Universidade Federal Rural do Semi-Árido
Pró-Reitoria de Pesquisa e Pós-Graduação
https://periodicos.ufersa.edu.br/index.php/caatinga

ISSN 0100-316X (impresso)
ISSN 1983-2125 (online)
http://dx.doi.org/10.1590/1983-21252023v36n406rc

Agronomic performance of cowpea cultivars inoculated with rhizobia in the brazilian semiarid region

Desempenho agrônomico de cultivares de caupi inoculadas com rizóbio no semiárido brasileiro

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ABSTRACT - This work aimed to evaluate the agronomic efficiency of cowpea cultivars inoculated with rhizobia strains recommended for cowpea in a semiarid environment. Experiments were carried out in Mossoró, Rio Grande do Norte, in two agricultural cycles: the first from January to April 2019, and the second from June to August of the same year. The experimental design adopted was randomized blocks with four replications arranged in a 4 × 4 x 2 factorial scheme, with treatments consisting of four cowpea cultivars: 'BRS Imponente', 'BRS Itaim', 'BRS Novaera', and 'BRS Tumucumaque' as well as three sources of N, two strains of *Bradyrhizobium* spp. registered for cowpea (BR 3262 and BR 3267) and, the application of mineral N (50 kg ha^{-1}), beyond an absolute control, without inoculation or application of mineral N. In addition to two planting seasons. The cultivar 'BRS Tumucumaque' showed greater productive potential. The inoculation of cowpea seeds with *Bradyrhizobium* strains BR 3262 and BR 3267 provided grain yields similar to those obtained with the application of mineral N.

Keywords: *Vigna unguiculata* L. (Walp). Biological nitrogen fixation. Inoculant.

RESUMO - Este trabalho teve como objetivo avaliar a eficiência agronômica de cultivares de feijão-caupi inoculadas com linhagens de rizóbios recomendadas para feijão-caupi em ambiente semiárido. Os experimentos foram conduzidos em Mossoró, Rio Grande do Norte, em duas safras agrícolas: o primeiro de janeiro a abril de 2019, e o segundo de junho a agosto do mesmo ano. O delineamento experimental adotado foi de blocos ao acaso com quatro repetições em esquema fatorial 4 × 4 x 2, com tratamentos constituídos por quatro cultivares de feijão-caupi: 'BRS Imponente', 'BRS Itaim', 'BRS Novaera' e 'BRS Tumucumaque' e três fontes de N, duas linhagens de *Bradyrhizobium spp*. registradas para o feijão-caupi (BR 3262 e BR 3267), aplicação de N mineral (50 kg ha^{-1}), além de uma testemunha absoluta, sem inoculação ou aplicação de N mineral. Além de duas épocas de plantio. A cultivar 'BRS Tumucumaque' apresentou maior potencial produtivo. A inoculação de sementes de feijão-caupi com as linhagens de *Bradyrhizobium* BR 3262 e BR 3267 proporcionou rendimentos de grãos semelhantes aos obtidos com a aplicação de N mineral.

Palavras-chave: *Vigna unguiculata* L. (Walp). Fixação biológica de nitrogênio. Inoculante.

Conflict of interest: The authors declare no conflict of interest related to the publication of this manuscript.

Received for publication in: May 26, 2023.
Accepted in: August 31, 2023.

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INTRODUCTION

Cowpea (*Vigna unguiculata* (L.) Walp.) is a crop of great importance for the northeastern region of Brazil due to its rusticity, as it adapts to different cultivation conditions and has good tolerance to water deficit when compared to other cultures. However, due to the low technological level used in production systems and the low availability of nutrients in tropical soils, especially nitrogen (CAVALCANTE et al., 2017), the Northeast region has productivity below the other producing regions of the country (EMBRAPA ARROZ E FEIJÃO, 2019).

Nitrogen (N) is an element highly demanded by crops, so it is the most limiting nutrient for plant growth (MICHEL et al, 2020). Legumes can interact symbiotically with bacteria that have the enzyme nitrogenase, known as rhizobia, and can fix atmospheric nitrogen through biological nitrogen fixation (BNF) (LEITE et al., 2018).

There are currently four strains of the genus *Bradyrhizobium* spp. authorized for use in inoculants in cowpea culture: UFLA 3-84 (SEMIA 6461), BR 3267 (SEMIA 6462), INPA 03-11B (SEMIA 6463), and BR 3262 (SEMIA 6464). These strains are part of the composition of inoculants that are normally sold in peat or liquid medium (BRASIL, 2011; SILVA JÚNIOR et al., 2014).

According to Brito, Muraoka and Silva (2011), BNF can fully replace nitrogen fertilization in cowpea culture. However, the interaction of culture with rhizobia is of low specificity, with a large number of native bacteria capable of nodulating the culture (SILVA et al., 2012). Native rhizobia, in addition to having variable efficiency in terms of BFN (BORGES et al., 2012), compete with the strains selected during nodulation (COSTA et al., 2014), generating inconsistent data on the efficiency of inoculation with rhizobia in the culture.

The evaluation of the interaction between macro and microsymbionts to determine efficient combinations between cultivars and strains of rhizobia may allow the most adequate indication of specific inoculants for the cowpea genotypes (MARINHO et al., 2014); however, the selected strains have not been tested with most cultivars under the edaphoclimatic conditions of the semiarid region of Northeast Brazil.

In this sense, it is essential to evaluate the agronomic efficiency of strains recommended for cowpea in different cultivars and planting regions. Therefore, this study aimed to evaluate the agronomic efficiency of four cowpea cultivars inoculated with rhizobia strains recommended for cultivation in a semiarid environment in two agricultural cycles.

MATERIAL AND METHODS

Site description

The experiments were conducted in the field, in two agricultural cycles, at the Rafael Fernandes Experimental Farm, located in the district of Alagoinha, Mossoró, Rio Grande do Norte, Brazil. The first crop was conducted from January to April 2019, and the second, from June to August of the same year. The experimental area is located at 5° 03’ 37” S; 37° 23’ 50” W, with an altitude of approximately 72 m.

According to the Köppen classification, the climate of the region is of the BSh type, that is, hot and very dry, with two climatic seasons: a rainy one that usually runs from February to May, and a dry one, from June to January (ALVARES et al., 2013). The climatic data of the experimental area are shown in Figure 1.

The soil in the experimental area was classified as Oxisol Red Yellow Argisolic, sandy loam (SANTOS et al., 2013), whose chemical analysis, from the 0–0.20 m layer, presented the following characteristics for the first crop: pH (H_2O) was 6.4, P was 7.5 mg dm^{-3}, K was 36.9 mg dm^{-3}, Na was 17 mg dm^{-3}, Ca was 0.8 cmol$_c$ dm^{-3}, Mg was 0.3 cmol$_c$ dm^{-3}, and Al was 0.00 cmol$_c$ dm^{-3}. The second crop had the following values: pH H_2O was 6.9, P was 14.1 mg dm^{-3}, K was 39.1 mg dm^{-3}, Na was 7.3 mg dm^{-3}, Ca was 1.1 cmol$_c$ dm^{-3}, Mg was 0.4 cmol$_c$ dm^{-3}, and Al was 0.00 cmol$_c$ dm^{-3}.

The first experiment was planted on January 29th and harvested on April 20th, 2019. A time that comprises the rainy season in the region. While the second experiment was planted on June 13th and harvested on August 29th, 2019. Being carried out within the dry season in the region. Totaling 81 and 77 days of cycles in the rainy and dry season, respectively.

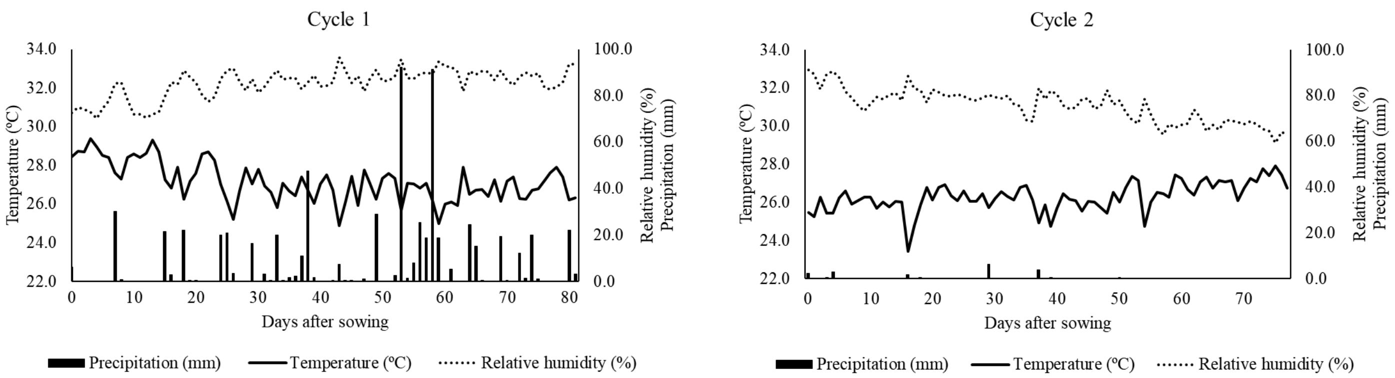

Figure 1. Climatic characterization of the experimental area in both agricultural crops. Mossoró, RN, Brazil. 2019.

Experimental design and treatments

The experimental design adopted was randomized blocks arranged in a 4 × 4 x 2 factorial scheme, with four replications. The treatments consisted of four cowpea cultivars (‘BRS Imponente’, ‘BRS Itaim’, ‘BRS Novaera’, and ‘BRS Tumucumaque’), four sources of N, and two strains of bacteria of the genus *Bradyrhizobium*: BR 3267 (SEMIA 6462) and BR 3262 (SEMIA 6464), both authorized for use in inoculants in the cowpea culture (BRASIL, 2011), in addition to two controls, one with a supply of mineral N in the dose of 50 kg of N ha^{-1}, referring to the recommended dose for the culture (LOPES et al., 2008) and another without application of N. In addition to two planting seasons.

The plots consisted of four planting lines 4 m long, spaced 0.5 m apart. The spacing adopted between pits was 0.20 m, with two plants per pit, resulting in a population density of 200,000 plants ha^{-1}. The useful area considered in each plot was constituted by the two central rows, discarding two plants at each end, corresponding to 72 plants in an area of 3.6 m^2.

Crop management

The soil preparation consisted of plowing and two harrows; the first was carried out shortly after plowing and the second one occurred the day before sowing. The management of fertilization was carried out based on the results of analyses of soil samples collected from the experimental area before the installation of each experiment and in compliance with the recommendation of Lopes et al. (2008) for the cultivation of irrigated cowpea, so that all plots received phosphate (40 kg ha^{-1} and 20 kg ha^{-1} of P_2O_5 in the first and second crops, respectively) and potassium (40 kg ha^{-1} of K_2O in both crops), whose sources were simple superphosphate and potassium chloride, respectively. The application of N in the plots was made according to the respective treatments, with urea being used as a source of mineral N. The plots whose treatment corresponded to the application of mineral N received 20 Kg of N ha^{-1} in the foundation and 30 Kg of N ha^{-1} in the top dressing, 25 days after sowing, through fertigation.

The inoculants containing the rhizobia strains were obtained from Embrapa Agrobiologia in a peat medium with a minimum concentration of 109 bacterial cells g^{-1} of inoculant. The inoculation process was carried out following the recommendation of Auras et al. (2018) for inoculating the cowpea culture with rhizobia in a solid peat medium.

The drip system used was drip irrigation, with 2 L h^{-1} flow emitters spaced 0.20 m and was maintained daily until harvesting the pods.Weed management was carried out through manual weeding so that the plants were kept clean for as long as possible, to minimize the interference of the weed community on the crop.

Harvest and evaluated variables

The planting took place through manual sowing, at a depth of approximately 0.05 m, placing four seeds per hole. At ten days after emergence (DAE), thinning was carried out, leaving only two plants per hole.

At 40 days after sowing (DAS), eight random plants were collected from the useful area of each plot, maintaining the integrity of the roots, and taken to the laboratory to evaluate the characteristics related to nodulation: number of nodules (NN); nodule dry matter (NDM); aerial part dry matter (APDM), and relative efficiency (RE).

For NN evaluation, the plant nodules were detached from the roots and later counted. Then, the nodules were washed and dried in an oven with forced air circulation at a temperature of 65°C, until a constant mass was obtained, and weighed in a precision analytical balance, to determine NDM.

For the aerial part evaluations, the collected plants were placed in an oven with forced air circulation at a temperature of 65 °C until a constant mass was obtained, for APDM determination. The RE was calculated using the formula proposed by Bergensen et al. (1971):

$$RE = \frac{APDM_{\ inoculated}}{APDM\ N_{mineral}} \times 100$$

where RE is relative efficiency (%), $APDM_{inoculated}$ is the dry matter of the aerial part of the inoculated material (g), and $APDM\ N_{mineral}$ is the dry matter of the aerial part of the material with the application of mineral N (g).

At the end of the crop cycle, dried pods were harvested and evaluated for pod length (PL), the number of grains per pod (NGP), the weight of 100 grains (W100G), grain yield (GY), and grain index (GI). The PL and NGP were measured through the random collection of ten pods, which were measured with the aid of a graduated ruler, and then were threshed to perform the grain count. The GY was determined after threshing the pods, followed by the weighing of all grains. To standardize the humidity of the harvested grains, a correction of humidity to 13% was carried out. The W100G was obtained by weighing a sample of 100 random grains from each plot. The GI was calculated through the relationship between the weight of the grains and the weight of the pods.

Statistical analysis

The data obtained were submitted to analysis of variance using the statistical software Sisvar® v. 5.3 (FERREIRA, 2008) for each experiment in isolation. Observing the assumptions for homogeneity of variances and normality of errors between the experiments, it was submitted to joint analysis of variance, the means being compared by the Tukey test at 5% probability.

RESULTS AND DISCUSSION

The summary of the variance analysis of the characteristics evaluated is presented below (Table 1). There was a significant effect of the triple interaction Crop x Cultivar x N source for dry matter of nodules and grain yield. Interaction Cultivar x N source for the number of nodules character. Interaction Cultivar x Crop for dry matter of the aerial part, relative efficiency and number of grains per pod. For the grain index, a double interaction N source x Crop was observed. And an isolated effect of Crop and Cultivar was verified for weight of 100 grain. And isolated Cultivar effect for length of podes.

There was no difference in the number of nodules between the cultivars, except for ‘BRS Novaera’, which had statistically inferior nodulation compared to the others in the second crop. The timing of the agricultural cycle also affected the nodulation of the cultivars, so that in the second, ‘BRS Imponente’ and ‘BRS Novaera’ presented less nodulation, whereas ‘BRS Itaim’ and ‘BRS Tumucumaque’ were not influenced by this factor (Table 2).

The efficiency of biological nitrogen fixation (BNF) can vary according to the cultivars studied, due to the genetic variability of macrosymbionts. Similar results were found by Costa et al. (2014) when evaluating the agronomic efficiency of different strains of rhizobia in cowpea cultivars in Bom Jesus, Piauí, Brazil. In this study, the authors also observed different nodulations between the cultivars studied.

Table 1. Summary of joint analysis of variance number of nodules (NN), dry matter of nodules (DMN), dry matter aerial part (DMAP), relative efficience (RE), length of podes (LP), number of grains per pod (NGPP), grins yield (GY), weight of100 grain (W100G) and grain index (GI) of cowpea cultivars inoculated with different strains of rhizobia.

SV	DF	Means Square								
		NN	DMN	DMAP	RE	LP	NGPP	GY	W100G	GI
Block (crop)	6	161.24	8335.69	4.25	1660.34	2.22	2.36	766600.34	7.61	2.62
Crop (Cr)	1	439.45**	314546.04**	545.37**	1540.37^{ns}	3.09^{ns}	69.60**	18133016.27**	449.06**	134.46**
Cultivar (C)	3	376.66**	32146.20**	25.80**	1155.36^{ns}	128.51**	21.35**	1310583.46**	357.43**	96.90**
N Source (N)	3	861.66**	39089.04**	14.58**	2628.63**	1.74^{ns}	2.59^{ns}	496004.03**	10.16^{ns}	3.70^{ns}
C x Cr	3	93.43**	6796.98*	30.64**	3546.65**	2.95^{ns}	4.12**	601453.68**	18.94^{ns}	23.70**
N x Cr	3	112.27*	11505.81**	1.90^{ns}	500.95^{ns}	2.14^{ns}	1.42^{ns}	719217.88**	30.08^{ns}	14.16**
C x N	9	43.64*	4835.23*	2.42^{ns}	268.95^{ns}	1.50^{ns}	0.98^{ns}	178025.27^{ns}	13.31^{ns}	4.09^{ns}
Cr x C x N	9	61.04^{ns}	6457**	2.57^{ns}	466.87^{ns}	1.06^{ns}	1.40^{ns}	347435.12**	21.27^{ns}	5.21^{ns}
Error	90	33.38	1919.30	1.63	448.10	1.12	1.01	100585.01	23.00	2.64
CV (%)		22.48	23.38	17.06	22.15	5.93	10.93	23.04	20.57	2.10

Table 2. Average values for the number of nodules of cowpea cultivars inoculated with different strains of rhizobia.

Cultivar/N source	Number of Nodules (nodules plant^{-1})	
	Crop 1	Crop 2
BRS Imponente	29.44 Aa	24.21 Ab
BRS Itaim	25.52 Aa	25.00 Aa
BRS Novaera	25.15 Aa	17.36 Bb
BRS Tumucumaque	30.09 Aa	28.82 Aa
Abscence of N	26.03 Ba	21.75 BCb
BR 3267	34.91 Aa	26.14 ABb
BR 3262	29.44 Ba	28.90 Aa
N mineral	19.82 Ca	18.59 Ca

Means followed by the same capital letter, in columns, and lower case in lines, do not differ by Tukey's test at 5% probability.

Biological nitrogen fixation is a process that is influenced by edaphoclimatic factors, which can bring benefits or losses to the process (MELO; ZILLI, 2009), such as those observed by Zilli et al. (2013), where the authors observed a reduction in the rhizobia density in the soil during the dry season in the Cerrado of Roraima, Brazil. Thus, possibly the environmental conditions, especially the lower rainfall found in the second crop, limited the nodulation of the cultivars 'BRS Imponente' and 'BRS Novaera', justifying the lower values obtained by these cultivars.

In both crops, the largest number of nodules was promoted by strains of rhizobia, while the lowest nodulation was promoted by the application of mineral N. The nodulation promoted by the control without application of N and with the application of mineral N reflects the presence and capacity of nodulation of the native community of rhizobia, varying from 18.59 nodules per plant, in treatments with an application of mineral N, to 26.03 nodules per plant, with the control without inoculation (Table 3). The greater nodulation promoted by the witness suggests that the application of mineral N restricts the nodulation promoted by the native rhizobia community.

Table 3. Number of nodules of cowpea cultivars inoculated with different strains of rhizobia.

N Source	Number of Nodules (nodules plant^{-1})			
	Absence of N	BR 3267	BR 3262	N mineral
BRS Imponente	25.00 Abc	32.97 ABa	28.92 ABab	20.40 Ac
BRS Itaim	24.73 Aab	28.46 BCa	26.95 Bab	20.90 Ab
BRS Novaera	19.95 Aab	24.06 Ca	25.09 Ba	15.92 Ab
BRS Tumucumaque	25.86 Ab	36.61 Aa	35.73 Aa	19.62 Ab

Means followed by the same letter, uppercase in the columns and lowercase in the rows, do not differ by Tukey's test at 5% probability.

According to Brito, Muraoka and Silva (2011) under the condition of high availability of mineral N, there is a reduction in BNF. This is because BNF is a process that depends on the activity of the enzyme nitrogenase, which in turn demands large amounts of adenosine triphosphate (ATP) as an energy source, making it a metabolically costly process for plants. Thus, there is a preference by plants for metabolic pathways with lower energy costs (RAVEN; EVERT; EICHHORN, 2014).

Regarding rhizobial strains, the results showed a wide capacity to promote nodulation in cowpea cultivars, where the number of nodules varied from 26.14 to 34.91 nodules per plant, both values obtained with the BR 3267 strain in the second and first crop, respectively. The strain BR 3262 had less variation relative to the crops (29.44 and 28.90 nodules per plant for the first and second crop, respectively) showing greater adaptability to the different edaphoclimatic conditions of each cultivation (Table 3).

In general, 'BRS Imponente' and 'BRS Tumucumaque' stood out with a greater number of nodules for both strains, while 'BRS Itaim' and 'BRS Novaera' achieved lower values for this characteristic. In the treatments without inoculation, no statistical differences were observed between the cultivars (Table 3), indicating the equal capacity of the cultivars in nodules with the community of native rhizobia.

Possibly, the greater nodulation achieved by 'BRS Tumucumaque' and 'BRS Imponente' with the rhizobial strains is associated with the specificity of strains in BNF. According to Costa et al. (2011), the symbiotic specificity of a rhizobia strain refers to its ability to induce the formation of nodules and fix N^2 when associated with specific hosts.

The dry matter of the nodules obtained in the first crop was higher than the second (Table 4). The greater dry matter of the nodules, especially in treatments without inoculation, indicates a greater contribution of the native rhizobia community in the formation of nodules during the first crop. In the first crop, 'BRS Tumucumaque' obtained greater dry matter from the nodules for all N sources, showing the great capacity that this cultivar has in establishing a symbiotic relationship with diazotrophic bacteria. In the second crop, in turn, the cultivars did not differ in terms of nodulation due to N sources, except 'BRS Novaera', which obtained less dry matter from the nodules when inoculated with the BR 3262 strain (Table 4).

Table 4. Dry matter of nodules of cowpea cultivars inoculated with different strains of rhizobia.

Crop	Cultivar	Dry matter of nodules (mg planta^{-1})			
		N source			
		Absence of N	BR 3267	BR 3262	N mineral
1	BRS Imponente	247.09 Bab	311.55 Aa	182.75Bbc	111.78 Bc
	BRS Itaim	313.70 ABa	212.33 BCb	233.32 Bab	176.47 ABb
	BRS Novaera	262.96 ABa	156.33 Cc	239.45 Bab	160.29 Bbc
	BRS Tumucumaque	332.42 Aab	241.37 ABc	356.08 Aa	253.09 Abc
2	BRS Imponente	125.85 Aa	148.97 Aa	165.33 ABa	116.19 Aa
	BRS Itaim	160.07 Aab	170.46 Aab	195.96 Aa	93.73 Ab
	BRS Novaera	104.22 Aa	116.17 Aa	110.28 Ba	64.43 Aa
	BRS Tumucumaque	144.55 Aa	186.82 Aa	186.69 ABa	114.95 Aa

Averages followed by the same capital letter compares the cultivars for each source of N, within each cycle, and means followed by the same lowercase letter in the lines compares the sources of N for each cultivar, in each cycle, by Tukey's test at 5% probability.

As for the sources of N, in the first crop, the lowest dry matter of the nodules was promoted by the application of mineral N in all cultivars studied (Table 4). This behavior is due to the inhibition effect of nodulation promoted by the application of mineral N (RAVEN; EVERT; EICHHORN, 2014) on the native rhizobial community, which resulted in less nodulation and, consequently, less dry matter in the nodules. In the second crop, there was an effect of N sources only in the cultivar 'BRS Itaim', where the application of mineral N promoted the lowest value for this characteristic (Table 4).

The results found are similar to those obtained by Melo and Zilli (2009) in Boa Vista, Roraima, Brazil when evaluating the BNF in five improved cowpea cultivars. In this study, the authors observed the dry matter of the nodules ranging from 90.4 to 216.1 mg per plant, with the application of mineral N and inoculation with the strain BR 3262, respectively.

There was a higher production of dry matter in the aerial part in the first crop, in all cultivars. On this occasion, the cultivar 'BRS Imponente' reached 12.12 g per plant, followed by 'BRS Tumucumaque', with 9.59 g per plant. 'BRS Novaera' and 'BRS Itaim' did not differ from each other; however, they presented the lowest values for this characteristic (8.27 and 8.21 g per plant, respectively). In the second crop, in turn, no statistical differences were found between cultivars (Table 5).

Table 5. Dry matter of the aerial part and relative efficiency of cowpea cultivars inoculated with different strains of rhizobia.

Cultivar	Dry matter of the aerial part ($g\ plant^{-1}$)		Relative efficiency (%)	
	Crop 1	Crop 2	Crop 1	Crop 2
BRS Imponente	12.12 Aa	5.14 Ab	109.70 Aa	84.58 Ab
BRS Itaim	8.21 Ca	5.67 Ab	76.59 Bb	97.01 Aa
BRS Novaera	8.27 Ca	4.97 Ab	101.00 Aa	99.70 Aa
BRS Tumucumaque	9.59 Ba	5.88 Ab	108.90 Aa	87.15 Ab

Means followed by the same letter, uppercase in the columns and lowercase in the rows, do not differ by Tukey's test at 5% probability.

The higher values found in the first crop are probably associated with greater efficiency of the BNF at that time, since the greater number and dry matter of the nodules were obtained in the first crop, suggesting a greater N input to the plants. In addition, the high rainfall reached during the crop cycle in the first crop (Figure 1) may have contributed to this result since, under high water availability, there may be greater vegetative growth of cowpea.

Among the cultivars, it was observed that the values for relative efficiency in the first crop were higher than 100%, except for ‘BRS Itaim’ (76.59%), indicating the good response of the cultivars to alternative sources to the application of mineral N. In the second crop, there were no differences between cultivars. Among the crops, it was observed that, except for ‘BRS Novaera’, which did not differ statistically due to the crops, changes in the response were observed in the other cultivars. For ‘BRS Imponente’ and ‘BRS Tumucumaque’, the highest values were obtained in the first crop, while for the second crop ‘BRS Itaim’ that stood out (Table 5).

As for the effect of sources of N on the dry matter of the aerial part, the rhizobial strains, together with the application of mineral N, stood out with higher values, whereas the control without application of N promoted lower values for this characteristic (Table 6).

Table 6. Dry matter of the aerial part and relative efficiency of cowpea cultivars inoculated with different strains of rhizobia.

N source	Dry matter of the aerial part ($g\ plant^{-1}$)	Relative efficiency (%)
Absence of N	6.53 B	82.93 B
BR 3267	8.03 A	103.76 A
BR 3262	7.50 A	95.63 AB
N mineral	7.88 A	100.00 A

Means followed by the same letter in the columns do not differ, by Tukey's test at 5% probability.

As for relative efficiency, no statistical differences were observed between rhizobia strains and the application of mineral N. The values obtained with the rhizobial strains were 95.63% and 103.76% for BR 3262 and BR 3267, respectively (Table 6). This result points to the capacity of rhizobia strains to promote increases in the biomass production of the aerial part of the plants relative to the control, similar to that obtained with the application of mineral N. Similar results were found by Gualter et al. (2011), where the authors observed that the relative efficiency of the inoculated treatments was statistically similar to the application of mineral N.

‘BRS Tumucumaque’ reached the longest pod lengths, with 20.80 cm, followed by ‘BRS Imponente’ and ‘BRS Itaim’ (17.44 and 17.16 cm, respectively), which did not differ between themselves, and ‘BRS Novaera’ (16.20 cm) which presented the shortest length among the tested cultivars (Table 7). The values obtained are similar to those found by Silva and Neves (2011) evaluating production components of cowpea genotypes that obtained the length of pods ranging from 17.60 cm to 21.80 cm.

For all cultivars, the highest number of grains per pod was obtained in the first crop, indicating that this characteristic was influenced by the environment. In the first crop, the highest number of grains per pod was achieved by the cultivar ‘BRS Tumucumaque’ (11.46 grains per pod), being statistically superior to the other cultivars, which did not differ from each other (Table 7).

Despite not differing from the other cultivars in the first crop, with the exception of BRS Tumucumaque, ‘BRS Imponente’ obtained a lower number of grains per pod in the second agricultural crop. This result is possibly associated with the characteristics of this cultivar, the first launched on the national market with extra-large grains, evidenced by the larger size at the expense of the number of grains.

Table 7. Pod length and grain number per pod of cowpea cultivars inoculated with different strains of rhizobia.

Cultivar	Pod length (cm)	Number of grains per pod (grains pod^{-1})	
		Crop 1	Crop 2
BRS Imponente	17.44 B	8.89 Ba	7.56 Bb
BRS Itaim	17.16 B	9.76 Ba	8.88 Ab
BRS Novaera	16.20 C	9.64 Ba	8.46 ABb
BRS Tumucumaque	20.80 A	11.46 Aa	8.95 Ab

Means followed by the same letter, uppercase in the columns and lowercase in the rows, do not differ by Tukey's test at 5% probability.

The weight of 100 grains suffered only the isolated effect of crops and cultivars. 'BRS Imponente' stood out among the other cultivars with the highest weight of 100 grains (27.69 g), while 'BRS Itaim' reached the lowest values for this characteristic (19.68 g) (Table 8). These results indicate that all cultivars reached satisfactory grain size for the market requirements, which according to Silva and Neves (2011), have a preference for genotypes with 100-grain weights over 18 g, however, the authors also highlight the grain size for the commercial price of the product, which has a preference for larger grains.

Table 8. Weight of 100 grains of cowpea cultivars inoculated with different strains of rhizobia.

Cultivar	Weight of 100 grains (g)		AVERAGE
	Crop 1	Crop 2	
BRS Imponente	25.80	29.58	27.69 A
BRS Itaim	18.88	20.47	19.68 C
BRS Novaera	21.03	26.14	23.59 B
BRS Tumucumaque	20.05	24.55	22.30 BC
Average	21.44 b	25.18 a	

Means followed by the same letter, uppercase in the columns and lowercase in the rows, do not differ by Tukey's test at 5% probability.

Among the crops, it was observed that in the second, the weight of 100 grains was 17.44% higher than the first (Table 8). This result indicates a greater filling of grains, and possibly fewer grains per pod found in the second crop contributed to this result.

In general, the grain yield achieved in all treatments was high relative to the national average productivity, ranging from 574.07 kg ha^{-1} to 2,562.42 kg ha^{-1} (Table 9). This result may be associated with the technological level employed in this study, characterized by the cultivars used, which have high productive potential compared to those normally used, the practice of irrigation and phosphate, and potassium fertilization. In addition, even treatments without the application of mineral N achieved considerable nodulation, showing the good response of native rhizobial strains to BNF.

Table 9. Grain yield of cowpea cultivars inoculated with different strains of rhizobia.

Crop	Cultivar	Grain yield (kg ha^{-1})			
		N source			
		Absence of N	BR 3267	BR 3262	N mineral
1	BRS Imponente	1141.36 Aab	1170.56 Aa	574.07 Bb	690.57 Bab
	BRS Itaim	943.95 Aab	875.62 Aab	1361.60 Aa	725.45 Bb
	BRS Novaera	948.46 Aab	1061.22 Aab	1379.93 Aa	740.20 Bb
	BRS Tumucumaque	987.07 Aa	958.73 Aa	967.21 ABa	1474.18 Aa
2	BRS Imponente	1315.51 Aa	1344.14 Ca	1417.93 Ba	1786.90 Aa
	BRS Itaim	1345.98 Ab	2087.68 ABa	1608.44 Bab	2144.61 Aa
	BRS Novaera	1206.06 Aa	1683.10 BCa	1580.22 Ba	1692.27 Aa
	BRS Tumucumaque	1663.72 Ab	2562.42 Aa	2331.97 Aa	2273.47 Aa

Averages followed by the same capital letter compares the cultivars for each source of N, within each cycle, and means followed by the same lowercase letter in the lines compares the sources of N for each cultivar, in each cycle, by Tukey's test at 5% probability.

In treatments without inoculation, in both crops, there were no differences between cultivars, except for the application of mineral N in the first crop, where 'BRS Tumucumaque' stood out from the other cultivars that did not differ from each other. Among the rhizobial strains, in the first crop, no statistical differences were found between cultivars when inoculated with strain BR 3267. With strain BR 3262, cultivars 'BRS Itaim' and 'BRS Novaera' stood out relative to 'BRS Imponente'. Still, in the first crop, 'BRS Tumucumaque' showed similar productivity to the others, in addition to higher productivity for both strains of rhizobia in the second crop (Table 9).

The behavior of cultivars varied according to the sources of N. In the first crop, the application of mineral N did not result in productivity increases compared to the other sources of N, whereas in the second, the cultivars 'BRS Itaim' and 'BRS Tumucumaque' responded positively to inoculation with rhizobial strains and the application of mineral N, differing statistically from the control without application of N. Compared the control, inoculation with strain BR 3267 promoted increments of 55.1% and 54.01% in the BRS cultivars Itaim and 'BRS Tumucumaque', respectively (Table 9).

As for the grain index, the cultivars obtained higher values for this characteristic in the second crop, except for 'BRS Tumucumaque', which did not differ due to the crops. Among the sources of N, the highest values for this characteristic were obtained in the second crop, except for strain BR 3267, which was not influenced by this factor (Table 10).

Table 10. Grain index of cowpea cultivars inoculated with different strains of rhizobia.

Cultivar/N source	Grain index (%)	
	Crop 1	Crop 2
BRS Imponente	75.46 Bb	77.49 Ba
BRS Itaim	77.80 Ab	81.94 Aa
BRS Novaera	76.04 Bb	78.14 Ba
BRS Tumucumaque	76.00 Ba	75.93 Ca
Abscence of N	76.42 ABb	78.16 Aa
BR 3267	77.68 Aa	77.98 Aa
BR 3262	75.79 Bb	78.72 Aa
N mineral	75.42 Bb	78.64 Aa

Means followed by the same letter, uppercase in the columns and lowercase in the rows, do not differ by Tukey's test at 5% probability.

In general, all cultivars showed grain index values above 75% (Table 10), indicating that there was a good translocation of the photoassimilates from the pods intended for filling the grains. However, the highest grain rates achieved in the second crop indicate that there was a greater filling of grains at this time of planting, evidenced by the greater productivity and 100-grain weights achieved.

'BRS Itaim' stood out from the other cultivars with the highest grain index in both crops, while 'BRS Tumucumaque' achieved the lowest values for this characteristic. Among the sources of N, it was observed that in the first crop, the highest grain index was promoted by strain BR 3267, standing out from the other sources of N, which did not differ statistically from each other. In the second crop, no statistical differences were identified between the respective sources of N (Table 10).

CONCLUSIONS

The cultivar 'BRS Tumucumaque' showed higher grain yield potential.

The inoculation of cowpea seeds with strains BR 3262 and BR 3267 of *Bradyrhizobium* spp. was able to provide grain yield similar to that obtained with the application of mineral N fertilizer.

ACKNOWLEDGEMENTS

The authors would like to thank the Coordenação de Aperfeiçoamento de Pessoal de Nível Superior (CAPES) for the financial support by granting scholarships to the authors of this work.

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