

Seed and leaf inoculation with *Azospirillum brasilense* and increasing nitrogen in wheat production

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ABSTRACT: The objective was to evaluate the efficiency of inoculation based on the bacteria *Azospirillum brasilense*, in wheat crop performance with increasing nitrogen rates, in temperate conditions. The experiment was conducted on the wheat crop in a no-tillage system at the Federal University of Santa Catarina - Brazil, over two years (2013 and 2014). The experimental design was randomized blocks, in a 3x4 factorial model, with four replicates, combining three forms of wheat inoculation (seed inoculation, no inoculation, and leaf inoculation) with four N rates (0, 75, 150 and 225 kg ha⁻¹ of N). A joint analysis of experiments (two assessment years) was used. Biometric parameters and yield components were evaluated. There was no three-way relationship among the factors. Inoculation effect was only significant for ear length and plant height, with different response accordingly to assessment year. The effect of N rate was a quadratic response for all variables. The best results for the variables were with the application of 150 to 225 kg N ha⁻¹. The application of 215 kg N ha⁻¹ with no inoculation resulted in the best management for wheat cultivation.

Key words: diazotrophic bacteria; grain yield; *Triticum aestivum*

Inoculação via semente e foliar com *Azospirillum brasilense* e incremento de nitrogênio na produção de trigo

RESUMO: O objetivo foi avaliar a eficiência da inoculação com bactérias *A. brasilense* no desempenho da cultura do trigo, com doses crescentes de nitrogênio, em condição de clima temperado. O experimento foi conduzido com a cultura do trigo em sistema plantio direto na Universidade Federal de Santa Catarina – Brasil, em dois anos (2013 e 2014). Foi utilizado o delineamento experimental de blocos ao acaso com esquema fatorial 3x4, com quatro repetições, combinando três formas de inoculação (inoculação na semente, sem inoculação e inoculação via foliar) com quatro doses de N (0, 75, 150 e 225 kg ha⁻¹ de N). Foi utilizada a análise conjunta de experimento (considerando os dois anos de avaliação). Foram avaliados os parâmetros biométricos e de rendimento. Não houve interação tripla entre os fatores. Efeito da inoculação foi apenas significativo para comprimento de espiga e altura de plantas, com diferentes resultados de acordo com o ano de avaliação. O efeito da dose de N foi quadrático para todas as variáveis. A inoculação na semente ou via foliar no trigo não afetou os componentes de rendimento ou produtividade de grãos de trigo. Os melhores resultados para as variáveis foram com a aplicação de 150 a 225 kg de N ha⁻¹. A aplicação de 215 kg de N ha⁻¹ sem inoculação, resultou na melhor opção de manejo para o cultivo do trigo.

Palavras-chave: bactérias diazotróficas; rendimento de grãos; *Triticum aestivum*

Introduction

Wheat (*Triticum aestivum*) is a vital winter crop for the South of Brazil, being one of the few income sources during the cold season. The states of southern Brazil are significant producers of wheat, representing around 92% of the national production (CONAB, 2015). However, Brazilian grain crop production in 2014 was 5,971 Mg compared to the demand of 12,209,000 Mg, requiring importation of this cereal (CONAB, 2015), a situation which has been repeated over the years. Therefore, it is necessary to develop techniques that allow better use of cultivated areas, to increase productivity, with soil fertility management, especially regarding to nitrogen (N) fertilization.

N is the nutrient required in most considerable quantity for the wheat crop, and it is the most limiting element to productivity, as it determines tiller development (Dobbelaere et al., 2002), increases yield and determines the concentration of protein in the grain. However, the plant uses only part of the N added through fertilization because this nutrient efficiency mainly depends on soil and climate conditions, as well the crop efficiency of N uptake. Studies show that wheat plants use only 50% of the N fertilizer applied, the other half is lost through processes such as volatilization, leaching, and denitrification (Dobbelaere et al., 2002).

On average, wheat exports 22 kg of N per Mg of grains, therefore, it is essential that this amount is provided at some crop development stage (SBCS and CQFS-RS/SC, 2016). However, close attention should be paid to fertilization concerning the application rate, as small quantities limit productivity, high rates can lead to the lodging of the crop, making harvesting difficult and resulting in a drop in productivity and grain quality (Teixeira Filho et al., 2010). Besides that, high N rates can cause significant environmental problems, with the emission of nitrous oxide, for example, one of the three main gases responsible for the greenhouse effect (Morais et al., 2013). Several studies have shown that when using N rates up to 70 kg ha⁻¹, positive results for yield were observed (Theago et al., 2014; Nunes et al., 2015). However, these results vary according to the type of preceding crop, the cultivar and climate conditions (Prando et al., 2013), taking into account the existence of any residual N from the previous crop.

The amount of N fertilizer applied to wheat is one of the leading factors increasing production costs of this cereal (Khakbazan et al., 2013), as well as contaminating the environment if managed improperly. Thus, for a sustainable agricultural model, efficient utilization of atmospheric N is required, with a consequent reduction in the consumption of N mineral fertilizers. Growth-promoting bacteria have therefore emerged as a potential alternative, guaranteeing high productivity at low cost (Hungria, 2011).

In this regard, an alternative to reduce N mineral fertilization can be due to the use of bacteria *Azospirillum brasilense*, which is capable of supplying part of the plant

demand for N, due to the biological nitrogen fixation (BNF) (Hungria, 2011). Studying this bacteria on wheat, Sala et al. (2007) highlighted that when using different N rates, in combination with seed inoculation with *A. brasilense*, a greater increase in grain productivity was seen, without additional N. The authors attributed these benefits caused by the bacteria, to the plant's capacity for phytohormone synthesis.

Although wheat inoculation with *A. brasilense* is becoming a common practice, as the productivity increases (Hungria et al., 2010), incompatibility problems with insecticides and fungicides for seed treatment, has been reported (Fukami et al., 2016). In this way, alternative methods for the use of this technology emerge to mention the inoculant leaf application, increasing the bacteria efficiency, as well reducing incompatibility with the pesticides in seeds treatment (Fukami et al., 2016).

Therefore, this study hypothesizes that with *Azospirillum brasilense* inoculation, the amount of N fertilizer applied in top dressing to wheat can be reduced, without affecting the expected grain yield. In this respect, the objective of this work was to evaluate the impact of *A. brasilense* inoculation applied to the seed and leaf, with increasing N rates, on the performance of wheat crop in a temperate climate.

Material and Methods

A field experiment was conducted over two growing seasons, winter of 2013 and 2014, in the experimental area of the Federal University of Santa Catarina (UFSC), Curitibanos, located in Southern Brazil latitude 27°S, longitude 50°W and at an altitude of 1050m, in an Inceptisol (Soil Survey Staff, 2014) with clayey texture (550g kg⁻¹ of clay). The area was cultivated in the no-tillage system for five years. The soil chemical characterization before the experiment implementation is in Table 1.

Information on rainfall and temperature during the experiment, over the two years of the study, can be found in Figure 1. According to Köppen, the climate of the region is classified as temperate Cfb, mean annual rainfall and temperature are 1600 mm and 16,5 °C, respectively.

The experimental design was randomized blocks, in a factorial arrangement of 3x4 with four replicates. The factors were: three forms of inoculation (seed inoculation, no inoculation and leaf inoculation in emergent leaves) with four nitrogen rates (0, 75, 150 and 225 kg ha⁻¹ of N, in urea form). The evaluations took place over two years of wheat cultivation (2013 and 2014). The dimensions of the plots

Table 1. Chemical characterization of the study area before implementation of the experiment.

OM ⁽¹⁾ (g dm ⁻³)	pH CaCl ₂	P ⁽²⁾ (mg dm ⁻³)	K ⁺⁽²⁾	Ca ²⁺⁽³⁾	Mg ²⁺⁽³⁾	Al ³⁺⁽³⁾	V (%)
49.59	5.90	20.75	0.18	10.20	3.10	0.00	82.05

OM= Organic Matter; V = Base Saturation. ⁽¹⁾ Walkley Black; ⁽²⁾ Mehlich-1; ⁽³⁾ KCl 1 mol L⁻¹; pH measured in solution CaCl₂ 0.01 mol L⁻¹.

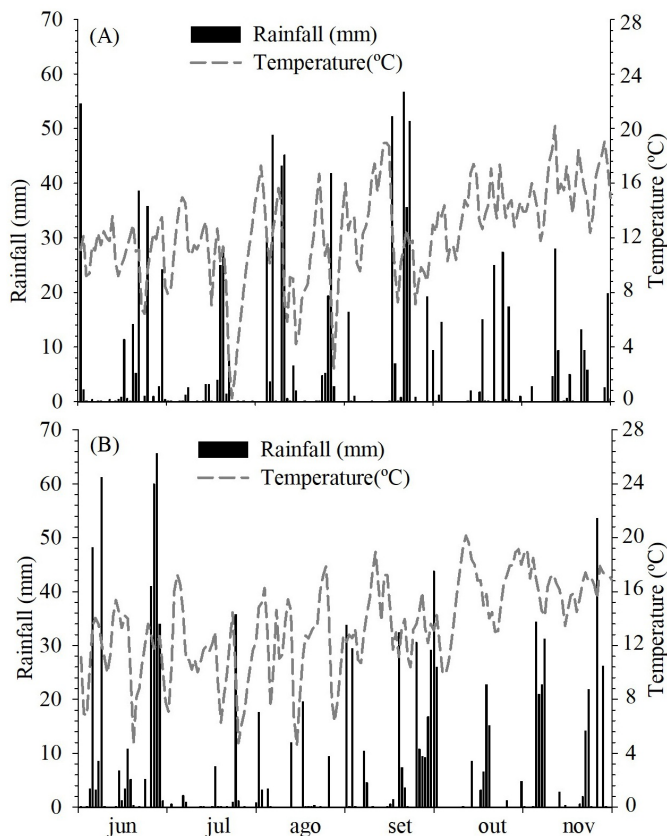


Figure 1. Rainfall and daily mean temperature for the months corresponding to the period of experiments at the Federal University of Santa Catarina. Where (A) 2013 and (B) 2014.

were 3 x 1.5 m, with 9 rows spaced at 0.17 m, with 45 seeds per linear meter. The wheat CD 108 cultivar was used, which was chosen due to its adaptation to climate conditions of the region, its resistant to lodging and its short emergence to maturation cycle, as well as moderately resistant to natural threshing (COODETEC, 2015).

In the first year, the experiment was implanted on field conditions, on 06/20/2013 and in the second year on 06/12/2014. For 2013 experiment, preceding crop was a mix of black oats and ryegrass straw (sown in June 2012), which was desiccated with the herbicide Glyphosate at a concentration of 1.44 kg ha⁻¹ of a.i. (active ingredient). In 2014, the wheat crop was sown following corn cultivation under a no-till system, in the same area as the previous year, after desiccation during pre-sowing with Glyphosate (1.44 kg ha⁻¹ of a.i.). Fungicides used in the seed treatment had chemical compositions of Carbendazim + Thiran (45 + 105 g a.i. in 100 kg of seeds). In the first year of the experiment, weed control was carried out through manual removing and in the second year using Metsulfurom Metilico (2.4 g ha⁻¹ of a.i.) and Methyl Iodosulfurom (3.5g ha⁻¹ of a.i.) post-emergence. For disease control Azoxystrobin + Cyproconazole (60 + 24 g ha⁻¹ of a.i.) was used, while for pest control was applied Imidacloprid + Beta-Cyfluthrin (50 + 6.25 g ha⁻¹ of a.i.). The products above were applied in solution using a crop sprayer

with carbon dioxide pump at a flow rate of 200 L ha⁻¹ using a flat-fan spray nozzle.

A basal application of 00-12-12 (N-P-K) fertilizer was used at 400 kg ha⁻¹, in both years. In the seed inoculation with *A. brasilense* process, a dosage of 125 mL of the commercial product Azototal[®] (2.0 x 10⁸ cfu mL⁻¹) was used for every 50 kg of wheat seeds. The inoculation was performed after the seed treatment with the use of a plastic bag, where the seeds were put in contact with the product and shaken to homogenize the distribution of the solution containing the bacteria. This activity was carried out just before sowing. For the leaf inoculation process, performed during the post-emergence period of the crop, 350 mL of the commercial product Azototal[®] (2.0 x 10⁸ cfu/mL) per hectare was used using a crop sprayer with a carbon dioxide gas pump at a flow rate of 150 L ha⁻¹, with a flat-fan spray nozzle. The leaf inoculation was applied on 3 DAE (days after emergence), in both years. The N fertilization for all the treatments was divided into two periods, the first 15 DAE and the second during tillering.

The plant height was measured at the beginning of the grain filling phase, taken as the distance from ground level to the end of the ears, excluding the awns. The wheat harvest was performed manually for each experimental unit after grain maturation, through the gathering of the four central lines of each plot, totaling an area of 4.5 m². The harvested material was mechanically threshed, weighed and transformed to kg ha⁻¹. The grain moisture was corrected to 14 %. The number of grains and spikelets per ear were performed by counting 10 ears sampled per experimental unit. The number of grains per spikelet was calculated by dividing the total number of grains per ear by the total number of spikelets per ear for each process. Additionally, the ear length was determined by taking 10 ears per experimental unit, measuring from the base of the ear to its end, not including the awns. The 1.000 grain mass was determined by counting 300 grains and correcting for grain moisture at 14 %.

The results were submitted to ANOVA for joint analysis of experiments (two assessment years) in a factorial (inoculation form *versus* N rate) randomized blocks. The means of the years and inoculation forms were compared using Tukey test (p<0.05) and regression analysis according to N rates, with 5 % probability level.

To carry out the economic analysis and calculate the Maximum Economic Efficiency (MEE), a value of US\$ 0.44 for each 1 kg of urea and US\$ 0.16 for each 1 kg of wheat was used. These values were obtained from local commercial trade during the experimental period. The maximum Technical Efficiency (MTE) and MEE were determined using the following equation:

$$y = a + bx + cx^2 \quad (1)$$

To determine the value for "x" in MTE the derivation of the implicit function was performed:

$$\frac{dy}{dx} = b - 2 \times c \times x \quad (2)$$

“x” for MEE was determined using the equation:

$$x = \frac{\left(\frac{\text{input}}{\text{product}} - b \right)}{2 \times c} \quad (3)$$

Results and Discussion

The variance analysis results did not show a three-way interaction between inoculation forms, N rates and assessment years (Table 2). There were no significant impacts of inoculation (Table 2). Considerable influence was observed for assessment year for the variables plant height, ear length, grains per spikelet, grains per ear and 1000 grain mass (Table 2). Nitrogen did not affect 1000 grain mass and grains per spikelet (Table 2).

Grains per spikelet and 1000 grain mass only had differences among the years of evaluation, with a decrease in 2014 (Table 3). It is assumed that these results were influenced by weather conditions for each year (Figure 1A and 1B), as the experimental area and cultivar remained the same. The occurrence of high temperatures during wheat cultivation shortened the cycle duration, the plant height and percentage of flower fertilization, as well as reduced the 1000 grain mass (Ribeiro et al., 2012). Accordingly, in 2014 the mean air temperature from June to October was 1.1 °C higher than in 2013 (Figure 1), which must have failed a portion of the flowers, reducing the number of grains per spikelet.

Many factors affect the efficiency of the inoculation practice. According to the results obtained it is suggested that the cultivar used and the growth-promoting bacteria had no positive interaction. Sala et al. (2007), found that the number of endophytic microorganisms in wheat roots was influenced by the interaction of the plant genotype with N

Table 3. Means for the variables analyzed according to assessment year for grains per spikelet and 1000 grain mass (g), in the wheat crop.

Variable	Year		CV (%)
	2013	2014	
Grains per spikelet	3.3 a	2.9 b	8.89
1000 grain mass	35.8 a	32.6 b	10.29

Means followed by the same lowercase letter on the line, do not differ significantly from each other according to the Tukey test at 5% probability.

fertilization. As such Lemos et al. (2013), observed different responses between wheat cultivars when *A. brasilense* was inoculated and associated with N fertilization, including the cultivar used in this study, which showed low interaction with *A. brasilense*.

A relevant factor, which may have contributed to the lack of response to the inoculation, relates to the climate conditions of this experiment (Figure 1A and 1B). Accordingly to Cattelan & Vidor (1990), climatic variations interfere in microbial growth, especially in topsoil layers, which are more susceptible to variations in moisture and temperature, leading to fluctuations in growth and multiplication of bacteria. Also, the occurrence of extreme temperatures can interfere in the bonding process, thus limiting the biological effects. Reis et al. (2015) reported that the ideal temperature for *A. brasilense* growth is 37 °C. In this study, the mean temperature during the experimental period was found to be 13.3 °C in 2013 (Figure 1A) and 15.3 °C in 2014 (Figure 1B). This demonstrates the necessity for more studies to assess the effects of *A. brasilense* under particular conditions as the temperate climate, occurring on the Santa Catarina's plateau.

There was an interaction between inoculation and assessment year only for ear length and plant height (Table 2). In 2013, seed inoculation resulted in ear length of 5.9 cm, which was significantly higher than the other treatments, whereby leaf inoculation resulted in the smallest ear length (5.3 cm) (Table 4). Conversely, in 2014 the inoculation forms showed 21% higher values for ear length than in 2013 (Table 3), although the values for leaf inoculation and no

Table 2. Variance analysis refers to plant height (H), ear length (EL), grains per spikelet (GS), spikelets per ear (SE), grains per ear (GE), 1000 grain mass (M1000) and wheat grain yield (Yield), cultivated in the years 2013 and 2014, with inoculation forms and nitrogen rates.

Sources of variation	GL	Mean square						
		H (cm)	EL (cm)	GS	SE	GE	M1000 (g)	Yield (kg ha ⁻¹)
Block (year)	6	24.68 ns	0.73**	0.04 ns	0.46 ns	5.09 ns	20.25 ns	1676483.27**
Year	1	169.41*	35.86**	3.03**	0.30 ns	224.11**	233.44**	143212.23 ns
Inoculation	2	57.27 ns	0.31 ns	0.11 ns	1.54 ns	28.42 ns	15.31 ns	1045087.65 ns
Nitrogen	3	371.36**	19.80**	0.09 ns	22.85**	278.91**	19.06 ns	15381025.04**
Year*Inoc.	2	354.79**	1.41**	0.05 ns	1.26 ns	13.26 ns	2.00 ns	290506.42 ns
Nitrogen*Year	3	117.37*	1.53**	0.05 ns	4.91**	61.07**	4.02 ns	104480.40 ns
Nitrogen*Inoc.	6	53.99 ns	0.55*	0.07 ns	0.71 ns	5.93 ns	13.85 ns	118019.09 ns
Year*Inoc.* Nitro.	6	80.22 ns	0.42 ns	0.07 ns	0.61 ns	14.63 ns	4.96 ns	105910.16 ns
Error	66	39.38	0.21	0.04	0.80	10.93	9.18	343261.95
Grand mean		78.84	6.24	3.10	9.76	30.33	34.20	2370.44
CV (%)		7.69	7.43	6.15	9.18	10.90	8.86	24.72

* signifying 5% and ** signifying 1% probability, according to the Tukey test; ns = not significant.

Table 4. Means for the interaction between assessment year and inoculation (inoc.) form with *A. brasilense* for ear length and height of wheat plants.

Year	Seed inoc.	No inoc.	Leaf inoc.	CV (%)
Ear length (cm)				
2013	5.9 bA	5.6 bB	5.3 bC	14.85
2014	6.6 aB	6.9 aA	6.9 aA	16.36
CV (%)	16.65	17.91	21.46	
Plant height (cm)				
2013	85.40 aA	77.10 aB	78.49 aAB	9.85
2014	77.27 bA	75.77 aA	78.01 aA	9.74
CV (%)	10.34	12.48	13.66	

Means followed by the same uppercase letter on the line and lowercase letter in the column, do not differ significantly from each other according to the Tukey test at 5% probability.

inoculation were higher than for seed inoculation (Table 4). These results demonstrate some level of instability about the effects associated with *A. brasilense* on wheat crop.

In a study by Lemos et al. (2013), on CD 108 cultivar, the higher ear lengths were obtained in two isolated circumstances: only with inoculation with *A. brasilense*, or just with N fertilization. On the other hand, Corassa et al. (2013), reported that seed inoculation with *A. brasilense* associated with N fertilization management did not result in differences in ear length. Our results (Table 4) may be related to the occurrence of low temperatures during wheat cultivation in 2013 (Figure 1). Low air temperatures result in higher tiller production (Rodrigues et al., 2011), meaning the higher quantity of ears per area, resulting in shorter ear length (Table 4) (Teixeira Filho et al., 2010).

For plant height, significant differences only occurred in 2013, where seed inoculation resulted in height increases relative to no inoculation, which was similar to leaf inoculation (Table 4). Significant differences were only found between the study years for seed inoculation, whereby in 2013 it was higher (85.4 cm) than in 2014 (77.27 cm) (Table 4). Height difference in wheat plants was not observed by Guimarães et al. (2015), when comparing the use of a commercial inoculant based on *A. brasilense* and the supply of 80 mg dm⁻³ for N in urea (approximately 72 kg N ha⁻¹).

The results for plant height in 2013 with the application of N were higher than those for 2014 (Figure 2A), except for 0 kg of N ha⁻¹. The result of N fertilization for this variable was quadratic, as the maximum response at 158 kg N ha⁻¹, for 2013 and 155 kg N ha⁻¹, for 2014, reaching maximum plant heights of 84.15 and 79.52 cm, respectively (Figure 2A). The increase in plant height observed in this study was related to stem elongation, which is caused by the addition of N. According to Marschner (1995), the application of increasing N rates during the initial stages of cereal development promotes an increase in the production of growth-promoting phytohormones responsible for cell division and expansion.

For some spikelets per ear (Figure 2B) and some grains per ear (Figure 2C), a decrease was observed in 2013 with the application of N rates higher than 157.2 and 164.5 kg N ha⁻¹, respectively. However, for 2014, it was observed an increase for spikelets up to the highest rate, being estimated

at 332.8 kg N ha⁻¹ for this variable and 222 kg N ha⁻¹ for grains per ear (Figure 2B and 2C).

In work carried by Coelho et al. (1998), for these parameters, a linear result was observed with an increase in N rates. The positive response to N application may be due to the fact that this nutrient provides an addition of gibberellic acid content to the plant, leading to greater protein synthesis and consequently stimulating greater flowering (Marschner, 1995), thereby increasing the number of flowers per ear and increasing ear length (Coelho et al., 1998).

According to the interaction between N rate and inoculation form for ear length, using the MTE calculation,

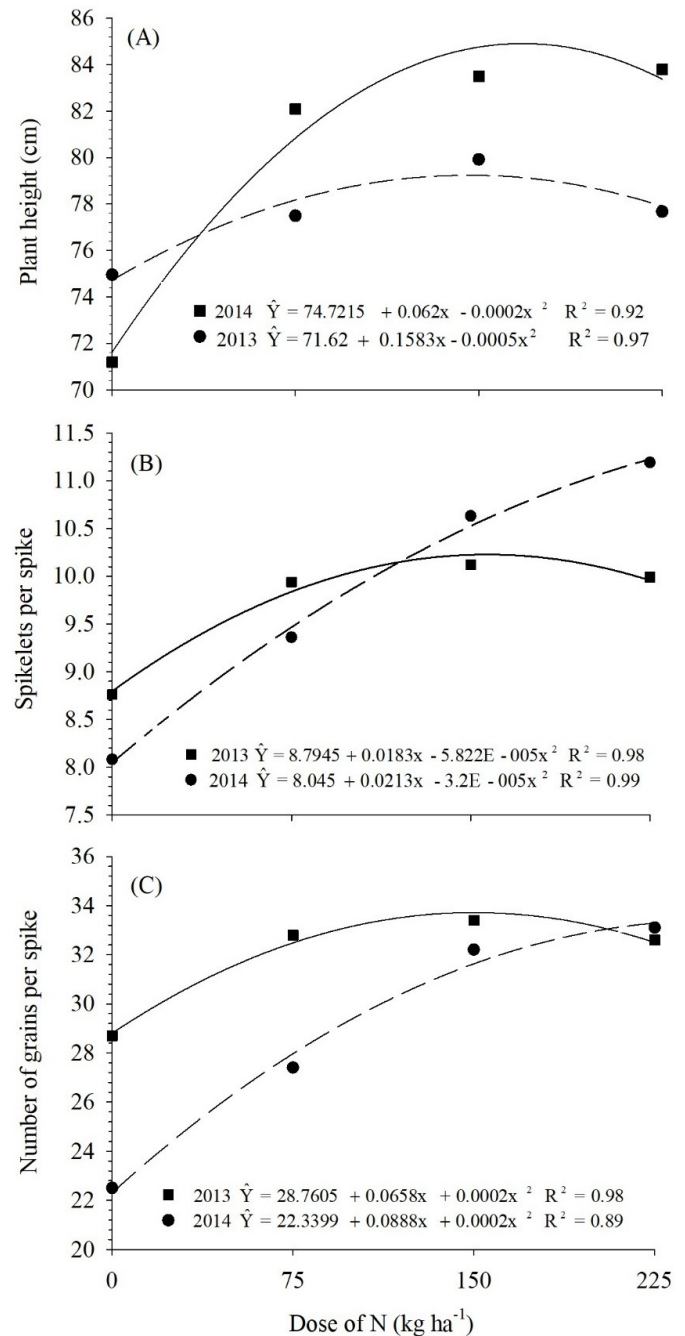


Figure 2. Regressions between the assessment year and N rate interaction, for plant height (A), spikelets per ear (B) and number of grains per ear (C) in wheat crop.

it can be observed that seed inoculation resulted in positive responses for the higher rates tested in this study (Figure 3A). While for leaf inoculation and no inoculation, MTE was estimated at 228.3 and 202 kg of N ha⁻¹, respectively, reaching maximum ear length of 7.07 and 7.02 cm (Figure 3A). For the treatment without N application, seed inoculation showed highest results, whereas leaf inoculation showed the lower. However, for 75 and 150 kg of N ha⁻¹ rates, no inoculation was the most efficient (Figure 3A). This result differs from those found by Corassa et al. (2013) that combining inoculation with N application, compared with no N application, no significant result was seen. For ear length, increasing the N rate to 215 kg of N ha⁻¹ in 2013 and 264 kg of N ha⁻¹ in 2014 resulted in highest ear length, reaching 6.17 and 8.05 cm, respectively.

No significant increase in wheat grain yield has been demonstrated with *A. brasilense* inoculation, in any of the forms studied (Table 2). The determining factor for the effectiveness of the inoculation concerns in the ability of diazotrophic bacteria to compete with the native soil community, on which the competitive efficiency is only maximized under low availability of N in soil conditions (Bashan & de Bashan et al., 2015). High organic matter content and the subsequent higher availability of N may mitigate the benefits of *Azospirillum* sp inoculation (Sangoi et al., 2015). Although these authors have studied the effect of inoculation on corn, it is assumed that the high content

of organic matter in this study area (Table 1) may have influenced in the efficiency of the inoculation in wheat.

The result of this work was different to the study carried out by Hungria et al. (2010), in which through the utilization of *A. brasilense*, AB V5, and AB V6 strains increased 13 to 18% in wheat grain yield, when compared with no inoculation. Corassa et al. (2013), evaluating wheat yield with seed inoculation, with and without N application, found higher yield when combining seed inoculation with the N application in top dressing, a different result was observed in this study.

N rates quadratically affected wheat grain yield (Figure 4). Therefore, MTE was calculated for 234 kg of N ha⁻¹. However, MEE was estimated to 215 kg N ha⁻¹, reaching a grain yield of 3.068 kg ha⁻¹ (Figure 4). In a study carried out by Theago et al. (2014), evaluating the crop response to N rates, the maximum yield was estimated in 134 and 128 kg of N ha⁻¹ for IAC 370 and Embrapa 21 cultivars, respectively. Nunes et al. (2015), evaluating rates of 20, 60, 100, 140 and 180 kg ha⁻¹ of N, obtained increases in wheat yield with rates up to 140 kg ha⁻¹ cultivated under soil condition with low N residual in succession to maize. The higher N rates of maximum grain yield in this experiment, compared to other studies, may be related to the fact of the preceding crop in both years were grass crops (maize and black oat plus ryegrass) with high C/N ratio, resulting in immobilization of a significant amount of applied N, thus reducing the efficiency N fertilization.

No positive impact was found on the 1000 grain mass for N rates or for inoculation (Table 2). Galindo et al. (2015), studying irrigated wheat in the Brazilian *cerrado*, observed that regardless of the *A. brasilense* inoculation form there was no significant impact on the 1000 grain mass or yield. Similar results were also observed in Corassa et al. (2013) assessing N rates in combination with *Azospirillum* inoculation, with no significant differences found, which corroborates with the results from this study.

Although inoculation with *A. brasilense* in wheat aids the N absorption, this practice is not capable of completely

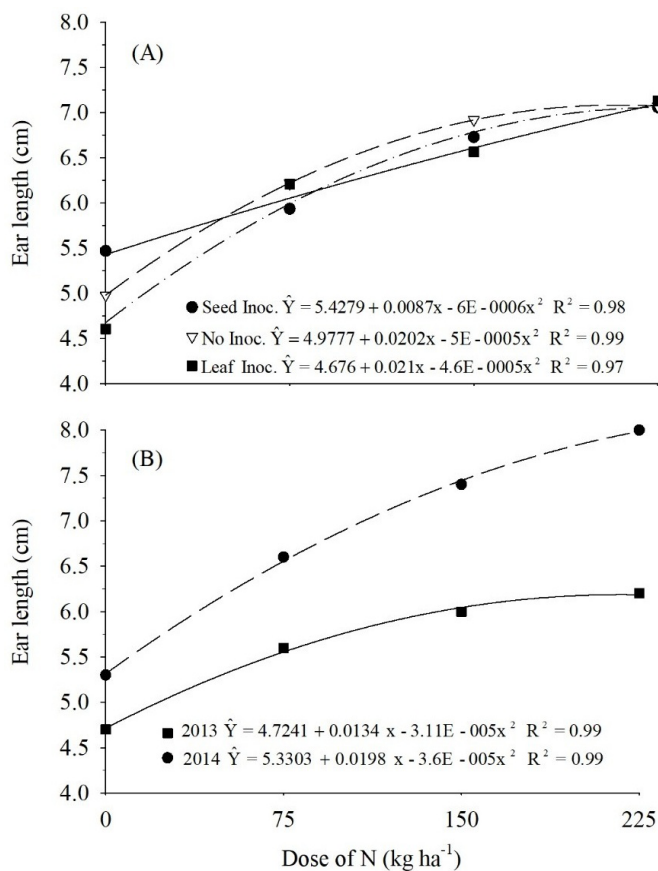


Figure 3. Interaction regressions between N rate and inoculation form (A) and between N rate and assessment year (B) for ear length in wheat crop.

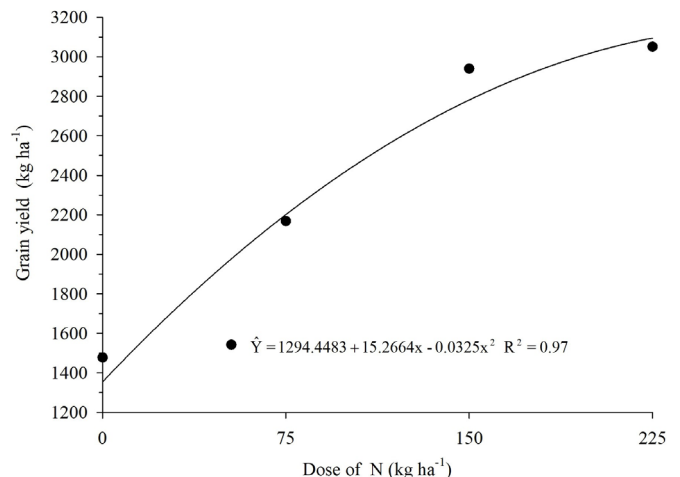


Figure 4. Mean wheat grain yield according to N rates in the two assessment years (2013 and 2014).

replacing mineral N fertilization (Hungria, 2011). As in the example of wheat, few studies demonstrate benefits from symbiotic interaction in grass leaves that are capable of providing the action of diazotrophic bacteria on entering the plant tissue and subsequently processing N, thus causing increases in productivity (Ferreira et al., 2014). The differences for the variables over the two years reflect the inconsistency obtained for inoculation of wheat with *A. brasilense*, the same was found by Nunes et al. (2015), who indicate that the results over two assessment years do not always show the same trend. Secondly, according to Hungria et al. (2010), despite advances in basic and applied research, the results obtained from field experiments on the agronomic efficiency of *Azospirillum* based inoculants are not consistent. Therefore, there is a need for more studies to be carried out, evaluating different cultivars in more diverse climates and soil conditions.

Conclusions

A. brasilense resulted in inconsistent results over the two years of evaluation.

The inoculation in seeds and leaves of wheat with *A. brasilense* promoted an increase in plant height, but it did not affect the yield components either grain yield.

The best results for all variables were between the rates of 150 and 225 kg N ha⁻¹.

Not inoculating the wheat with *A. brasilense* and applying 215 kg N ha⁻¹ was the best management of nitrogen fertilization, under the conditions of this study.

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