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Bradyrhizobium brasilense as an efficient soybean microsymbiont in two contrasting soils of the southwestern region of Piauí (Cerrado biome)

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ABSTRACT: The goal of this study was to evaluate the efficiency of *Bradyrhizobium brasilense* strains native to the soil of the semiarid region in northeastern Brazil in symbiosis with soybeans in two contrasting soils in southwestern Piauí. A pot experiment was conducted using a randomized block design in a 10 × 2 factorial scheme. There were 10 nitrogen (N) sources: six native *Bradyrhizobium* strains [UFLA06-13, UFLA06-15, UFLA06-19, UFLA06-21, UFLA06-22 (*B. brasilense*), and UFLA06-24 (*Bradyrhizobium* sp.)], two controls with strain SEMIA 5019 (*B. elkanii*) and a commercial inoculant [SEMIA 5079 (*B. japonicum*) + SEMIA 5080 (*B. diazoefficiens*)] recommended for soybeans, and two controls without the application of an inoculant (one with and the other without the application of mineral N). The second experimental factor corresponded to the use of two soils (Oxisol and Quartzarenic Neosol). All strains showed increased nodulation and shoot nitrogen content in soybean plants in both soils. Most strains promoted higher nitrogen fixation when inoculated in the Oxisol. UFLA06-19, UFLA06-22, and UFLA06-24 were efficient in nitrogen accumulation in the shoots of soybeans in the Oxisol. This is the first report regarding the efficiency of *B. brasilense* strains in symbiosis with soybeans under different soil conditions.

Key words: biological nitrogen fixation; *Glycine max*; inoculation; symbiosis

Bradyrhizobium brasilense como um eficiente microssimbionte de soja em dois solos contrastantes da região Sudoeste do Piauí (bioma Cerrado)

RESUMO: Este trabalho teve como objetivo avaliar a eficiência de estirpes de *Bradyrhizobium brasilense* nativas de solo da região semiárida do Nordeste brasileiro em simbiose com soja em dois solos contrastantes do Sudoeste do Piauí. Foi conduzido um experimento de vasos, usando um delineamento em blocos casualizados, em esquema fatorial 10 x 2. Foram 10 fontes de nitrogênio (N): seis estirpes de *Bradyrhizobium* nativas [UFLA06-13, UFLA06-15, UFLA06-19, UFLA06-21, UFLA06-22 (*B. brasilense*) e UFLA06-24 (*Bradyrhizobium* sp.)]; dois controles compostos pela estirpe SEMIA 5019 (*B. elkanii*) e um inoculante comercial [SEMIA 5079 (*B. japonicum*) + SEMIA 5080 (*B. diazoefficiens*)] recomendados para a soja, e dois controles sem a aplicação do inoculante (um com e o outro sem a aplicação de N mineral). O segundo fator experimental correspondeu ao uso de dois solos (Latossolo Amarelo e Neossolo Quartzarênico). Todas as estirpes aumentaram a nodulação e o teor de nitrogênio na parte aérea de plantas de soja, em ambos os solos. A maioria das estirpes promoveu maior fixação de nitrogênio quando inoculadas na soja no Latossolo Amarelo. As estirpes UFLA06-19, UFLA06-22 e UFLA06-24 foram eficientes no acúmulo de nitrogênio da parte aérea de soja no Latossolo Amarelo. Este é o primeiro relato de eficiência de estirpes da espécie *B. brasilense* em simbiose com soja em diferentes condições de solo.

Palavras-chave: fixação biológica de nitrogênio; *Glycine max*; inoculação; simbiose

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Introduction

Soybean (*Glycine max* L.) crops are widespread in Brazil and worldwide and are considered one of the main commodities in the agricultural sector. Soybeans encompass approximately 45% of the cropped area in Brazil, with a production output of 121,522.363 tons in the 2019/2020 crop season (IBGE, 2020). Soybean production in Piauí State (2,447.620 tons) represents only 2% of the Brazilian crop production (IBGE, 2020). However, in this state, especially in the southwestern mesoregion, which is considered one of the last agricultural frontiers in the country, there has been an extensive increase in soybean production and yield over the last few years (Alcântara Neto et al., 2018).

Large amounts of nutrients are required to cultivate soybeans, such as nitrogen (N), which is considered the most important nutrient because it participates in several metabolic processes in plants. Generally, 80 kg of N is required to produce 1,000 kg of soybean grains (Hungria & Mendes, 2015), provided by mineral fertilizers obtained by industrial fixation (Haber-Bosch process). However, in addition to high costs, these fertilizers may result in a high degree of environmental pollution when not managed properly. Alternatively, the process of biological nitrogen fixation (BNF) performed by *Bradyrhizobium* strains in symbiosis with soybeans is an economical and sustainable alternative for the supply of N to the crop, providing average annual savings of nearly US$ 15 billion in Brazil (Hungria & Mendes, 2015).

Soybeans can establish symbiosis with diverse *Bradyrhizobium* species, including *B. japonicum*, *B. elkanii*, *B. diazoefficiens*, *B. liaoningense*, *B. yuanmingense*, *B. huanghuaihaiense*. *B. daqingense*, *B. ottawaense*, and *B. amphicarpaeae* (Delamuta et al., 2013; Wang et al., 2013; Yu et al., 2014; Ribeiro et al., 2015; Bromfield et al., 2019). These nine species have been described based on the strains from temperate regions. However, recently, strains isolated from soybean nodules in the state of Piauí (Cerrado biome) were found to be efficient for fixing nitrogen in symbiosis with soybeans during experiments conducted under axenic conditions (Ribeiro et al., 2015). Some of these strains were classified as *Bradyrhizobium brasilense* (symbiovar sojae) (Costa et al., 2020).

Studies on the BNF for soybeans in Brazil have increased its cultivation in the country, ensuring its competitiveness in the external market. *Bradyrhizobium* strains delivered in commercial inoculants, when in symbiosis with soybeans, are able to partially or completely cater to the N demand of the crop (Hungria & Mendes, 2015; Kaschuk et al., 2016).

Although the use of inoculant strains has been consolidated in soybeans, it is important to consider that these microorganisms can be significantly influenced by several factors, such as climatic conditions, pH, nutrient availability, humidity, soil texture, and competition with the native communities of N_2-fixing bacteria (rhizobia) for the occupation of infection sites in the roots of the host plants (Kaschuk et al., 2016; Castro et al., 2017; Thilakarathna & Raizada, 2017). However, there may be strains with desirable symbiotic characteristics for selection in these autochthonous communities, which can be used as inoculants of soybeans (Rufini et al., 2014; Ribeiro et al., 2015; Bromfield et al., 2017; Chibeba et al., 2017). These facts reinforce the need for recurrent selection of new efficient *Bradyrhizobium* strains.

In Brazil, four *Bradyrhizobium* strains have been selected and authorized by the Ministry of Agriculture, Livestock, and Supply (MAPA) to produce soybean inoculants (Brasil, 2011). They have been isolated from soils of the south (*B. elkanii* SEMIA 587), southeastern (*B. elkanii* SEMIA 5019), and midwestern (*B. diazoefficiens* SEMIA 5080 and *B. japonicum* SEMIA 5079) regions (Boddey & Hungria, 1997). During the selection process, assessing the agronomic efficiency of each strain under different edaphoclimatic conditions is very important because inherent factors affect their adaptability that could increase cell mortality when introduced at specific sites.

Based on the importance of the BNF process for soybean crops, and considering the edaphoclimatic diversity of the agricultural systems, it is necessary to perform studies for the selection of new strains for soybean inoculation, especially in the northeastern region of Brazil, because the currently recommended strains are from other regions and may not fully express a high symbiotic efficiency in this environment. In the present study, five *Bradyrhizobium brasilense* strains (UFLA06-13, UFLA06-15, UFLA06-19, UFLA06-21, and UFLA06-22) and one *Bradyrhizobium* sp. strain (UFLA06-24) from the semiarid region in northeastern Brazil, which are efficient soybean microsymbionts under axenic conditions, were used to evaluate their N_2 fixation efficiency in symbiosis with soybeans in two soils in southwestern Piauí.

Materials and Methods

This experiment was conducted for 40 days, between November and December 2018, in a greenhouse at the Federal University of Piauí (UFPI), Campus Professora Cinobelina Elvas, in pots filled with non-sterile soil samples as the substrate. The mean internal temperatures registered in the greenhouse varied from 26 to 34 °C during the experimental period.

The experimental design was randomized blocks, in a 10 × 2 factorial scheme, totaling 20 treatments, with four replications. The first tested factor corresponded to 10 nitrogen (N) sources: six native *Bradyrhizobium* strains from the semiarid region of northeastern Brazil (UFLA06-24, UFLA06-13, UFLA06-15, UFLA06-19, UFLA06-21, and UFLA06-22) isolated by Ribeiro et al. (2015) and classified as *B. brasilense* (Costa et al., 2020), except for strain UFLA06-24 (*Bradyrhizobium* sp.); two controls consisting of the SEMIA 5019 strain (*B. elkanii*) and a commercial inoculant [*B. japonicum* (SEMIA 5079) + *B. diazoefficiens* (SEMIA 5080)] authorized by MAPA for the inoculation of the soybean crop (Brasil, 2011); and two controls without applying an inoculant, one without and one with the application of mineral N. The control with mineral N received a total dose of 300 mg dm^{-3}, which was provided over three applications, using NH_4NO_3 as the source. The second experimental factor corresponded to the use of two soils: an Oxisol (Yellow Latosol) and a Quartzarenic Neosol .

Samples of each soil were collected from the 0–20 cm layer in areas without previous utilization of microbial inoculants in southwestern Piauí. The Quartzarenic Neosol was collected in an area cultivated with pasture for more than 20 years in the municipality of Santa Luz, Piauí (8°55′1.4124″W, 44°9′41.778″S). The Oxisol was collected in an area of native forest, at the "Vô Desidério" Farm, municipality of Bom Jesus, Piauí (9°13′28.9344″ W, 44°44′44.4192″ S). After collection, each soil sample was air-dried, ground, sieved (4 mm mesh sieve), and aliquots of 5 kg of each soil were added to plastic pots. Some of the samples were subjected to chemical and granulometric analyses (Table 1).

Liming was performed in all pots to increase the base saturation to 60% using dolomitic limestone with a relative total neutralization power of 91%. Thirty days after limestone application, fertilization with phosphorus (P) and potassium (K) was performed according to the availability of the elements and the recommendation for crops in Cerrado soils (Sousa & Lobato, 2004). We used 75 mg pot^{-1} of P_2O_5 and 100 mg pot^{-1} of K_2O in the Quartzarenic Neosol, and 400 mg pot^{-1} of P_2O_5 and 400 mg pot^{-1} of K_2O in Oxisol using monoammonium phosphate and potassium chloride as sources. In all plots, fertilization with micronutrients was performed by applying 0.5, 1.5, 3.6, 5, and 0.1 mg dm^{-3} of B, Cu, Mn, Zn, and Mo, respectively, according to the recommendations of Malavolta et al. (1989).

The soybean genotype used was FT 4280 IPRO, cycle 8.0, widely employed in the region. The seeds were superficially disinfected with 70% alcohol for 30 s and 2% sodium hypochlorite for 2 min, with 10 successive washes in autoclaved water. Four seeds were sown per pot, and thinning was performed 5 days after emergence, allowing two plants per pot.

For the treatments inoculated with the UFLA strains and SEMIA 5019, the strains were grown in 79 liquid medium (Fred & Waksman, 1928) under constant stirring at 110 rotations per minute (rpm) and a temperature of 28 °C for 5 days (with approximately 1 × 10^8 cells mL^{-1}). Each treatment received 1 mL of the inoculant on each seed. For treatment with the commercial inoculant, 1 mL of the liquid inoculant was used per seed, with a minimum concentration of 5 × 10^9 cells mL^{-1}. Irrigation of the pots was performed by maintaining the moisture at approximately 60% of field capacity.

At 40 days after sowing, corresponding to the beginning of flowering (R1 stage), the chlorophyll *a*, *b*, and *total* contents were evaluated using a Clorofilog chlorophyll meter (Falker CFL 1030, São Paulo, Brazil), collected from the first fully expanded trifoliate leaf at the apex. Subsequently, the plants were collected to evaluate the number of nodules per plant (NN, nodules plant^{-1}), nodule dry mass (NDM, mg plant^{-1}), shoot dry matter (SDM, g plant^{-1}), root dry matter (RDM, g plant^{-1}), shoot nitrogen content (SNC, %), shoot nitrogen accumulation (SNA, mg N plant^{-1}), and relative efficiency (RE, %).

To determine the NDM, SDM, and RDM, the nodules, shoots, and roots were placed in a forced-air oven at 60 °C until they reached a constant weight. The SNA was calculated by multiplying the SDM by the N content in the shoots. The N content in the shoots was determined using the semi-micro Kjeldahl method (Liao, 1981). The relative efficiency of each treatment was calculated according to the expression: RE = (SDM of each treatment*100)/SDM of the treatment with mineral N within the same block.

According to the Shapiro–Wilk test, the data presented a normal distribution and were subjected to analysis of variance by employing a statistical analysis system, SISVAR 5.3 (Ferreira, 2011). The treatment means were compared using the Scott–Knott test at a 5% probability level.

Results and Discussion

A significant interaction was observed between the N sources and types of soils for the NN, NDM, SDM, RE, SNC, and SNA (Table 2). The strains UFLA06-13, UFLA06-15, UFLA06-19, UFLA06-22, and UFLA06-24 provided higher NN when inoculated in the Oxisol (Table 2). However, for the NDM, all inoculated treatments provided higher values in the Oxisol. These results indicate that the nodulation process by strains of *Bradyrhizobium* is influenced by intrinsic characteristics of the soils used in the study, such as the content of organic matter and texture, corroborating previous studies (Bromfield et al., 2017; Chibeba et al., 2017).

Table 1. Chemical and granulometric characterization of the samples of Quartzarenic Neosol and Oxisol used in the experiment, collected from within the 0–20 cm depth layer in the municipalities of Santa Luz and Bom Jesus, respectively, in southwestern Piauí.

Soils	pH	P	H+Al	Al^{3+}	Ca^{2+}	Mg^{2+}	K^+
	(H_2O)	(mg dm^{-3})	(cmol$_c$ dm^{-3})				
Quartzarenic Neosol	4.5	45.23	1.40	0.00	0.31	0.10	0.13
Oxisol	4.6	4.54	7.22	1.80	0.05	0.03	0.04

Soils	SB	T	V	m	OM	Clay	Silt	Sand
	(cmol$_c$ dm^{-3})		(%)		(g kg^{-1})			
Quartzarenic Neosol	0.54	1.94	27.8	0.0	2.6	85	15	900
Oxisol	0.12	7.34	1.6	93.7	21.5	244	11	745

P and K - extractor Mehlich- 1; Ca, Mg, and Al - extractor KCl - 1 mol/L; H + Al - extractor calcium acetate at pH 7.0; SB - sum of bases; CTC (T) - cation exchange capacity at pH 7; V - base saturation; m - aluminum saturation; OM - organic matter, Walkey–Black method.

Table 2. Average values of nodule number (NN), nodule dry mass (NDM), shoot dry matter (SDM), relative efficiency (RE), shoot nitrogen content (SNC), and shoot nitrogen accumulation (SNA) obtained in soybean plants as a function of the different forms of nitrogen supply in soils from southwestern Piauí, in a greenhouse[(1)].

Soils	N Sources	NN (N plant⁻¹)	NDM (mg plant⁻¹)	SDM (g plant⁻¹)	RE (%)	SNC (%)	SNA (mg plant⁻¹)
Quartzarenic Neosol	UFLA06-13	78.37 B b	137.12 B b	2.88 A b	170.71 A a	1.36 B b	39.41 A b
	UFLA06-15	87.37 B b	186.75 A b	3.04 A b	175.94 A a	1.33 B a	40.54 A b
	UFLA06-19	135.87 A b	211.37 A b	3.13 A b	186.50 A a	1.24 B b	38.99 A b
	UFLA06-21	136.00 A a	182.50 A b	3.08 A b	180.57 A a	1.35 B a	41.55 A b
	UFLA06-22	82.25 B b	138.00 B b	2.91 A b	169.07 A a	1.37 B b	40.21 A b
	UFLA06-24	64.37 B b	120.87 B b	2.67 A b	154.26 A a	1.42 B b	37.59 A b
	SEMIA 5019	133.50 A a	218.87 A b	3.36 A b	200.58 A a	1.28 B b	43.40 A b
	Commercial[(2)]	87.87 B a	122.62 B b	2.66 A b	157.56 A a	1.35 B b	39.09 A b
	Without N[(3)]	16.50 C a	54.87 C a	3.73 A b	210.34 A a	1.03 C b	38.38 A b
	With N[(4)]	0.00 C a	0.00 C a	1.72 B b	100.00 B a	3.21 A a	54.95 A b
Oxisol	UFLA06-13	160.37 B a	307.75 A a	4.97 B a	104.39 A b	1.61 C a	80.39 C a
	UFLA06-15	144.12 B a	349.50 A a	5.80 A a	122.17 A b	1.43 C a	82.19 C a
	UFLA06-19	190.12 A a	351.00 A a	5.12 B a	107.44 A b	1.70 C a	87.27 B a
	UFLA06-21	157.75 B a	350.75 A a	4.78 B a	100.40 A b	1.54 C a	72.96 C a
	UFLA06-22	155.62 B a	315.00 A a	4.85 B a	101.80 A b	2.09 B a	101.76 B a
	UFLA06-24	204.25 A a	337.25 A a	5.68 A a	119.74 A a	2.24 B a	127.09 A a
	SEMIA 5019	131.00 B a	370.87 A a	5.74 A a	120.90 A b	1.68 C a	96.79 B a
	Commercial	125.25 B a	280.00 A a	5.45 A a	114.36 A b	1.63 C a	88.51 B a
	Without N	38.62 C a	115.12 B a	5.47 A a	115.59 A b	1.24 D a	67.67 C a
	With N	0.00 D a	0.00 C a	4.82 B a	100.00 A a	2.80 A b	135.21 A a
CV%		24,89	25.59	11.62	17.61	9.80	14.94

[(1)] Means followed by the same letter, uppercase in the columns and within each soil, and lowercase in the columns between the soils, belong to the same group according to the Scott–Knott test at a 5% probability. [(2)] Commercial = commercial inoculant (SEMIA 5079 + SEMIA 5080); [(3)] Without N = without inoculation and without mineral nitrogen; [(4)] with N = without inoculation and with mineral nitrogen (300 mg dm⁻³).

Among the N sources within the soils, it was verified that in the Quartzarenic Neosol, the highest means of NN and NDM were obtained in the plants inoculated with the strains UFLA06-21, UFLA06-19, and SEMIA 5019, in addition to UFLA06-15 for NDM, which was higher than that in other treatments (Table 2). The strains UFLA06-19 and UFLA06-24 induced higher NN values compared to other Oxisol treatments.

Similar to the NN, all inoculated treatments also had a higher NDM than that in the controls with and without N in both soils (Table 2). The absence of nodules observed in this study in the control with N showed the inhibitory capacity of nitrogen on nodulation in soybeans, corroborating the results of Kaschuk et al. (2016). The nodulation that occurred in the control without inoculation and N proves the existence of native rhizobial communities that can form a symbiosis with soybeans in the studied soils.

NN and NDM are important variables for selecting rhizobia because they reflect the competitive capacity and symbiotic efficiency of the strains. Nodulation data, especially NDM, are usually positively correlated with the fixation of N_2 and N accumulation in legumes (Costa et al., 2017a; Sulieman et al., 2019). However, it should be noted that good nodulation does not always reflect the efficiency of the strains because several edaphoclimatic factors can interfere with the survival and efficiency of these microorganisms (Moreira et al., 2010; Kaschuk et al., 2016; Castro et al., 2017).

On evaluating the interaction between factors for SDM, it was verified that all inoculated treatments, as observed for the NDM, induced higher values in the Oxisol (Table 2). The two controls without inoculation were also highlighted in the Oxisol. For the RE, most treatments presented higher values in the Quartzarenic Neosol, except for the control with N and the treatment inoculated with strain UFLA06-24, which induced similar values in both soils.

On evaluating the treatments in the Quartzarenic Neosol, there were no differences between the inoculated treatments and the control without N, which provided higher SDM and RE values compared to the control with N (Table 2). However, in the Oxisol, the highest SDM means were observed in the plants inoculated with strains UFLA06-15, UFLA06-24, and SEMIA 5019, the commercial inoculant, and in the control without N. For the RE, there were no differences between treatments within the Oxisol.

Soil factors, besides the texture and content of organic matter, which differed between soils (Table 1), could influence the performance of the strains in the production of SDM (Moreira et al., 2010; Soares et al., 2014; Costa et al., 2017a). However, better adaptation of the soybean cultivars in the Oxisol should also be considered because even the non-inoculated treatments presented higher production of SDM in this soil. Some studies conducted with rhizobial strains isolated from Oxisols in different regions of Brazil have shown that these soils represent an important genetic resources for obtaining and selecting strains that have efficient N_2 fixation abilities (Marra et al., 2012; Costa et al., 2014; Ribeiro et al., 2015; Coelho et al., 2018). The *Bradyrhizobium* strains used in this study were isolated from nodules of soybeans grown in an Oxisol in the state of Piauí (Cerrado biome) (Ribeiro et al., 2015).

Several factors could also interfere with the availability of nutrients in soils, reflecting higher or lower absorption by the plants (Marschner, 2012). In the present study, it was observed that, in the Quartzarenic Neosol, a dose of 300 mg of N dm^{-3}, even split into three applications, promoted visual symptoms of toxicity in the plants, reflecting the lower SDM and RE (Table 2). This probably occurred because of the low cation exchange capacity of this soil (Table 1), resulting in lower electrostatic adsorption of N, and consequently, greater availability in solution because closed pots were used to avoid leaching. The absence of a response to inoculation for SDM in both soils may be related to the length of the experiment because the cultivar has a short cycle.

On evaluating the interaction between factors for SNC and SNA, the control without N and the inoculated treatments presented higher values in the Oxisol, except for the treatments inoculated with strains UFLA06-15 and UFLA06-21, which produced similar values for the SNC in both soils (Table 2). The control with N induced a higher SNC in the Quartzarenic Neosol, which may have resulted in the toxicity that was visually observed in the plants after this treatment.

The improved performance for SNC and SNA in the Oxisol, which is also expressed in the production of NDM and SDM (Table 2), allows the inference of a direct relationship between these variables. This may be related, in part, to the better adaptation of the strains to this soil because it has closer physical and chemical characteristics to the soil from which they were isolated by Ribeiro et al. (2015). The best adaptation of the cultivar to Oxisol should also be considered, as mentioned above.

The SNC was higher in the control treatment with N compared to all other treatments in both soils (Table 2). In the Quartzarenic Neosol, all inoculations were superior to the control without N. Regarding the Oxisol, strains UFLA06-22 and UFLA06-24 were superior among inoculations, differing from the commercial inoculant by 22.00% and 27.23%, respectively. It is noteworthy that, as verified in the Quartzarenic Neosol, all inoculated treatments were better than the control without N in the Oxisol, indicating the greater efficiency of N_2 fixation by the inoculated strains compared to the native rhizobial communities.

For SNA, as observed for the SDM and SNC within the Quartzarenic Neosol, there were no differences between the inoculated treatments (Table 2). However, there were no differences in the inoculated treatments compared to the two control treatments without inoculation. In the Oxisol, the control with N and the UFLA06-24 strain-induced higher values, promoting an increase of 30% compared to the commercial inoculant. Strains UFLA06-19 and UFLA06-22 were also noteworthy for proportioning SNA similar to the inoculant strain SEMIA 5019 and the commercial inoculant, leading to SNA values higher than those obtained by other strains and the control without N.

For the variables RDM and chlorophyll *a*, *b*, and *total*, there was no significant interaction between factors, although there was an individual effect of N sources and soil types (Table 3). All variables presented higher mean values in the Oxisol, which may be related to the better adaptation of the soybean cultivar to this soil, as shown by most variables presented above (Tables 2).

The RDM of soybean plants inoculated with UFLA06-13 and UFLA06-19 was higher than that of the remaining UFLA strains and the control with N. However, their RDM was similar to that of the SEMIA 5019, the commercial inoculant, and the control without N (Table 3). The higher root production in plants inoculated with strains UFLA06-13, UFLA06-19, SEMIA 5019, the commercial inoculant, and the control without N may be related to the efficiency of these strains and the native rhizobial communities in promoting the root growth of soybean plants by other processes than N_2-fixation.

For the contents of chlorophyll *a, b*, and *total*, plants inoculated with strain SEMIA 5019 were noteworthy among all inoculated treatments and were similar to the control with N for chlorophyll *b* and *total* (Table 3). Except for the plants inoculated with UFLA06-13 and the commercial inoculant, the inoculated treatments presented superior chlorophyll *a* and *total* contents to the control without N. For the content of chlorophyll *b*, all treatments inoculated with UFLA strains and the commercial inoculant showed a similar performance to each other and with the control without N but were worse than the control with N. UFLA06-24 promoted higher contents of chlorophyll *a* and *total* compared to the remaining UFLA strains. It is worth noting that the plants inoculated with strain UFLA06-24 presented the highest SNA in the Oxisol, being equivalent to the control with N (Table 2). N is a chemical component of chlorophyll molecules, and a positive correlation is generally observed between the content of chlorophyll and SNA and/or SNC in legumes (Costa et al., 2017b; Mendes et al., 2020).

Table 3. Average values of root dry matter (RDM) and content of chlorophyll *a* (Chl *a*), *b* (Chl *b*), and *total* (Chl *total*) obtained in soybean plants as a function of the different forms of nitrogen supply and soils from southwestern Piauí, in a greenhouse[(1)].

Factors	RDM (g $plant^{-1}$)	Chl *a*	Chl *b*	Chl *total*
N Sources				
UFLA06-13	3.71 A	28.37 D	9.02 B	37.39 D
UFLA06-15	3.16 B	28.94 C	9.24 B	38.18 C
UFLA06-19	3.83 A	29.20 C	9.39 B	38.59 C
UFLA06-21	3.08 B	29.31 C	9.49 B	38.80 C
UFLA06-22	2.55 B	29.33 C	9.54 B	38.88 C
UFLA06-24	3.16 B	30.07 B	9.71 B	39.97 B
SEMIA 5019	3.82 A	31.41 A	10.62 A	42.03 A
Commercial[(2)]	3.81 A	28.03 D	8.97 B	37.00 C
Without N[(3)]	4.46 A	27.87 D	9.58 B	37.45 D
With N[(4)]	2.66 B	30.41 B	10.94 A	41.04 A
Soils				
Quartzarenic Neosol	2.13 B	28.07 B	8.87 B	36.94 B
Oxisol	4.72 A	30.45 A	10.43 A	40.89 A
CV (%)	28.95	3.22	6.82	4.00

[(1)] Averages followed by the same letter in columns within the nitrogen sources and within the soils belong to the same group by the Scott–Knott test at 5% probability. [(2)] Commercial = commercial inoculant (SEMIA 5079 + SEMIA 5080); [(3)] Without N = without inoculation and without mineral nitrogen; [(4)] With N = without inoculation and with mineral nitrogen, (300 mg dm^{-3}).

B. brasilense species was initially described as including rhizobia strains isolated from *Vigna unguiculata* and *Macroptilium atropurpureum* nodules in Brazilian soils (Costa et al., 2017b). However, a recent study on the *Bradyrhizobium* strains used in this study classified them as *B. brasilense* (Costa et al., 2020). Despite the economic importance of soybeans throughout Brazil, *B. brasilense* is the first species native to Brazilian soils whose strains have been identified to be efficient in fixing nitrogen in symbiosis with soybeans under axenic (Ribeiro et al., 2015) and original soil conditions (in this study). In addition to the strains used in this study, Ribeiro et al. (2015) isolated other *Bradyrhizobium* spp. strains from nodules of soybeans grown in soils from different Brazilian regions that exhibited a high symbiotic efficiency with soybean plants. Other studies conducted on strains from soils in the semiarid region of northeastern Brazil have shown the high symbiotic efficiency of *Bradyrhizobium* spp. strains isolated from nodules of cowpeas, indicating that these soils represent an important repository of symbiotically efficient rhizobia with promising biotechnological potential (Marinho et al., 2017; Oliveira et al., 2020; Sena et al., 2020).

Overall, UFLA06-19, UFLA06-22 (*B. brasilense*), and UFLA06-24 (*Bradyrhizobium* sp.) were the most efficient soybean microsymbionts in the Oxisol. However, it should be noted that the experiment was conducted in a greenhouse. In future studies, these strains will be evaluated under field conditions to confirm their potential for use as inoculants in soybean crops.

Conclusions

All strains showed increased nodulation and shoots nitrogen content of soybean plants in the Quartzarenic Neosol and Oxisol. Most strains presented better symbiotic performance when inoculated in soybeans cultivated in the Oxisol.

The strains UFLA06-19, UFLA06-22 (*Bradyrhizobium brasilense*), and UFLA06-24 (*Bradyrhizobium* sp.) were efficient in shoots nitrogen accumulation in soybeans in the Oxisol, indicating their potential for use as soybean inoculants.

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Compliance with Ethical Standards

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