

## Organic and Mineral Source of Nitrogen Associated With *Azospirillum brasilense* in Culture of Wheat

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### Abstract

Nitrogen fertilization in wheat is the item that most impacts production costs when it comes to fertilization, due to its importance and demand for this crop. Thus, organic fertilization and nitrogen fixing bacteria can be management strategies to supply the nitrogen demand. The objective of this study was to evaluate the impacts of the bacterium *A. brasilense* associated with organic fertilization and mineral fertilization on some plant parameters and on the final productivity of wheat grains. The experimental design was a randomized block, with 8 treatments and 8 repetitions, with the following treatments: T1: Control; T2: *A. brasilense*; T3: 100% N-mineral; T4: 100% N-mineral + *A. brasilense*; T5: 50% N-mineral + 50% N-organic; T6: 50% N-mineral + 50% N-organic + *A. brasilense*; T7: 100% N-organic; T8: 100% N-organic + *A. brasilense*. The parameters of the plant and the final productivity of wheat grains showed that the use of the bacterium *A. brasilense* was not an efficient strategy, however, in relation to the fertilization sources in the final grain productivity, the use of urea isolated or associated with organic fertilization did not differ from each other and were superior to the other treatments.

**Keywords:** mineral nutrition, nitrogen fixation, organic fertilization, *Triticum aestivum*

### 1. Introduction

In the 2019 harvest, Brazil produced 5,200 million tons of wheat (*Triticum aestivum*) for an internal demand of 12,000 million tons. Among the producing regions, the South stands out with 88% of national production, with the Paraná, Rio Grande do Sul and Santa Catarina states responsible for 42.1; 42.5 and 3.4%, respectively of that amount (CONAB, 2019). This lack of production to meet domestic demand is mainly due to the cost of production, devaluation of the product and the lack of proper incentive by government agencies. Regarding fertilization, Nitrogen (N) is the nutrient with the greatest demand in the crop.

The application of a mineral source is the most common way to meet the demand for N in the wheat crop, due to its practicality, high solubility and rapid availability to the plants, in addition to the moment of application factor that may be in greater synchronism with the demand of the culture.

Based on this, new, more sustainable alternatives are gaining ground in production systems, among which the use of organic residues from animal production stands out, and more recently, studies such as the use of nitrogen-fixing and growth-promoting bacteria with an emphasis on of the *Azospirillum* genus (Milléo & Cristófoli, 2016), as a way to minimize the cost of production and the impact on the environment seeking a more sustainable agriculture (Sá et al., 2017). *Azospirillum brasilense* produce various stimuli in plants as the biological fixation of N, besides production of developmental hormones such as cytokinins, gibberellins and auxins, promoting greater root growth and consequently increasing the water and nutrient absorption area (Kazi et al., 2016).

As it is a recent technology, there is still little and sometimes contradictory information in the literature regarding the use of bacteria of the genus *Azospirillum* in seed treatment. This lack of response in a more concise manner regarding the use of bacteria of the genus *Azospirillum*, may be associated with biotic and abiotic factors,

however studies regarding the use of this inoculant via seed associated with an organic source are lacking. Therefore, the objective of the study was to evaluate sources of nitrogen associated with the use of *A. brasilense*, on some plant parameters and on the final productivity of wheat grains.

## 2. Methodology

The experiment was carried out in 2018, in the city of Frederico Westphalen, located in the Middle Upper Uruguay region at an altitude of 483 m. The region's characteristic climate is humid subtropical, Cfa, average annual temperature of 19°C and average annual rainfall of 1,880 mm approximately according to Köeppen. The soil in the experimental area is characterized as a typical dystrophic Red Latosol, with a clay texture (EMBRAPA, 2013), which at the time of the experiment had the following physical and chemical characteristics.

Table 1. Chemical and physical characterization of the experimental area in the 0-20 cm layer. Frederico Westphalen, 2018

pH	O.M	Clay	Sand	Silt	P-meh	K <sup>+</sup>	Ca <sup>2+</sup>	Mg <sup>2+</sup>	Al <sup>3+</sup>	CTC	V
water	dag Kg <sup>-1</sup>	----- mg dm <sup>-3</sup> -----			----- cmol <sub>c</sub> dm <sup>-3</sup> -----			----- (%) -----			
5.9	2.7	710	199	96	4.8	69.5	6.6	2.9	0.0	14.1	68.7

Note. OBS.: pH in water; P-meh and K<sup>+</sup> mehlich<sup>1</sup> extractor; Ca<sup>2+</sup>, Mg<sup>2+</sup> and Al<sup>3+</sup> extractor KCl 1 mol L<sup>-1</sup>; OM organic matter determined by the Walkley-Black method; CTC cation exchange capacity; V base CTC saturation.

The accumulation of rain during the conduction of the experiment was around 700 millimeters, well distributed throughout the crop cycle and meeting the demand of the same. After sowing and applying the treatments, a 12 mm irrigation was carried out to dissolve the mineral N applied over the sowing furrow and to have enough moisture to guarantee good germination and seedling emergence. Temperatures were around 20 °C, ideal for a good development of the culture. Figure 1 shows precipitation and temperature data during the conduct of the experiment.

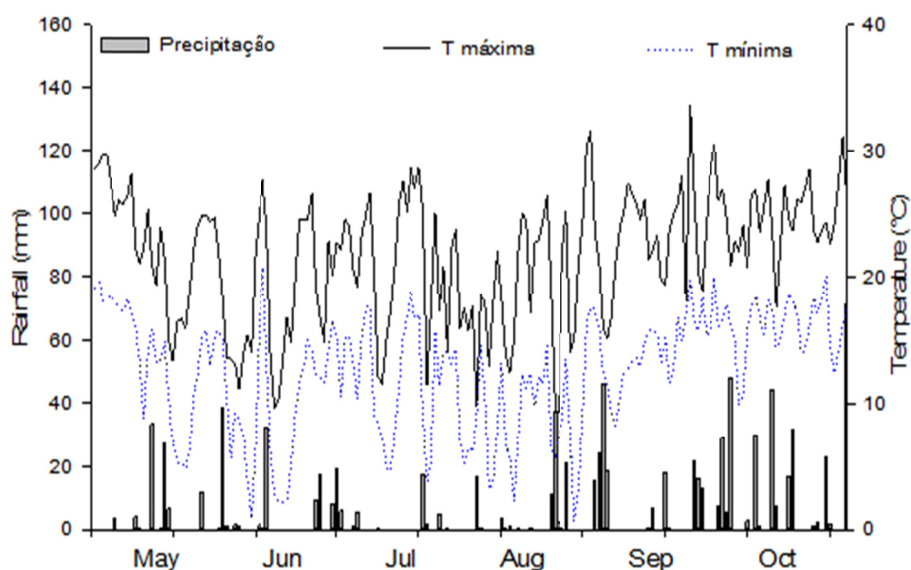


Figure 1. Average daily temperatures and daily precipitation during the months of the experiment Frederico Westphalen (RS), 2018

The experimental design was a randomized blocks (CRBD) with 8 treatments and 8 repetitions, and the treatments consisted of associations of sources of mineral nitrogen (urea 45%) and organic nitrogen (poultry litter 2.7% of N) associated or not with *A. brasilense*: T1 = control without application of any nitrogen source; T2 = *A. brasilense*; T3 = 100% N-mineral; T4 = 100% N-mineral + *A. brasilense*; T5 = 50% N-mineral + 50% N-organic; T6 = 50% mineral N + 50% organic N + *A. brasilense*; T7 = 100% N-organic; T8 = 100% N-organic

+ *A. brasilense*. The amount of N applied to the crop was 60 kg of N ha<sup>-1</sup>, based on the expected yield of 4000 kg ha<sup>-1</sup>, according to the Liming and Fertilization Manual for the states of Rio Grande do Sul and Santa Catarina (CQFS-RS/SC, 2016), however for organic fertilization 100% of mineralized N was considered during cultivation.

The experimental area had a 5-year history of no-tillage, with straw from the predecessor crop soybeans (*Glycine max*) and ryegrass (*Lolium multiflorum*) at an early stage of development, thus drying with glyphosate (Roundup®) 3.5 L ha<sup>-1</sup> was done. 22 days after drying wheat was sown - cultivar TBIO TORUK with density of 3.4 million seeds ha<sup>-1</sup> on 26/05/2018. The wheat seeds were inoculated 30 minutes before sowing with *Azospillum brasilense* (Azototal®), which has strains AbV5 and AbV6 and a concentration of 2.0 × 10<sup>8</sup> colony-forming unit—CFU, ml<sup>-1</sup> developed by Embrapa and Total Biotecnologia, applying 2.5 ml kg<sup>-1</sup> of wheat seeds. For this operation, a winter seeder with 16 rows spaced with 17 cm between them was used, with the plots being 2.72 meters wide and 6 meters long. The seeds were treated with the insecticide Vitavax Thiram 200 SC 3 ml kg<sup>-1</sup> of seed and fungicide Much 600 FS 1 ml kg<sup>-1</sup> of seed one day before sowing.

First, the sowing of the non-inoculated seeds was carried out and then the inoculated seeds. In this same operation, 110 kg ha<sup>-1</sup> of P<sub>2</sub>O<sub>5</sub> (triple superphosphate) was applied to the sowing furrow and after that operation, on the same day, 70 kg ha<sup>-1</sup> of K<sub>2</sub>O were applied in haul to all plots. The day after the sowing, 20 kg ha<sup>-1</sup> of mineral N was applied over the sowing furrow in the treatments with mineral N and the whole poultry litter 1.111 and 2.222 kg ha<sup>-1</sup> poultry beds for 50% and 100% of N, respectively in treatments that involved the application of this source in an isolated and/or associated way. Afterwards, irrigation was carried out in the evening to minimize possible nitrogen losses through volatilization. The rest of the mineral N 40 Kg ha<sup>-1</sup> was equally divided in the treatments that involved the use of this source in the phenological stages of wheat tillering and elongation.

For crop management, when it came to plant protection practices via chemical control of pests, diseases and weeds, these were carried out with the help of a backpack sprayer adapted with a 4-nozzle bar and a capacity of 20 L, following the recommendation according to the technical indications for the wheat crop, without interfering in the results (RCBPTT, 2018).

As for the plant characteristics, the following evaluations were made in each experimental unit: Dry mass of plants of the aerial part (DM): the plants were collected close to the ground with a table of known area of 0.25 m<sup>2</sup> in R1 stage, taking them to an oven at 60 °C until a constant mass is obtained, weighed and extrapolated to Kg ha<sup>-1</sup>. Total Nitrogen of the aerial part of the plant (TN): it was determined from a coarse grind of the dry mass and later a finer grind was obtained in a Wiley mill from a small part, a homogeneous portion was taken for the determination according to Tedesco et al. (1995), the results being expressed in g Kg<sup>-1</sup>. Total chlorophyll content (TCC) determined in 5 random plants in the flag leaf in stage R1, with the aid of the portable chlorophyll meter CLOROFILOG®, the data being expressed in an FCI dimensionless index (Falker chlorophyll index).

Other evaluations were carried out after the physiological maturation point of the wheat, where it was determined: Number of Ears per Square Meter (NESM): ears of 2 m linear and extrapolated to m<sup>2</sup> were counted, resulting in units. The main tiller was cut into 10 plants, flush to the ground randomly to carry out the following evaluations: Plant height (PH): it was measured from the base to the peak of the ear, discounting the edges, measured with a graduated ruler. Ear length (EL): it was measured from the base of the ear to the apex of the ear discounting the edges, measured with a graduated ruler. Number of spikelets per ear (NSE): the number of fertile spikelets on the ear is counted. Number of grains per ear (NGE): thresh the ear manually to count all grains, resulting in units.

For the thousand grains mass (TGM): this variable was determined through eight subsamples of one hundred grains from each plot that determined the grain yield, where the average and adjustment for the thousand grains mass at 13% humidity was obtained, in grams. Hectoliter weight (HW): was determined using a hectoliter weight scale, results in kg hl<sup>-1</sup>. Grains Final Productivity (GFP): it was determined by harvesting all the plants in the useful area of the experimental unit (4.34 m<sup>2</sup>), which were tracked with a tractor mixer. Subsequently, the samples were cleaned and the weight corrected to 13% humidity and later the productivity data was extrapolated in Kg ha<sup>-1</sup>.

The results were subjected to analysis of variance and when the variables showed significance, the means were grouped by the Scott-Knott test at p > 0.05 probability. The analysis were performed with the aid of the statistical software SISVAR (Ferreira, 2011).

### 3. Results and Discussion

In Table 2, the analysis of variance revealed a significant difference at 5% probability of error, using the F test, for the variables PH, EL, NSE, NGE, NESM, HW, TCC, TN, DM and GFP, while on the other hand, the TGM variable did not present a significant difference between all treatments studied, not even for the control without nitrogen fertilization. This lack of response may be linked to the material's genetic load.

Table 2. Summary of analysis of variance with information regarding the characteristics: plant height (PH), ear length (EL), number of spikelets per ear (NSE), number of grains per ear (NGE), number of ears per meter square (NESM), thousand grain mass (TGM), hectoliter weight (HW), total chlorophyll content (TCC), total nitrogen of the aerial part of the plant (TN), dry mass of the aerial part of the plant (DM) and final grain productivity (GFP) obtained from wheat as a result of organic and mineral nitrogen fertilization associated with *A. brasilense*, Frederico Westphalen, 2018 harvest

VF <sup>1</sup>	DF	MS					
		PH	EL	NSE	NGE	NESM	TGM
Blocks	7	25.77*	0.21 <sup>ns</sup>	0.44 <sup>ns</sup>	17.59*	14.683.0*	2.93 <sup>ns</sup>
Treatments	7	25.22*	0.55*	0.90*	30.35*	6.026.5*	3.90 <sup>ns</sup>
Error	49	8.99	0.16	0.37	7.03	1.489.6	2.11
Total	63						
CV-%		4.23	6.60	4.60	9.44	7.90	4.40

VF <sup>1</sup>	DF	MS				
		HW	TCC	TN	DM	GFP
Blocks	7	2.63*	11.38*	4.79 <sup>ns</sup>	1.533.677.23*	739.127.40*
Treatments	7	6.51*	43.32*	57.53*	1.984.727.17*	2.697.223.23*
Error	49	0.91	4.04	3.22	366.941.74	107.710.15
Total	63					
CV-%		1.21	4.42	8.88	10.23	9.22

Note. VF<sup>1</sup>: Variation Factor, DF: degrees of freedom; CV-%: coefficient of variation; MS: mean square of the variables; \* Significant values for F test at 5% probability of error; ns: non-significant value.

Table 3 shows that there was a significant difference between treatments. Regarding treatments without the addition of any nitrogen source (Control and *A. brasilense*), all treatments with addition of nitrogen regardless of the source, were statistically superior for the variables PH, EL and NSE. In the treatments using 100% N-organic and 50% N-mineral + 50% N-organic, there was no significant difference when compared to the 100% N-mineral treatment, showing that for these variables, organic fertilization (poultry litter) can be a viable alternative as a source of nitrogen. However, the use of *A. brasilense* in isolation or associated with some source of nitrogen (mineral and/or organic), did not present a significant response to the variables studied.

Table 3. Effect of mineral and organic nitrogen sources associated to the *A. brasilense* about the plant height (PH), ear length (EL) and number of spikelets/ear (NSE), in the wheat. Frederico Westphalen (RS), in the year of 2018

Tratamentos	PH (cm)	EL (cm)	NSE (n°)
Control	67.86 b	5.61 b	12.66 b
<i>A. brasilense</i>	68.20 b	5.73 b	12.75 b
100%N-mineral	71.69 a	6.42 a	13.54 a
100%N-mineral + <i>A. brasilense</i>	72.58 a	6.26 a	13.51 a
50%N-mineral + 50%N-organic	71.62 a	6.11 a	13.45 a
50%N-mineral + 50%N-organic + <i>A. brasilense</i>	71.59 a	6.11 a	13.32 a
100%N-organic	71.63 a	6.06 a	13.22 a
100%N-organic + <i>A. brasilense</i>	71.66 a	6.04 a	13.31 a
CV-%	4.23	6.60	4.60

Note. Averages followed by distinct letter in the columns differ from each other through the Scott Knott Test of 5% of probability. CV-%: Coefficient of Variation. N: Nitrogen.

For the PH, its growth occurred due to the use of nitrogen on the early stages of development, because it rises the production of phytohormones of growing promotion as the auxins and therefore, increasing the cell expansion and division (Alves et al., 2017), the same behavior was observed for the EL, due to the using of the nitrogen fertilization. As for the NSE, according to Demari et al. (2016), the management of the N influences the NSE, besides that often the soil conditions that provide a propitious environment for the plants, make any response impossible due to the implemented management practices (Munareto et al., 2019).

For the variables NGE, NESM and HW presented in the Table 4, the same behavior is observed for all treatments as observed for Table 3, that regardless of the source and management used, all of the treatments with addition of N were superior to the control and to the treatment only with seed inoculation with *A. brasilense*.

Table 4. Effect of mineral and organic nitrogen sources associated to the *A. brasilense* about the number of grains per ear (NGE), number of ears per square meter (NESM) and hectoliter weight (HW), in the wheat. Frederico Westphalen (RS), in the year of 2018

Tratamentos	NGE (n°)	NESM (n°) (cm)	HW
Control	25.83 b	442.92 b	76.73 b
<i>A. brasilense</i>	24.82 b	451.03 b	77.24 b
100%N-mineral	30.35 a	511.74 a	78.95 a
100%N-mineral + <i>A. brasilense</i>	30.47 a	513.28 a	79.38 a
50%N-mineral + 50%N-organic	28.54 a	505.93 a	78.89 a
50%N-mineral + 50%N-organic + <i>A. brasilense</i>	28.57 a	506.68 a	78.80 a
100%N-organic	28.16 a	490.74 a	78.67 a
100%N-organic + <i>A. brasilense</i>	28.35 a	486.32 a	78.76 a
CV-%	1.96	4.40	3.89

Note. Averages followed by distinct letter in the columns differ from each other through the Scott Knott Test of 5% of probability. CV-%: Coefficient of Variation. N: Nitrogen.

The variables NGE and EL showed a strong correlation, since the greater the length of the ear, the greater the number of grains per ear and also the better nutritional conditions of the plant favor at the time of the floral differentiation (Alves et al., 2017). As for the NESM and according to Hossain et al. (2013), high temperatures influence this variable in the early stages of the culture, reducing the number of tillers. However, this condition may be associated with the plasticity of wheat genotypes as well as a greater nutritional condition provided by the addition of N, increasing the fertility of the tillers.

The HW is important for the commercialization of wheat, as it implies in the quality and the lower the HW (less than 78 kg hl<sup>-1</sup>) the lower the price paid for the kilogram of the product. In this study, a significant difference was observed only in relation to the use of some nitrogen source. This may be associated with the availability factor of N in the soil, since this element is responsible for the production and maintenance of the leaf area and consequently, increased the photosynthetic capacity of the plant increasing the drain of photoassimilates for the grain. The HW also experiences variations due to the genotype-environment interaction (Franceschi et al., 2009), high levels of precipitation at the end of the reproductive period or even the lodging of the crop leaving the ears close to the ground receiving constant moisture, activate enzymatic processes in the seed (Munareto et al., 2019). According to Franceschi et al. (2009), when the enzymes are active, they make changes in the proteins and starch of the seeds, initiating the germination process, thus reducing its quality.

In the Table 5, one can observe that there was a significant difference between the treatments following the same tendency observed in the Tables 3 and 4. Regarding the treatments without adding any nitrogen source (Control and *A. brasilense*), all of the treatments with adding of nitrogen regardless of the source were statistically superior for the variables TCC and TN. However, the utilization of *Azospirillum* alone or associated with some source of nitrogen in the organic or mineral form, did not show a significant response in relation to all of the treatments studied.

Table 5. Effect of mineral and organic nitrogen sources associated to the *A. brasilense* about the total chlorophyll content (TCC) total nitrogen of the aerial part of plant (TN), in the wheat. Frederico Westphalen (RS), in the year of 2018

Tratamentos	TCC (FCI)	TN (g Kg <sup>-1</sup> )
Control	41.86 b	15.59 b
<i>A. brasilense</i>	42.00 b	16.44 b
100%N-mineral	47.41 a	21.59 a
100%N-mineral + <i>A. brasilense</i>	48.06 a	23.15 a
50%N-mineral + 50%N-organic	46.46 a	21.55 a
50%N-mineral + 50%N-organic + <i>A. brasilense</i>	46.52 a	21.26 a
100%N-organic	45.80 a	20.94 a
100%N-organic + <i>A. brasilense</i>	45.87 a	21.09 a
CV-%	4.42	8.88

Note. Averages followed by distinct letter in the columns differ from each other through the Scott Knott Test of 5% of probability. CV-%: Coefficient of Variation. N: Nitrogen.

The TCC observed in this work is attributed to the use of nitrogen as this characteristic occurs due to the increase in the chlorophyll concentration promoted by the greater availability of nitrogen by the plant (Galindo et al., 2017). Thus, the significant correlation is confirmed when observing the TN of wheat in this study with the TCC obtained by Clorofilog. Although the present study has not shown a significant response, numerically, the best treatment was 100% N-mineral + *A. brasilense*. According to Ali et al. (2002), the capture of atmospheric nitrogen for the plants by the N biological fixation can improve root development promoted by substances produced by rhizobacteria and, consequently, improve the nutrient absorption capacity of the wheat plants.

In the Table 6, the DM variable followed the same tendency of the Tables 3, 4 and 5. All of the treatments with addition of some N source did not differ from each other and were statistically superior to the treatments without addition of nitrogen (control and *A. brasilense*) and also the utilization of *Azospirillum* alone or associated with some source of nitrogen in the organic or mineral form, did not show a significant response in relation to all of the studied treatments. As observed regarding the utilization of nitrogen fertilization in the present study and as stated by Malavolta (2006), the increase in N availability reflects on a greater production of dry matter due to the nitrogen as a constituent of nucleic acids and proteins that form the plant structure.

Table 6. Effect of mineral and organic nitrogen sources associated to the *A. brasilense* about the dry mass of the aerial part of plant (DM) and grains final productivity (GFP), in the wheat. Frederico Westphalen (RS), in the year of 2018

Tratamentos	DM (kg ha <sup>-1</sup> )	GFP (kg ha <sup>-1</sup> )
Control	4915.13 b	2656.47 c
<i>A. brasilense</i>	5160.15 b	2668.27 c
100%N-mineral	6220.12 a	3994.58 a
100%N-mineral + <i>A. brasilense</i>	6405.27 a	4067.37 a
50%N-mineral + 50%N-organic	6320.76 a	3908.82 a
50%N-mineral + 50%N-organic + <i>A. brasilense</i>	6205.94 a	3973.89 a
100%N-organic	6051.36 a	3648.92 b
100%N-organic + <i>A. brasilense</i>	5847.29 a	3539.85 b
CV-%	10.23	9.22

Note. Averages followed by distinct letter in the columns differ from each other through the Scott Knott Test of 5% of probability. CV-%: Coefficient of Variation. N: Nitrogen.

For the GFP, it is observed that all treatments with application of N regardless of the source were superior to the control and to the treatment with only *A. brasilense*. When comparing nitrogen sources, it is clear that the highest yields were obtained when part or all of the N was applied in the mineral form, these yields being higher than those obtained in treatments with full application of the N via organic fertilization. On average, in the treatments

with application of part or all of the N via mineral source, the increase in the final grain yield was 10.9% higher than that observed for the average in the treatments, where only organic fertilization was used.

This lack of a more significant response in wheat yield with the utilization of the organic fertilizer as a source of nitrogen may be associated with the lack of synchronism between the demand for the culture and the availability of mineral nitrogen in the soil solution due to the question of mineralization and release of the N. For the highest grain yield (4.067 kg ha<sup>-1</sup>) in the treatment (100% N-mineral + *A. brasilense*), there was an increase of 53.1% compared to the control (without N), which shows the importance of nitrogen for high wheat yields.

When comparing the treatments 100% N-mineral + *A. brasilense* with 100% N-organic + *A. brasilense*, we observed a 13% reduction in the wheat yield in the treatment with the organic source associated with *A. brasilense*. This reduction in the yield when using organic fertilizer plus *A. brasilense*, may be associated with the competition of the bacteria in the soil microbial community, as the *A. brasilense* is a rhizospheric bacterium and consequently, like the native bacteria, are more adapted (Nunes et al., 2015) and (Munareto et al., 2019). Besides, the addition of a carbon source activates the soil biota, favoring those more adapted to the environment, in this case the native ones. On the second hand, other factors can negatively contribute such as inoculation rate, soil physical-chemical characteristics, pesticides and also of the plant genotypes themselves (Ludwig et al., 2018). Perhaps the wheat cultivar used in this study (TBIO TORUK) was not efficient in this interaction with these bacteria since they, as growth promoters, depend on the production of plant hormones that are synthesized and stimulate root growth by increasing the water and nutrients absorption capacity, which reflects on a greater resistance to water and salt stress (Hungria et al., 2010). Therefore, the lack of exposure of the culture to these stresses may be associated with the lack of a more significant response to the seeds inoculation with *A. brasilense*.

#### 4. Conclusion

The utilization of the *Azospirillum* did not prove to be an efficient practice, this season, for the analyzed plant variables and in the wheat grains final productivity and when a source of N was used, a significant response was observed in comparison to the treatments without N in most of the variables analyzed. It is suggested to conduct similar experiments over different seasons to assume the real efficiency of *A. brasilense*. In the comparison between the sources of N and for the grains final productivity, all of the treatments with the utilization of mineral N applied alone or associated with organic fertilization were significantly superior to the other treatments. This allows us to conclude that the organic fertilization must always be associated with mineral fertilization and not applied alone.

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#### References

- Ali, N. A., Darwish, S. D., & Mansour, S. M. (2002) Effect of *Azotobacter chroococcum* and *Azospirillum brasilense* inoculation an anhydrous ammonia on root colonization, plant growth and yield of wheat plant under saline alkaline cognition. *Journal of Agricultural Science Mansoura Univ.*, *Egypt*, *27*(8), 5575-5591.
- Alves, C. J., Arf, O., Ramos, A. F., Galindo, F. S., Nogueira, L. M., & Rodrigues, R. A. F. (2017). Irrigated wheat subjected to inoculation with *Azospirillum brasilense* and nitrogen doses as top-dressing. *Revista Brasileira de Engenharia Agrícola e Ambiental*, *21*(8), 537-542. <https://doi.org/10.1590/1807-1929/agriambi.v21n8p537-542>
- CONAB (Companhia Nacional de Abastecimento). (2019). *Acompanhamento da safra brasileira: Grãos, V. 7—Safra 2019/20—N.3—Terceiro levantamento*. Brasília: CONAB.
- CQFS-RS/SC (Comissão de Química e Fertilidade do Solo-RS/SC). (2016). *Manual de adubação e de calagem para os estados do Rio Grande do Sul e de Santa Catarina*. Porto Alegre: CQFS-RS/SC.
- Demari, G. H., Carvalho, I. R., Nardino, M., Follmann, D. N., Souza, V. Q. de; Somavilla, L., & Basso, C. J. (2016). Cama de aves como alternativa para adubação nitrogenada em trigo. *Revista Cultivando o Saber*, *9*(2), 224-242. Retrieved from [https://www.fag.edu.br/upload/revista/cultivando\\_o\\_saber/57bb33c3ea1bd.pdf](https://www.fag.edu.br/upload/revista/cultivando_o_saber/57bb33c3ea1bd.pdf)

- EMBRAPA (Empresa Brasileira de Pesquisa Agropecuária). (2013). *Sistema brasileiro de classificação de solos*. Brasília: EMBRAPA.
- Ferreira, D. F. (2011). Sisvar: a computer statistical analysis system. *Ciência e Agrotecnologia*, 35(6), 1039-1042. <https://doi.org/10.1590/S1413-70542011000600001>
- Franceschi, L. de, Benin, G., Guarienti, E., Marchioro, V. S., & Martin, T. N. (2009). Fatores pré-colheita que afetam a qualidade tecnológica de trigo. *Ciência Rural*, 39(5), 1624-1631. <https://doi.org/10.1590/S0103-84782009005000060>
- Galindo, F. S., Teixeira Filho, M. C. M., Buzetti, S., Santini, J. M. K., Alves, C. J., & Ludkiewicz, M. G. Z. (2017). Wheat yield in the Cerrado as affected by nitrogen fertilization and inoculation with *Azospirillum brasilense*. *Pesquisa Agropecuária Brasileira*, 52(9), 794-805. <https://doi.org/10.1590/s0100-204x2017000900012>
- Hossain, A., Sarker, M. A. Z., Saifuzzaman, M., Silva, J. A. T. da, Lozovskaya, M. V., & Akhter, M. M. (2013). Evaluation of growth, yield, relative performance and heat susceptibility of eight wheat (*Triticum aestivum* L.) genotypes grown under heat stress. *International Journal of Plant Production*, 7(3), 615-636. Retrieved from [http://ijpp.gau.ac.ir/article\\_1121\\_473a2481e1babb454f403aa4979faefd.pdf](http://ijpp.gau.ac.ir/article_1121_473a2481e1babb454f403aa4979faefd.pdf)
- Hungria, M., Campo, R. J., Souza, E. M., & Pedrosa F. O. (2010). Inoculation with selected strains of *Azospirillum brasilense* and *A. lipoferum* improves yields of maize and wheat in Brazil. *Plant Soil*, 331, 413-425. <https://doi.org/10.1007/s11104-009-0262-0>
- Kazi, N., Deaker, R., Wilson, N., Muhammad, K., & Trethowan, R. (2016). The response of wheat genotypes to inoculation with *Azospirillum brasilense* in the field. *Field Crops Research*, 196, 368-378. <https://doi.org/10.1016/j.fcr.2016.07.012>
- Ludwig, R. L., Martin, T. N., Stecca, J. D. L., Cunha, V. de S., Nunes, U. R., & Grando, L. F. T. (2018). Action specificity of chemical treatment and inoculation with *Azospirillum brasilense* in wheat seed on the crop initial growth. *Revista Ceres*, 65(5), 407-414. <https://doi.org/10.1590/0034-737x201865050005>
- Malavolta, E. (2006). *Manual de nutrição mineral de plantas* (1st ed.). São Paulo: Editora.
- Milléo, M. V. R., & Cristófoli, I. (2016). Avaliação da eficiência agrônômica da inoculação de *Azospirillum* sp. na cultura do milho. *Revista Scientia Agraria*, 17(3), 14-23. <https://doi.org/10.5380/rsa.v17i3.44630>
- Munareto, J. D., Martin, T. N., Fipke, G. M., Cunha, V. dos S., & Rosa, G. B. da. (2019). Nitrogen management alternatives using *Azospirillum brasilense* in wheat. *Pesquisa Agropecuária Brasileira*, 54, e00276. <https://doi.org/10.1590/S1678-3921.pab2019.v54.00276>
- Nunes, P. H. M. P., Aquino, L. A., Santos, L. P. D. dos., Xavier, F. O., Dezordi, L. R., & Assunção, N. S. (2015). Produtividade do trigo irrigado submetido à aplicação de nitrogênio e à inoculação com *Azospirillum brasilense*. *Revista Brasileira de Ciência do Solo*, 39(1), 174-182. <https://doi.org/10.1590/01000683rbcbs20150354>
- RCBPTT (Reunião da Comissão Brasileira de Pesquisa de Trigo e Triticale). (2018). *Informações técnicas para trigo e triticale—Safrá 2018/XI Reunião da Comissão Brasileira de Pesquisa de Trigo e Triticale, 2018*. Cascavel: RCBPTT.
- Sá, J. C. de M., Lal, R., Cerri, C. C., Lorenz, K., Hungria, M., & Carvalho, P. C. de F. (2017). Low-carbon agriculture in South America to mitigate global climate change and advance food security. *Journal Environment International*, 98, 102-112. <https://doi.org/10.1016/j.envint.2016.10.020>
- Tedesco, M. J., Gianello, C., Bissani, C. A., Bohnen, H., & Volkweiss, S. J. (1995). *Análise de solo, plantas e outros materiais*. Porto Alegre: Universidade Federal do Rio Grande do Sul.

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