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Azospirillum brasilense in corn grown single and intercropped with *Urochloa* in two contrasting soils

Abstract – The objective of this work was to evaluate the agronomic performance of corn (*Zea mays*) grown single and intercropped with *Urochloa ruziziensis* in succession to soybean (*Glycine max*) inoculated with *Bradyrhizobium japonicum*, as well as the inoculation and reinoculation of corn with *Azospirillum brasilense*, in two contrasting soils. The experiment was carried out in two crop years, in a randomized complete block design, in a 2x2x3 factorial arrangement, with five replicates. The first evalauted factor was composed of the two constrasting soils: a clayey Rhodic Eutrustox and a sandy Rhodic Haplustox. The second factor was corn cultivation: single and intercropped with *U. ruziziensis*. The third factor consisted of the inoculation onto soybean and corn in succession, as follows: no inoculation in both crops, inoculation with *B. japonicum* onto soybean and with *A. brasilense* onto corn, and reinoculation with *B. japonicum* + *A. brasilense* onto soybean and with *A. brasilense* onto corn. In both crop years, the highest corn yields are observed in clayey soil with seeds reinoculated with *A. brasilense*. The annual reinoculation with *A. brasilense* results in a higher corn grain yield in the intercropping with *U. ruziziensis* in the clayey soil. Inoculation and reinoculation reduce corn yield losses caused by the competition with *U. ruziziensis* in intercropping.

Index terms: *Bradyrhizobium japonicum*, *Zea mays*, cultivation systems, growth-promoting bacteria, inoculation.

Azospirillum brasilense em milho solteiro e consorciado com *Urochloa* em dois solos contrastantes

Resumo – O objetivo deste trabalho foi avaliar o desempenho agronômico de milho (*Zea mays*) solteiro e consorciado com *Urochloa ruziziensis* em sucessão à soja (*Glycine max*) inoculada com *Bradyrhizobium japonicum*, bem com a inoculação e a reinoculação do milho com *Azospirillum brasilense*, em dois solos contrastantes. O experimento foi realizado em duas safras, em delineamento de blocos ao acaso, em arranjo fatorial 2x2x3, com cinco repetições. O primeiro fator avaliado foi composto pelos dois solos contrastantes: Latossolo Vermelho eutrófico argiloso e Latossolo Vermelho distrófico arenoso. O segundo fator foi o cultivo de milho: solteiro e consorciado com *U. ruziziensis*. Já o terceiro fator consistiu na inoculação em soja e milho em sucessão, como a seguir: sem inoculação nos dois cultivos, inoculação com *Bradyrhizobium japonicum* em soja e com *A. brasilense* em milho, e reinoculação com *B. japonicum* + *A. brasilense* em soja e com *A. brasilense* em milho. Nos dois anos de cultivo, as maiores produtividades

Odair Honorato de Oliveira[1 ✉],
Gessi Ceccon[2],
Denise Prevedel Capristo[1],
Ricardo Fachinelli[3] and
Amanda Gonçalves Guimarães[1]

[1] Universidade Federal da Grande Dourados, Programa de Pós-Graduação em Agronomia, Rodovia Dourados/Itahum, Km 12, Unidade II, Caixa Postal 364, CEP 79804-970 Dourados, MS, Brazil.
E-mail: odairhonorato2020@gmail.com, denise_prevedel@hotmail.com, amandaguimaraes@ufgd.edu.br

[2] Embrapa Agropecuária Oeste, BR 163, Km 253,6, Caixa Postal 449, CEP 79804-970 Dourados, MS, Brazil.
E-mail: gessi.ceccon@embrapa.br

[3] Universidade Federal da Grande Dourados, Departamento de Ciência Agrárias, Rodovia Dourados/Itahum, Km 12, Unidade II, Caixa Postal 364, CEP 79804-970 Dourados, MS, Brazil.
E-mail: rfachinelli@hotmail.com

✉ Corresponding author

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de milho são observadas no solo argiloso com sementes reinoculadas com *A. brasilense*. A reinoculação anual com *A. brasilense* resulta em maior produtividade de grãos de milho no consórcio com *U. ruziziensis* no solo argiloso. A inoculação e a reinoculação reduzem as perdas de produtividade do milho causadas pela competição com *U. ruziziensis* no consórcio.

Termos para indexação: *Bradyrhizobium japonicum*, *Zea mays*, sistemas de cultivo, bactérias promotoras do crescimento, inoculação.

Introduction

Brazil is one of the largest producers of grains, meat, and fibers due to the incorporation of pastures into the country's production systems. However, for a sustainable production of the by-products of grains and pastures, it is important to use cultivation techniques that aim to maintain the balance of the environment (Silva et al., 2021). In this scenario, intercropping emerges as a viable alternative since more than one species is cultivated in an area, contributing to improvements in soil physical, chemical, and biological characteristics through nutrient cycling, which allows an increase in the yield of the crop grown in succession.

Intercropping between forage and cereal species, such as *Urochloa* spp. and corn (*Zea mays* L.), has shown excellent results in improving the physicochemical quality of the soil, which is attributed to the residual effect of the organic matter deposited on its surface. According to Méndez et al. (2019), the consortium between corn and tropical forage grasses brings benefits to the chemical and biochemical parameters of the soil that are related to carbon cycling in the surface layer and to mycorrhizal symbiosis, allowing a greater cycling of nutrients.

Since two grasses are usually used in intercropping, when there is a high C/N ratio, there will be a deficiency in N, which is one of the main elements linked to grain production (Manevski et al., 2015). The high demand for this nutrient by the plant and its low availability in Brazilian soils has made N fertilization an indispensable practice, in which inorganic fertilizers stand out as the main way of adding the nutrient to the soil (Dartora et al., 2013). Duarte & Cantarella (2007) found that an average of 30 to 50 kg ha^{-1} N is applied after soybean cropping due to the biological N fixation by the crop and to the small chance of nitrate loss by leaching by the corn crop cultivated under a lower rainfall volume in the study period. However, new techniques should be incorporated for the rational use of those fertilizers since the demand for N is high in intercropping between forages and the cost of the industrial process for the production of N fertilizers is also high (Dartora et al., 2013).

A sustainable alternative to influence plant development are plant-growth promoting bacteria, which are diazotrophic and show the ability of reducing atmospheric nitrogen (N_2) to ammonia (NH^{+3}) by breaking the triple bond of N with the enzyme nitrogenase (Taiz et al., 2017). Of these bacteria, those of the genera *Azospirillum* and *Rhizobium* have been found in grasses and legumes, respectively.

The inoculation with the *Azospirillum brasilense* bacterium increases grain yield without significantly increasing production costs, which, consequently, leads to an increase in the profitability of the final product (Vendruscolo et al., 2018). In studies of corn intercropped with *Urochloa decumbens* (Stapf) R.D.Webster (Syn. *Brachiaria decumbens* Stapf), there was a higher accumulation of dry mass and number of leaves when strains of *Azospirillum* were inoculated onto the forage (Guimarães et al., 2011); however, there are few known researches with *Urochloa ruziziensis* (R.Germ. & C.M.Evrard) Morrone & Zuloaga (Syn. Brachiaria ruziziensis R.Germ. & C.M.Evrard).

The objective of this work was to evaluate the agronomic performance of corn grown single and intercropped with *U. ruziziensis* in succession to soybean inoculated with *Bradyrhizobium japonicum*, as well as the inoculation and reinoculation of corn with *A. brasilense*, in two contrasting soils.

Materials and Methods

The experiment was carried out in the autumn-winter of 2019 and 2020 in a screened house without air conditioning and with a glass cover to allow ambient light and with sides coated with galvanized wire for free air circulation, located at Embrapa Agropecuária Oeste, in the municipality of Dourados, in the state of Mato Grosso do Sul, Brazil (22°16'33.1"S, 54°48'55.6"W, at an altitude of 430 m). The climate of the region is humid mesothermal, with hot summers and dry winters, according to Köppen-Geiger's classification. Temperatures were recorded at a climatological station located 500 m from the screened house (Figure 1).

The experimental design used was randomized complete blocks, in a 2x2x3 factorial arrangement, with five replicates. The first factor was composed of two contrasting soils classified according to Santos et al. (2018): Latossolo Vermelho distrófico, i.e., Rhodic Haplustox, with a sandy texture, with 726 g kg^{-1} sand and 223 g kg^{-1} clay; and Latossolo Vermelho eutrófico, i.e., Rhodic Eutrustox, with a clayey texture, with 160 g kg^{-1} sand and 723 g kg^{-1} clay. The second factor consisted of types of corn cultivation: single and intercropped with *U. ruziziensis*. The third factor were the different inoculations: no inoculation, both in soybean and corn; inoculation with *B. japonicum* onto soybean and *A. brasilense* onto corn; and reinoculation with *B. japonicum* + *A. brasilense* onto soybean and reinoculation with *A. brasilense* onto corn.

Before the implementation of the experiment, liming and fertility standardization were carried out according to the soil analysis, which showed: pH water 5.8, 18.12 mg dm^{-3} P, 0.97 $cmol_c$ dm^{-3} Ca^{2+}, 0.44 $cmol_c$ dm^{-3} Mg^{2+}, 0.11 $cmol_c$ dm^{-3} K^+, cation exchange capacity of 3.49 $cmol_c$ dm^{-3}, base saturation of 43%, and sum of bases of 1.52 $cmol_c$ dm^{-3} for Latossolo Vermelho eutrófico; and 5.8 pH water, 22.20 mg dm^{-3} P, 3.27 $cmol_c$ dm^{-3} Ca^{2+}, 1.55 $cmol_c$ dm^{-3} Mg^{2+}, 0.59 $cmol_c$ dm^{-3} K^+, cation exchange capacity of 11.5 $cmol_c$ dm^{-3}, base saturation of 48%, and sum of bases of 5.40 $cmol_c$ dm^{-3} for Latossolo Vermelho distrófico. P was extracted by Mehlich-1, and Ca^{2+} and Mg^{2+} were extracted by KCl 1.0 mol L^{-1}. At sowing, fertilization was performed with 3.5 g (equivalent to 200 kg ha^{-1}) of the N-P_2O_5-K_2O 4-18-18 formula.

For the experiment, polyvinyl chloride (PVC) pots with 60 cm height x 40 cm diameter, containing 60 kg air-dried soil, were used. On 3/26/2019 and 3/5/2020, four seed per pot of the AG8480 corn hybrid were sown in single and intercropped treatments, respectively. For intercropping, six seed of *U. ruziziensis* (Sementes Oeste Paulista, Presidente Prudente, SP, Brazil) were sown per pot. In the inoculation treatments, the AbV5

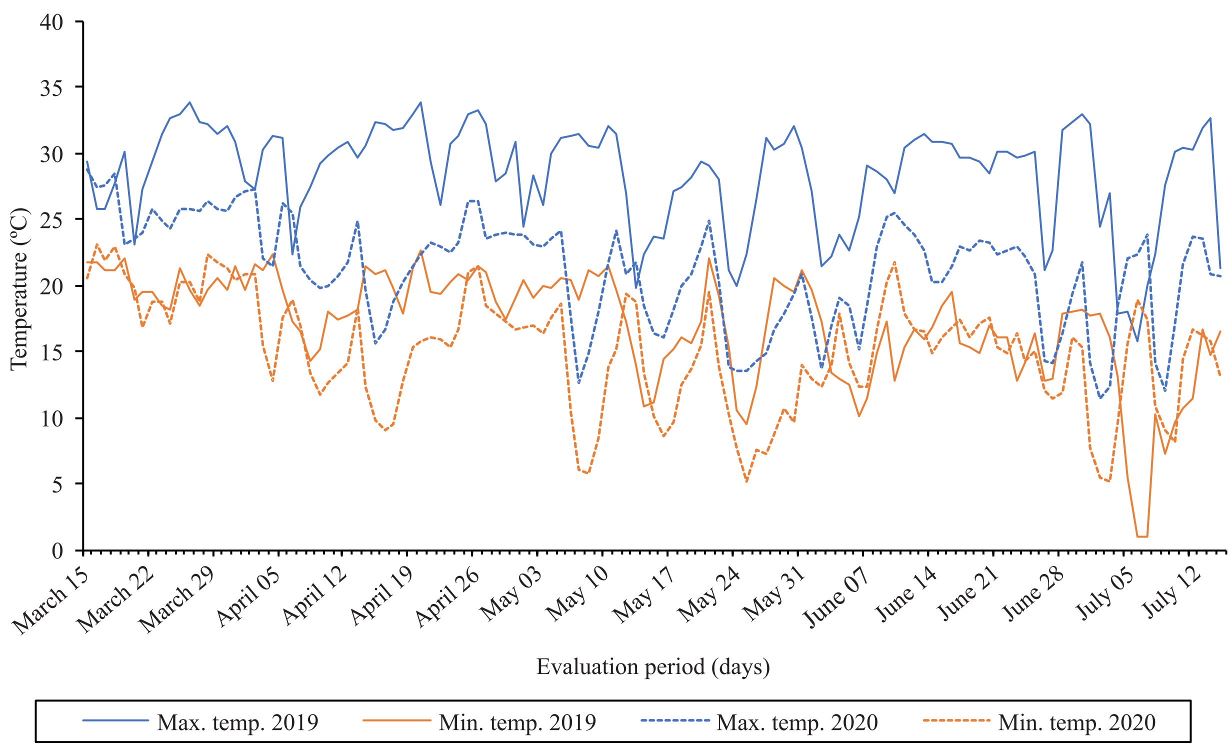

Figure 1. Maximum (Max. temp.) and minimum (Min. temp.) temperatures obtained at the experimental station of Embrapa Agropecuária Oeste, located 500 m from the screened greenhouse where the experiment was carried out in 2019 and 2020, in the municipality of Dourados, in the state of Mato Grosso do Sul, Brazil. Source: Embrapa Agropecuária Oeste (2020).

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and AbV6 *A. brasilense* strains, with 2x10^8 colony-forming units (CFU) per milliliter, were used. At ten days after emergence, two seedlings of corn and four of *U. ruziziensis* were left per pot, equivalent to the commercial population of the species.

In the summer of 2019 and 2020, soybean was sown in 72 pots after the desiccation of *U. ruziziensis* with glyphosate at a rate of 1.44 kg ha^{-1} a.i. Six seed were deposited per pot and inoculated with the Semia 5080 *B. japonicum* strain, with 6x10^1 CFU per gram, and with the AbV5 and AbV6 *A. brasilense* strains, with 2x10^8 CFU mL^{-1}, all obtained from commercial products registered at Ministério da Agricultura, Pecuária e Abastecimento (Brasil, 2011). After emergence, four soybean seedlings were left per pot.

Soybean seed in the summer and corn and *U. ruziziensis* seed in the autumn-winter were deposited in 3.0 cm deep furrows. The inoculants were applied directly on the seeds using a manual sprayer, at a rate equivalent to 150 mL ha^{-1} diluted in 100 L water as recommended by the manufacturer. Then, the seeds were covered with soil, which was slightly moistened to facilitate seed germination.

Soil moisture was kept close to 80% of field capacity by drip irrigation using the rainwater captured by the gutters of the screened house. Pest and disease control was carried out in the two crop years through the application of the deltamethrin insecticide, at a rate of 0.005 L ha^{-1}, at 10 and 30 days after corn emergence.

Harvests were performed in the pots on 7/17/2019 and 7/10/2020. The corn and *U. ruziziensis* cycles lasted 112 and 107 days in 2019 and 2020, respectively. For the two corn plants and the six *U. ruziziensis* plants left in each pot, the following characteristics were evaluated: number of grains per ear, corn grain yield in kg ha^{-1}, mass of one hundred grains, corn dry mass in kg ha^{-1}, and *U. ruziziensis* dry mass in kg ha^{-1}. Grains were corrected to 13% moisture, and, then, yield was calculated (Brasil, 2009). An analytical balance was used to weigh the material. The obtained dough was dried in an oven with forced-air circulation, at 60°C, for 72 hours.

The results were subjected to the analysis of variance for each evaluation factor in 2019 and 2020, and, when differences were detected by the F-test, at 5% probability, treatment means from each evaluation were compared by Tukey's test, also at 5% probability.

Results and Discussion

In the harvest carried out in 2019, the interactions were: significant between soil class and cropping systems for grain yield and number of grains per ear; significant between cropping systems and inoculation treatments for corn dry mass, grain yield, and number of grains per ear; and triple between soil classes, cropping systems, and inoculation for corn dry mass and grain yield (Table 1).

In the harvest in 2020, the interactions were: significant between soil classes and cropping systems for corn dry mass, grain yield, and number of grains per ear; significant between soil classes and inoculation for corn dry mass and *U. ruziziensis* dry mass; and not significant/not triple between soil classes, cropping systems, and inoculation for the analyzed variables (Table 1).

Corn dry mass yield in the treatment with *B. japonicum* inoculated onto soybean and with *A. brasilense* reinoculated onto corn was 14.8% higher than that of the treatment without inoculation. When corn was grown single in the clayey soil, 15.8% more dry mass was produced under no inoculation than with the inoculation of *B. japonicum* + *A. brasilense* onto soybean and with the reinoculation of *A. brasilense* onto corn (Table 2).

In 2019, 14.3% more dry mass was produced by corn grown in single cropping than in intercropping in the clayey soil without any inoculation. This gain in yield in the single cropping system is expected due to the lack of competition between plants in the same space for nutrient resources, water, and light. However, in the treatment with the reinoculation of *A. brasilense* in the clayey soil, intercropped corn produced 16.3% more dry mass than the single one.

When comparing the treatments with inoculation in both soil classes and cropping systems, the highest plant dry mass of 8,144 kg ha^{-1} was obtained by reinoculation in the clayey soil and the lowest one of 5,263 kg ha^{-1} in the sandy soil under intercropping and without any inoculation (Table 2). This shows the benefit of inoculation with *A. brasilense* to corn due to its synergistic effect on the mass yield of this cereal. Quadros et al. (2014) also observed increases in the dry mass of corn plants when inoculated with *A. brasilense*. According to Bashan et al. (2004), *Azospirillum* has the ability to excrete phytohormones, mainly indoleacetic acid, which have an essential role in promoting plant growth.

Regarding yield in 2019, the highest value of 5,744 kg ha^{-1} was found for corn grown in the clayey soil under intercropping and reinoculation with *A. brasilense*, whereas the lowest value of 3,440 kg ha^{-1} was verified in the treatment without any inoculation (Table 2). In intercropping in the sandy soil, reinoculation provided a 38.3% increase in grain yield compared with no inoculation. In both studied soils, the treatments with inoculation and reinoculation led to a significant increase in yield.

In the same year, both single and intercropped corn, inoculated in both soils, showed higher grain yields of 5,171 and 5,744 kg ha^{-1}, respectively, than the corn that did not receive any inoculation (Table 2). Lana et al. (2012) reported that inoculation with *Azospirillum* provided increases of up to 14.0% in corn grain yield, even without the addition of N. Plant growth-promoting bacteria, such as *A. brasilense*, can lead to consistent gains without the need for large investments in N fertilizers (Ferreira et al., 2020).

In the 2020 harvest, the highest grain yield was observed in the clayey soil in the single cropping system (Table 3). In this soil class, dry mass, grain yield, and the number of grains per ear were higher both in the 2019 and 2020 crop years. However, in 2019, the dry mass of *U. ruziziensis* and the mass of one hundred corn grains did not differ significantly between soils. Pandolfo et al. (2015) did not find differences in the mass of one thousand grains due to the inoculation with *A. brasilense* nor an increase in grain yield. According to the authors, the environment directly interferes with the agronomic performance of corn and the performance of bacteria.

The average yield of corn grains was 772 and 3,017 kg ha^{-1} higher in 2019 and 2020, respectively, in pots with the clayey soil (Table 4), which can be attributed to the fact that clay soils are more fertile than the sandy ones (Niranjana et al., 2018). Ronquim (2010) concluded that sandy-textured soils are more susceptible to erosive processes and nutrient leaching due to their greater pore space and lower cation exchange capacity, which contributes to a reduced crop yield.

Table 1. Summary of the analysis of variance for shoot dry mass (DM), grain yield (GY), number of grains per ear (NGE), and mass of one hundred grains (M100G) of corn (*Zea mays*) grown single and intercropped with *Urochloa ruziziensis*, as well as shoot dry mass of *U. ruziziensis* (DMU), as a function of inoculation and reinoculation in two contrasting soils – a clayey Latossolo Vermelho eutrófico and a sandy Latossolo Vermelho distrófico – in the 2019 and 2020 crop seasons in the municipality of Dourados, in the state of Mato Grosso do Sul, Brazil[(1)].

Source of variation	Character				
	DM	Grain yield	NGE	M100G	DMU
			2019 crop season		
Soil (S)	*	*	*	ns	ns
Cultivation (C)	ns	*	ns	*	-
Inoculation (I)	ns	*	ns	ns	ns
S x C	ns	*	*	ns	-
S x I	ns	ns	ns	ns	ns
C x I	*	*	*	ns	-
S x C x I	*	*	ns	ns	-
CV (%)	12.54	11.05	40.22	13.66	35.78
			2020 crop season		
Soil (S)	*	*	*	*	*
Cultivation (C)	*	*	*	*	-
Inoculation (I)	*	ns	ns	ns	ns
S x C	*	*	*	ns	-
S x I	*	ns	ns	ns	*
C x I	ns	ns	ns	ns	-
S x C x I	ns	ns	ns	ns	-
CV (%)	11.11	12.89	15.84	18.64	16.65

[(1)]CV, coefficient of variation; and -, character not evaluated in single corn. *Significant by the F-test, at 5% probability. [ns]Nonsignificant.

Table 2. Average value for dry mass of the aerial part and for grain yield of corn (*Zea mays*) grown single and intercropped with *Urochloa ruziziensis* as a function of inoculation and reinoculation with *Bradyrhizobium japonicum* and *Azospirillum brasilense* in two contrasting soils – a clayey Latossolo Vermelho eutrófico and a sandy Latossolo Vermelho distrófico – in the 2019 and 2020 crop seasons, in the municipality of Dourados, in the state of Mato Grosso do Sul, Brazil[(1)].

Treatment	Sandy		Clayey	
	Intercropping	Single	Intercropping	Single
	Dry mass of the aerial part of corn (kg ha^{-1}) – 2019 harvest			
No inoculation	6,938aBα	8,097aAα	5,263aAβ	5,680aAβ
B. japonicum + *A. brasilense*	8,144aAα	6,812bBα	5,519aAβ	6,020aAα
B. japonicum + *A. brasilense* + *A. brasilense*	6,931aAα	7,259abAα	5,596aAβ	5,486aAβ
Coefficient of variation (%)	12.54			
	Corn grain yield (kg ha^{-1}) – 2019 harvest			
No inoculation	3,440bBα	4,699aAα	2,432bBβ	4,757aAα
B. japonicum + *A. brasilense*	5,744aAα	4,698aBα	2,945bBβ	5,171aAα
B. japonicum + *A. brasilense* + *A. brasilense*	5,326aAα	5,057aAα	3,943aBβ	5,081aAα
Coefficient of variation (%)	11.05			
	Corn grain yield (kg ha^{-1}) – 2020 harvest			
No inoculation	6,371aBβ	9,229aAα	2,199aBβ	7,288aAα
B. japonicum + *A. brasilense*	6,668aBβ	9,293aAα	2,569aBβ	7,228aAα
B. japonicum + *A. brasilense* + *A. brasilense*	6,639aBβ	9,625aAα	3,167aBβ	7,272aAα
Coefficient of variation (%)	12.95			

[(1)]Means followed by equal letters – lowercase in the columns comparing inoculation in each cropping system and soil, uppercase in the lines comparing soils within each inoculation treatment and cropping system, and Greek in the lines comparing cropping system in each soil and inoculation – do not differ by Tukey's test, at 5% probability.

Table 3. Average value for dry mass (DM) of the aerial part, grain yield, number of grains per ear (NGE), and mass of one hundred grains (M100G) of corn (*Zea mays*), as well as dry mass of the aerial part of *Urochloa ruziziensis* (DMU), in two contrasting soils – a clayey Latossolo Vermelho eutrófico and a sandy Latossolo Vermelho distrófico – in single and intercropping systems in 2019 and 2020, in the municipality of Dourados, in the state of Mato Grosso do Sul, Brazil[(1)].

Soil	DM (kg ha^{-1})	Grain yield (kg ha^{-1})	NGE	M100G (g)	DMU (kg ha^{-1})
			2019 harvest		
Sandy	5,594b	4,055b	256b	19.9a	1,858a
Clayey	7,363a	4,827a	337a	20.2a	1,779a
Cultivation					
Single	6,559a	4,910a	298a	21.4a	-
Intercropping	6,398a	3,972b	295a	18.7b	-
Coefficient of variation (%)	12.99	13.16	40.22	13.66	35.78
Soil			2020 harvest		
Sandy	5,916b	4,954b	134b	22.4b	7,334a
Clayey	9,545a	7,971a	193a	26.2a	4,383b
Cultivation					
Single	8,454a	8,322a	201a	26.7a	-
Intercropping	7,007b	4,602b	126b	21.9b	-
Coefficient of variation (%)	14.17	12.95	15.84	18.64	16.28

[(1)]Means followed by equal letters, in the columns, do not differ by Tukey's test, at 5% probability.

The dry mass of *U. ruziziensis* was 4.2 and 42% higher in the clayey soil in 2019 and 2020, respectively (Table 4). Bezerra et al. (2020) observed improved characteristics of this species in clayey soil, which was favored, in part, by the greater nutrient availability in this soil. Forage plants produce large amounts of biomass in the dry season of the year, which can be used as pasture or straw to cover the ground, and the combination of corn and *Urochloa* spp. at the end of the rainy season allows drawing water from the deeper parts of the soil to promote photosynthesis during the dry season (Campanhola & Pandey, 2019).

Mass yield is an important tool for the recovery of degraded soils in tropical regions, as the process of straw decomposition is accelerated in these soils due to climatic conditions. Therefore, a higher straw production allows the soil to be covered for a longer period, providing a more favorable environment for plant development. In addition, *Urochloa* spp. improves the soil physical conditions for water infiltration and allows a greater exploitation of the soil profile, reducing the erosive process and also maintaining the production system (Chioderoli et al., 2012).

Overall, corn yield was greater under single cropping. In 2019, single corn showed an average yield 19.6% higher than that of intercropped corn, i.e., 4,910 vs. 3,972 kg ha^{-1}. In 2020, this difference was of 45.6%, with single corn producing 8,332 kg ha^{-1} and the intercropped one 4,602 kg ha^{-1} (Table 3).

The low grain yield in intercropped corn was already expected due to the greater population of *U. ruziziensis* plants and, therefore, competition between species, but this reduction is offset by the total dry mass of corn and forage when the aim is straw production. The low corn grain yield in 2019, which was of 4,910 kg ha^{-1} under cropping, may be associated with the climate, whose higher temperatures were unfavorable for corn cultivation (Figure 1). According to Renato et al. (2013), the increase in air temperature can lead to a reduction in the photosynthetic rate, an increase in the respiration and transpiration rates, and a reduction in the crop cycle, causing a drop in grain yield even in crops with C_4 metabolic behavior.

The interaction between soil classes and cropping systems (Table 4) showed a lower grain yield of 3,107 kg ha^{-1} and a reduced number of grains per ear of 213 in 2019 when corn was intercropped on sandy soil. Regarding the interaction of grain yield as a function of soil within each cropping treatment, single corn produced 25% more than the intercropped one.

For grain yield within each inoculation, single corn produced 19% more when subjected to reinoculation than to no inoculation. Under intercropping, corn produced 30% more than when not inoculated. Contrarily, Mumbach et al. (2017) did not observe significant responses to the variables analyzed as a function of the inoculation with *A. brasilense*. However, Fukami et al. (2016) concluded that the lower N rates

Table 4. Average value for dry mass of the aerial part, grain yield, and number of grains per ear of corn (*Zea mays*) grown single and intercropped with *Urochloa ruziziensis* in two contrasting soils – a clayey Latossolo Vermelho eutrófico and a sandy Latossolo Vermelho distrófico – in the 2019 and 2020 crop seasons, in the municipality of Dourados, in the state of Mato Grosso do Sul, Brazil[(1)].

Cultivation	Dry mass of the aerial part of corn (kg ha^{-1})		Corn grain yield (kg ha^{-1})		Number of grains per ear	
	Sandy	Clayey	Sandy	Clayey	Sandy	Clayey
			2019 harvest			
Intercropping	-	-	3,107bB	4,837aA	213bB	376aA
Single	-	-	4,936aA	4,951aA	298aA	297aA
CV (%)			11.94		40.22	
			2020 harvest			
Intercropping	4,768bB	7,064bA	2,645bB	7,263bA	85bB	166aA
Single	9,245aA	9,844aA	6,559aB	9,382aA	183aB	219aA
CV (%)	11.11		12.95		15.84	

[(1)]Means followed by equal letters, lowercase in the columns comparing the soil within each cropping system and uppercase in the rows comparing cropping system within each soil class, do not differ by Tukey's test, at 5% probability. CV, coefficient of variation.

associated with the inoculation with *A. brasilense* result in yields similar to those obtained with total N fertilization at topdressing, making inoculation with *A. brasilense* an important technique in the search for more profitable agricultural systems with a rational use of inputs.

In 2020, corn dry mass was higher (7,064 kg ha^{-1}) in the clayey soil and with intercropping with *U. ruziziensis*. Ngwira et al. (2012) found that intercropped corn produced more than twice as much plant biomass in drier years and 33% more grains than single corn. In the present study, dry mass yield in the sandy soil was 48% higher in single corn. Grain yields were also higher in corn grown single in the sandy and clayey soils, with values of 6,559 and 9,382 kg ha^{-1}, respectively. However, considering the soils factor, both intercropped and single corn had higher yields of 7,263 and 9,382 kg ha^{-1}, respectively, in the clayey soil (Table 4). The lower values found for intercropped corn may be associated with its competition with *U. ruziziensis* for water, light, and nutrients.

In the evaluation of the inoculation treatments and cropping systems in 2019, the dry mass of corn (6,888 kg ha^{-1}) was higher in the single cropping system without inoculation, with no differences in the treatments with inoculation (Table 5). In the same year, intercropped and reinoculated corn showed higher averages than noninoculated corn, with a dry mass of 6,263 kg ha^{-1}, grain yield of 4,634 kg ha^{-1}, and number of grains per ear of 395. Growth-promoting bacteria, such as *A. brasilense* and *B. japonicum*, may have promoted an increase in the synthesis of hormones, such as auxin, which stimulates plant growth. According to Taiz et al. (2017), the production of hormones, including auxins, is essential for plant growth and development.

Corn dry mass did not differ significantly in the sandy soil due to inoculations, but was higher in the clayey soil under cropping with inoculation and reinoculation in 2020 (Table 5). When evaluating mass as a function of inoculation within each soil, the highest yields were observed in the clayey soil, with an average of 10,211 kg ha^{-1} with reinoculation. Reinoculated corn was beneficial for *U. ruziziensis* mass yield in the sandy soil. Oliveira et al. (2007) observed that 'Marandu' *Urochloa brizantha* (A.Rich.) R.D.Webster inoculated with diazotrophic bacteria showed a higher forage yield than when not inoculated.

Table 5. Average value for dry mass of the aerial part, grain yield, and number of grains per ear of corn (*Zea mays*), as well as dry mass of the aerial part of *Urochloa ruziziensis*, in single cropping and intercropping with *Urochloa ruziziensis*, with and without inoculation with *Bradyrhizobium japonicum* (BJ) and *Azospirillum brasilense* (AB), in two contrasting soils – a clayey Latossolo Vermelho eutrófico and a sandy Latossolo Vermelho distrófico – in the 2019 and 2020 crop seasons, in the municipality of Dourados, in the state of Mato Grosso do Sul, Brazil(1).

Treatment	Dry mass of the aerial part of corn (kg ha^{-1})		Corn grain yield (kg ha^{-1})		Number of grains per ear	
	Single	Intercropping	Single	Intercropping	Single	Intercropping
			2019 harvest			
No inoculation	6,888aA	6,101aB	4,728aA	2,936bB	324aA	188bB
BJ + AB	6,416aA	6,831aA	4,934aA	4,344aB	283aA	301abA
BJ, AB + AB	6,372aA	6,263aA	5,069aA	4,634aA	286aB	395aA
CV (%)	16.26		11.94		40.22	
Treatment	Dry mass of the aerial part of corn (kg ha^{-1})		Dry mass of the aerial part of *U. ruziziensis* (kg ha^{-1})			
	Sandy	Clayey	Sandy	Clayey		
			2020 harvest			
No inoculation	5,879aB	8,776bA	7,858aA	4,078aB	-	-
BJ + AB	5,562aB	10,211aA	7,401aA	3,765aB	-	-
BJ, AB + AB	6,306aB	9,647abA	6,743aA	5,305aB	-	-
CV (%)	14.17		16.65		-	

(1)Means followed by equal letters, lowercase in the columns comparing the split of cropping system or soil within each inoculation and uppercase in the rows comparing the split of inoculation within each soil or cropping system, do not differ by Tukey's test, at 5% probability. CV, coefficient of variation.

Bacteria inoculated onto corn seeds were beneficial for the growth of *U. ruziziensis* under intercropping in the sandy soil. Guimarães et al. (2011) verified that the dry mass of *U. decumbens* inoculated with diazotrophic bacteria was close to that of *U. ruziziensis* fertilized with N, showing that bacteria play an important role in the growth of this forage species.

Conclusions

1. Single corn (*Zea mays*) has a higher grain yield when reinoculated with *Azospirillum brasilense* and grown in a clayey soil – Latossolo Vermelho eutrófico – in both crop years.
2. A higher grain yield is obtained with the annual reinoculation of *A. brasilense* onto corn intercropped with *Urochloa ruziziensis* in the clayey soil.
3. The inoculation and reinoculation with *A. brasilense* reduces the corn yield losses caused by competition with *U. ruziziensis*.

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