), as these are the two most important components of soybean grain yield. There was also a reduction in pod mass, which is directly related to seed number and mass. However, no perfect correlation was found between maintaining high A levels during stress and productive performance. This only occurred in T3, one of the treatments with the highest grain mass, but not in T4 or T7. Conversely, treatments with reduced A, such as T14, exhibited high seed mass. This response may be due to gas exchange being measured at specific times or to the plant's ability to recover concerning gas exchange and the emission of new leaves or reproductive structures, which are influenced by co-inoculations.

Upon analyzing soybean cultures inoculated with *A. brasilense* under drought stress, Bulegon, Almeida, et al. (2019) found that foliar application mitigated production losses. Comparing the drought-stressed treatments with the irrigated control revealed a 28.36% reduction in production per plant for the dry control and a 13.77% reduction with the foliar application of *A. brasilense*. The same study also found that water deficit significantly affected the number of seeds per pod but had no impact on the NP per plant or the mass of 100 seeds.

This study highlights the significant impact of multifunctional microorganism inoculation on soybean gas exchange, growth, and productivity under water-deficit stress. In particular, co-inoculated treatments BRM 063574+BRM 67205+BRM 034008, BRM 063574+BRM 063573+ESA 441, BRM 063574+BRM 063573+BRM 034008, and Ab-V5+BRM 063573+ESA 441 showed improved gas exchange rates compared to single inoculation, especially at the last day of the induced stress. Treatments BRM 063574+BRM 063573+ESA 441 and BRM 034008 emerged as standout performers in several growth and productivity metrics, including shoot and RDWs, NP, and GW. Principal component analysis highlighted the clear benefits of co-inoculation, particularly in gas exchange efficiency, while single inoculation showed notable benefits in yield components.

Overall, these results suggest that the consortium of different PGPR can enhance soybean productivity and adaptability under challenging environmental conditions. Moreover, the results support further exploration of these inoculation strategies to optimize agricultural practices and enhance crop resilience to abiotic stresses and highlight the potential of PGPR management as a tool for sustainable agriculture.

5 | CONCLUSIONS

Co-inoculation alleviates the effects of water stress on gas exchange, plant growth, and productivity in soybeans more than single inoculation.

Parameters related to gas exchange are less impacted by water-deficit stress in the co-inoculations. BRM 063574+BRM 67205+BRM 034008, BRM 063574+BRM 063573+ESA 441, BRM 063574+BRM 063573+BRM 034008, and Ab-V5+BRM 67205+BRM 034008;

The co-inoculations BRM 063574+BRM 67205+ESA 441, and BRM 063574+BRM 063573+ESA 441 provide the best results for root and shoot growth of soybean subjected to water-deficit stress.

The yield components of soybean under water-deficit stress show good results with the co-inoculations BRM 063574+BRM 67205+ESA 441, BRM 063574+BRM 063573+ESA 441, and Ab-V5+BRM 67205+BRM 034008;

Considering the effects of inoculations on gas exchange, plant growth, and grain production, co-inoculations BRM 063574+BRM 063573+ESA 441 and BRM 063574+BRM 67205+ESA 441 provide soybean with greater tolerance to water-deficit stress;

From a farmer's perspective, co-inoculation with specific microbial strains offers a low-cost, scalable, and easy-to-adopt solution that enhances the drought resilience of soybeans, improving growth, yield, and sustainability in water-limited regions.

AUTHOR CONTRIBUTIONS

Michel Aldrighi: Data curation; formal analysis; investigation; validation; visualization; writing—original draft; writing—review and editing. **Juliana Domingues Lima**: Formal analysis; methodology; validation; visualization; writing—review and editing. **Paulo Ivan Fernandes-Júnior**: Formal analysis; methodology; validation; visualization; writing—review and editing. **Enderson Petrônio de Brito Ferreira**: Conceptualization; data curation; formal analysis; funding acquisition; methodology; project administration; resources; supervision; validation; visualization; writing—review and editing.

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CONFLICT OF INTEREST STATEMENT

The authors declare no conflicts of interest.

DATA AVAILABILITY STATEMENT

The data supporting this study's findings are available from the corresponding author upon reasonable request.

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REFERENCES

Ahluwalia, O., Singh, P. C., & Bhatia, R. (2021). A review on drought stress in plants: Implications, mitigation and the role of plant growth promoting rhizobacteria. *Resources, Environment and Sustainability*, *5*, 100032. https://doi.org/10.1016/j.resenv.2021.100032

Ahmad, H. M., Fiaz, S., Hafeez, S., Zahra, S., Shah, A. N., Gul, B., Aziz, O., Mahmood-Ur-Rahman, Fakhar, A., Rafique, M., Chen, Y., Yang, S. H., & Wang, X. (2022). Plant growth-promoting rhizobacteria eliminate the effect of drought stress in plants: A review. *Frontiers in Plant Science*, *13*, Article 875774. https://doi.org/10.3389/fpls.2022.875774

Almeida, D. S., Menezes-Blackburn, D., Zhang, H., Haygarth, P. M., & Rosolem, C. A. (2019). Phosphorus availability and dynamics in soil affected by long-term ruzigrass cover crop. *Geoderma*, *337*, 434–443. https://doi.org/10.1016/j.geoderma.2018.09.056

Asobia, P. C., Paula, K. L. M., Oliveira, M. I., Bittencourt, C. D., Wendland, A., & Ferreira, E. P. B. (2025). Co-inoculation of beneficial microorganisms in upland rice cultivated at different phosphorus levels. *Canadian Journal of Soil Science*, *105*, 1–13. https://doi.org/10.1139/cjss-2024-0054

Astorga-Eló, M., Gonzalez, S., Acuña, J. J., Sadowsky, M. J., & Jorquera, M. A. (2021). Rhizobacteria from 'flowering desert' events contribute to the mitigation of water scarcity stress during tomato seedling germination and growth. *Scientific Reports*, *11*(1), 13745. https://doi.org/10.1038/s41598-021-93303-8

Barbosa, M. R., Silva, M. M. A., Willadino, L., Ulisses, C., & Camara, T. R. (2014). Geração e desintoxicação enzimática de espécies reativas de oxigênio em plantas. *Ciência Rural*, *44*(3), 453–460. https://doi.org/10.1590/S0103-84782014000300011

Battisti, R., & Sentelhas, P. C. (2014). New agroclimatic approach for soybean sowing dates recommendation: A case study. *Revista Brasileira de Engenharia Agrícola e Ambiental*, *18*(11), 1149–1156. https://doi.org/10.1590/1807-1929/agriambi.v18n11p1149-1156

Bittencourt, C. D., Messias, M., Wendland, A., & de Brito Ferreira, E. P. (2024). Phosphate-solubilizing inoculant improves agronomic performance of common bean with reduced phosphate fertilizer dose. *Journal of Soil Science and Plant Nutrition*, *24*(3), 5815–5828. https://doi.org/10.1007/s42729-024-01943-2

Bulegon, L. G., Almeida, V. F., Inagaki, A. M., Battistus, A. G., Offemann, L. C., & Souza, A. K. P. (2019). Respostas da soja ao *Azospirillum brasilense* e reguladores vegetais em condições de déficit hídrico. *Revista Brasileira de Ciências Agrárias—Brazilian Journal of Agricultural Sciences*, *14*(4), 1–10. https://doi.org/10.5039/agraria.v14i4a5678

Bulegon, L. G., Guimarães, V. F., Battistus, A. G., Inagaki, A. M., & Costa, N. V. (2019). Mitigation of drought stress effects on soybean gas exchanges induced by *Azospirillum brasilense* and plant regulators. *Pesquisa Agropecuária Tropical*, *49*(e52807), 1–9.

Bulegon, L. G., Rampim, L., Klein, J., Kestring, D., Guimarães, V. F., Battistus, A. G., & Inagaki, A. M. (2016). Componentes de produção e produtividade da cultura da soja submetida à inoculação de Bradyrhizobium E *Azospirillum*. *Terra Latinoamericana*, *34*(2), 169–176.

Buso, P. H. M., de Oliveira, R. A., Daros, E., Zambon, J. L. C., Venancio, W. S., Souchie, E. L., Buso, E. K. R. P. D. M., & Díaz-Zorita, M. (2021). Plant growth analysis describing the soybean plants response on dryland field to seed co-inoculation. *Ciência Rural*, *51*, 1–9. https://doi.org/10.1590/0103-8478cr20190642

Cassán, F., Vanderleyden, J., & Spaepen, S. (2014). Physiological and agronomical aspects of phytohormone production by model plant-growth-promoting rhizobacteria (PGPR) belonging to the genus *Azospirillum*. *Journal of Plant Growth Regulation*, *33*(2), 440–459. https://doi.org/10.1007/s00344-013-9362-4

Ciampitti, I. A., & Salvagiotti, F. (2018). New insights into soybean biological nitrogen fixation. *Agronomy Journal*, *110*(4), 1185–1196. https://doi.org/10.2134/agronj2017.06.0348

CONAB. (2023). *Acompanhamento da safra brasileira – grãos: Monitoramento agrícola, safra 2022/2023*. Companhia Nacional de Abastecimento.

de Oliveira-Paiva, C. A., Bini, D., de Sousa, S. M., Ribeiro, V. P., dos Santos, F. C., de Paula Lana, U. G., de Souza, F. F., Gomes, E. A., & Marriel, I. E. (2024). Inoculation with *Bacillus megaterium* CNPMS B119 and *Bacillus subtilis* CNPMS B2084 improve P-acquisition and maize yield in Brazil. *Frontiers in Microbiology*, *15*, 1426166. https://doi.org/10.3389/fmicb.2024.1426166

Fernandes-Júnior, P. I., Aidar, S. D. T., Morgante, C. V., Gava, C. A. T., Zilli, J. É., Souza, L. S. B. D., Marinho, R. D. C. N., Nóbrega, R. S. A., Brasil, M. D. S., Seido, S. L., & Martins, L. M. V. (2015). The resurrection plant *Tripogon spicatus* (Poaceae) harbors a diversity of plant growth promoting bacteria in northeastern Brazilian Caatinga. *Revista Brasileira de Ciência do Solo*, *39*(4), 993–1002. https://doi.org/10.1590/01000683rbcs20140646

Ferreira, D. F. (2019). SISVAR: A computer analysis system to fixed effects split plot type designs. *Revista Brasileira de Biometria*, *37*(4), 529–535. https://doi.org/10.28951/rbb.v37i4.450

Grover, M., Bodhankar, S., Sharma, A., Sharma, P., Singh, J., & Nain, L. (2021). PGPR mediated alterations in root traits: Way toward sustainable crop production. *Frontiers in Sustainable Food Systems*, *4*, 618230. https://doi.org/10.3389/fsufs.2020.618230

Hong, Z., Yan, J., Jiang, J., Li, J., & Xu, R. (2021). Direct quantification of sorption thermodynamics of phosphate on four soil colloids through isothermal titration calorimetry. *ACS Earth and Space Chemistry*, *5*(2), 295–304. https://doi.org/10.1021/acsearthspacechem.0c00281

Hungria, M., Ribeiro, R. A., & Nogueira, M. A. (2018). Draft genome sequences of *Azospirillum brasilense* strains Ab-V5 and Ab-V6, Commercially Used in Inoculants for Grasses and Legumes in Brazil. *Genome Announcements*, *6*(20), 1. https://doi.org/10.1128/genomeA.00393-18

Ilyas, N., Mumtaz, K., Akhtar, N., Yasmin, H., Sayyed, R. Z., Khan, W., Enshasy, H. A. E., Dailin, D. J., Elsayed, E. A., & Ali, Z. (2020). Exopolysaccharides producing bacteria for the amelioration of drought stress in wheat. *Sustainability*, *12*(21), 8876. https://doi.org/10.3390/su12218876

Jabborova, D., Kannepalli, A., Davranov, K., Narimanov, A., Enakiev, Y., Syed, A., Elgorban, A. M., Bahkali, A. H., Wirth, S., Sayyed, R. Z., & Gafur, A. (2021). Co-inoculation of rhizobacteria promotes growth, yield, and nutrient contents in soybean and improves soil enzymes and nutrients under drought conditions. *Scientific Reports*, *11*(1), 22081. https://doi.org/10.1038/s41598-021-01337-9

Jan, M., Muhammad, S., Jin, W., Zhong, W., Zhang, S., Lin, Y., Zhou, Y., Liu, J., Liu, H., Munir, R., Yue, Q., Afzal, M., & Wang, G. (2024). Modulating root system architecture: Cross-talk between auxin and phytohormones. *Frontiers in Plant Science*, *15*, 1343928. https://doi.org/10.3389/fpls.2024.1343928

Kamran, M. A., Bibi, S., Chen, B., Jiang, J., & Xu, R.-K. (2022). Elucidating the mechanisms determining the availability of phosphate by application of biochars from different parent materials. *Environmental Geochemistry and Health*, *44*(11), 4191–4200. https://doi.org/10.1007/s10653-021-01184-7

Kumar, A., & Verma, J. P. (2018). Does plant—Microbe interaction confer stress tolerance in plants: A review? *Microbiological Research*, *207*, 41–52. https://doi.org/10.1016/j.micres.2017.11.004

Lenin, G., & Jayanthi, M. (2012). Indole acetic acid, gibberellic acid and siderophore production by PGPR isolates from rhizospheric soils of Catharanthus roseus. *International Journal of Pharmaceutical and Biological Archives*, *3*(4), 933–938.

Masciarelli, O., Urbani, L., Reinoso, H., & Luna, V. (2013). Alternative mechanism for the evaluation of indole-3-acetic acid (IAA) production by *Azospirillum brasilense* strains and its effects on the germination and growth of maize seedlings. *Journal of Microbiology*, *51*(5), 590–597. https://doi.org/10.1007/s12275-013-3136-3

Mohamed, H. I., & Latif, H. H. (2017). Improvement of drought tolerance of soybean plants by using methyl jasmonate. *Physiology and Molecular Biology of Plants*, *23*(3), 545–556. https://doi.org/10.1007/s12298-017-0451-x

Morcillo, R., & Manzanera, M. (2021). The effects of plant-associated bacterial exopolysaccharides on plant abiotic stress tolerance. *Metabolites*, *11*(6), 337. https://doi.org/10.3390/metabo11060337

Natali, L., Vangelisti, A., Guidi, L., Remorini, D., Cotrozzi, L., Lorenzini, G., Nali, C., Pellegrini, E., Trivellini, A., Vernieri, P., Landi, M., Cavallini, A., & Giordani, T. (2018). How *Quercus ilex* L. saplings face combined salt and ozone stress: A transcriptome analysis. *BMC Genomics*, *19*(1), 872. https://doi.org/10.1186/s12864-018-5260-2

Pereira, R. C., Guimarães, C. M., Heinemann, A. B., Lanna, A. C., & Lopes Junior, S., Narciso, M. G., Stone, L. F., Vianello, R. P., & Castro, A. P. (2017). *SITIS—Plant phenotyping platform*. https://ainfo.cnptia.embrapa.br/digital/bitstream/item/165780/1/CNPAF-2017-p47.pdf

Poudel, S., Vennam, R. R., Shrestha, A., Reddy, K. R., Wijewardane, N. K., & Reddy, K. N. (2023). Resilience of soybean cultivars to drought stress during flowering and early-seed setting stages. *Scientific Reports*, *13*, 1277. https://doi.org/10.1038/s41598-023-28354-0

Prando, A. M., Barbosa, J. Z., Oliveira, A. B. D., Nogueira, M. A., Possamai, E. J., & Hungria, M. (2024). Benefits of soybean co-inoculation with *Bradyrhizobium* spp. and *Azospirillum brasilense*: Large-scale validation with farmers in Brazil. *European Journal of Agronomy*, *155*, 127112. https://doi.org/10.1016/j.eja.2024.127112

Proque, A. L. (2019). Interdependência do Brasil e dos Estados Unidos no mercado da soja em grão: Uma análise da situação e das perspectivas. *Revista de Desenvolvimento e Políticas Públicas*, *3*(1), 41–60. https://doi.org/10.31061/redepp.v3n1.41-60

Puente, M. L., Gualpa, J. L., Lopez, G. A., Molina, R. M., Carletti, S. M., & Cassán, F. D. (2018). The benefits of foliar inoculation with *Azospirillum brasilense* in soybean are explained by an auxin signaling model. *Symbiosis*, *76*(1), 41–49. https://doi.org/10.1007/s13199-017-0536-x

Qaseem, M. F., Qureshi, R., Muqaddasi, Q. H., Shaheen, H., Kousar, R., & Röder, M. S. (2018). Genome-wide association mapping in bread wheat subjected to independent and combined high temperature and drought stress. *PLoS One*, *13*(6), e0199121. https://doi.org/10.1371/journal.pone.0199121

Silva, J. A. D., Santos, P. A. B. D., Carvalho, L. G. D., Moura, E. G., & Andrade, F. R. (2020). Gas exchanges and growth of soybean cultivars submitted to water deficiency. *Pesquisa Agropecuária Tropical*, *50*, 1. https://doi.org/10.1590/1983-40632020v5058854

Silva, J. F. (2017). *Caracterização polifásica de bactérias promotoras de crescimento vegetal associados ao sorgo (Sorghum bicolor (L.) Moench) e ao milheto (Penissetum glaucum (L.) R. Brown) cultivados no Sertão Pernambuco*. http://www.cpgrnsa.univasf.edu.br/uploads/7/8/9/0/7890742/disserta%C3%A7%C3%A3o__jessica_fernanda_da_silva.pdf

Silva, M. A., Nascente, A. S., Filippi, M. C. C. D., Lanna, A. C., Silva, G. B. D., & Silva, J. F. A. E. (2020). Individual and combined growth-promoting microorganisms affect biomass production, gas exchange and nutrient content in soybean plants. *Revista Caatinga*, *33*(3), 619–632. https://doi.org/10.1590/1983-21252020v33n305rc

Siqueira, A. F., Ormeño-Orrillo, E., Souza, R. C., Rodrigues, E. P., Almeida, L. G. P., Barcellos, F. G., Batista, J. S. S., Nakatani, A. S., Martínez-Romero, E., Vasconcelos, A. T. R., & Hungria, M. (2014). Comparative genomics of *Bradyrhizobium japonicum* CPAC 15 and *Bradyrhizobium diazoefficiens* CPAC 7: Elite model strains for understanding symbiotic performance with soybean. *BMC Genomics [Electronic Resource]*, *15*(1), 420. https://doi.org/10.1186/1471-2164-15-420

Teixeira, P. C., Donagemma, G. K., Fontana, A., & Teixeira, W. G. (Eds.). (2017). *Manual de Métodos de Análise de Solo*. Embrapa.

Tian, J., Lu, X., Chen, Q., Kuang, X., Liang, C., Deng, L., Lin, D., Cai, K., & Tian, J. (2022). Phosphorus fertilization affects soybean rhizosphere phosphorus dynamics and the bacterial community in karst soils. *Plant and Soil*, *475*(1–2), 137–152. https://doi.org/10.1007/s11104-020-04662-6

Tombesi, S., Nardini, A., Frioni, T., Soccolini, M., Zadra, C., Farinelli, D., Poni, S., & Palliotti, A. (2015). Stomatal closure is induced by hydraulic signals and maintained by ABA in drought-stressed grapevine. *Scientific Reports*, *5*, 12449. https://doi.org/10.1038/srep12449

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