# Evaluation of phyllite and basalt associated with *Azospirillum brasilense* inoculation in oat cultivation<sup>1</sup>

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**ABSTRACT** - The objective of this study was to evaluate the performance of white oat in the presence of phyllite and basalt dusts associated with *Azospirillum brasilense* inoculation. The experiment was conducted in a completely randomized design (n = 5) with the following treatments: FB, fertilizer + basalt dust; FP, fertilizer + phyllite dust; FA, fertilizer + Azospirillum brasilense; FBA, fertilizer + basalt dust + Azospirillum brasilense; FPA, fertilizer phyllite dust + Azospirillum brasilense; C, Control. The following variables were evaluated: plant variables - emergence speed index (ESI), Fresh matter (FM), dry mass (DM), root length (RL), plant height (H) and root dry mass (RDM); and soil variables - phosphorus (P), potassium (K), calcium (Ca), magnesium (Mg), aluminum (Al), potential soil acidity (PSA), base saturation (BS) and cation exchange capacity (CEC). Phyllite application resulted in lower RL, but promoted the highest H and FM. On the other hand, basalt application increased RDM production when associated with inoculation. In addition, there was an increase in the base content of the soil when phyllite dust and basalt dust were applied. It can be observed that, throughout the cuts, the association of rock dusts with inoculation allows more stability of FM and DM production, increasing the forage potential of oat in the third cut by up to 20%. It is concluded that the association of phyllite and basalt with *Azospirillum brasilense* affects the chemical properties of the soil, promoting benefits to white oat cultivation.

Key words: Rock powder. Alternative inputs. Avena sativa L.. Beneficial bacteria.

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## **INTRODUCTION**

White oat (*Avena sativa* L.) is a winter cereal widely cultivated in southern Brazil because it is suitable for several purposes (Malanchen *et al.*, 2019). Oat grains and forage are among the main sources of feed used in livestock activities (Kaspary *et al.*, 2015), and phytomass production makes its cultivation an excellent option as ground cover (Silva *et al.*, 2021). The crop is part of the group of species classified as demanding in terms of fertility, so the availability of macro and micronutrients is directly related to its yield (Fontaneli; Santos; Fontaneli, 2012).

Chemical fertilizers represent the main source of nutrients applied to soils to meet crop requirements (Franco Junior *et al.*, 2023), but these inputs make up a significant portion of the cost of agricultural production and trigger environmental discussions because are not a sustainable option. On the other hand, remineralizers are natural inputs (Viana *et al.*, 2021) that promote the replacement of nutrients to soils (Benevides Filho *et al.*, 2023) and allow for increase economic indices in agricultural production (Galina *et al.*, 2023).

There are several rocks with potential for agricultural use (Theodoro *et al.*, 2021). Among them, basalt and phyllite have the potential to improve soil fertility (Rezende *et al.*, 2021) and favor the development of forage species. Basalt is an igneous rock, while phyllite is a metamorphic rock formed from sediments (Costa, 2021). Both rocks contain in their mineralogical composition silicate minerals with potassium (K<sub>2</sub>O), calcium (CaO), magnesium (MgO), iron (Fe<sub>2</sub>O<sub>3</sub>), copper (CuO), zinc (ZnO), manganese (MnO) and aluminum (Al<sub>2</sub>O<sub>3</sub>) oxides (Costa, 2021), which can be gradually released into the soil profile during the weathering process.

The release of nutrients that are retained in inorganic forms in rocks is a complex and heterogeneous process (White *et al.*, 2017), but it can be accelerated by the biological action of organisms that live in the soil due to their action on mineral weathering (Sharma *et al.*, 2023). Plant growth-promoting rhizobacteria (PGPR) are microorganisms that live in symbiosis in the rhizosphere, promoting benefits due to the ability to release specific substances (amino acids, siderophores, phytohormones, organic acids) during the metabolization of exudates released by the roots, stimulating plant growth and development (Santos *et al.*, 2021).

Among the PGPR, *Azospirillum brasilense* stands out, a free-living bacterium that also has the ability to perform Biological Nitrogen Fixation (BNF) (Spolaor *et al.*, 2016), especially in grasses. In addition, some species of bacteria of the genus *Azospirillum* spp. have the ability to act in the solubilization of phosphate  $(P_2O_5)$  and  $K_2O$  (Fukami *et al.*, 2016). Therefore, *Azospirillum* 

*brasilense* can be used via inoculation in order to improve the action of phyllite and basalt application in the soil.

The hypothesis was investigated that the application of phyllite and basalt dusts affects the chemical properties of the soil and the vegetative development of white oat, with inoculation being a catalyst for the effects. The objective of this study was to evaluate the performance of white oat cultivated with phyllite and basalt dusts associated with *Azospirillum brasilense* inoculation and the influence of this management on soil chemical attributes.

# **MATERIAL AND METHODS**

#### Experimental design and cultural practices

The experiment was conducted with pots in an environment under controlled temperature  $(23 \pm 2 \text{ °C})$  and photoperiod (12 h light and 12 h dark), using a lighting system with LED Grow light lamps (Murphy *et al.*, 1992). Each experimental unit was represented by 1 dm<sup>-3</sup> polyethylene pots with capillary system after soil saturation to 60% of field capacity. A completely randomized design was used with 5 replicates of six treatments (Table 1).

The soil used was collected in an agricultural area cultivated under no-till in the municipality of Quilombo - SC (26° 42' 6.72' S e 52° 46' 47.21"W), in 0-10 cm depth, classified in Cambissolo húmico for Brazilian System of Soil Classification (SiBCS) proposed by Santos et al. (2018), and Inceptisol to according to Soil Taxonomy (Soil Survey Staff, 1999). The soil was sieved through a 4 mm mesh with field capacity (FC) corrected to 60% at the time of sowing. Soil chemical analyzes used the methodology proposed by on the Manual of Methods and Analyses of the Official Network of Laboratories (Rede Oficial de Laboratórios - ROLAS) (Silva et al., 2019): clay 39.15%, hydrogen potential (pH) 5.30, SMP 6.60, organic matter 2.90%, phosphorus (P) 49.90 mg dm-3, potassium (K) 156.00 mg dm-3, calcium (Ca) 10.23 cmol<sub>a</sub> dm<sup>-3</sup>, magnesium (Mg) 4.20 cmol<sub>a</sub> dm<sup>-3</sup>, cation exchange capacity at pH7 (CEC) 30.96 cmol dm-3.

Table 1 - Description of the treatments in the research

Treatments	Description		
С	Control (Fertilizer)		
FB	Fertilizer + basalt dust		
FP	Fertilizer + phyllite dust		
FA	Fertilizer + Azospirillum brasilense		
FBA	Fertilizer + basalt + Azospirillum brasilense		
FPA	Fertilizer + phyllite + Azospirillum brasilense		

Fertilization was calculated based on soil analysis for an expected yield (green mass) of 36 t ha<sup>-1</sup>, adjusted to 1 dm<sup>-3</sup> based on the Liming and fertilization manual for the states of Rio Grande do Sul and Santa Catarina (Gatiboni; Silva; Anghinoni, 2016). The fertilizers [triple superphosphate (TSP), urea, chlorinated urea (NK) and potassium chloride (KCl)] were weighed on an analytical precision scale and incorporated into the soil before sowing, through homogenization in plastic bags and distributed to the pots, followed by sowing. The equivalent of 30 kg ha<sup>-1</sup> of N was applied for the planting fertilizer and 120 kg ha<sup>-1</sup> was applie on the surface in three applications of 40 kg ha<sup>-1</sup> after each cut of the oats. The soil was not limed.

Studies with remineralizers of different compositions tested doses ranging from 1 to 100 t ha-1 (Swoboda; Doring; Hamer, 2022), however the most used dose for experiments is 2.5 t ha<sup>-1</sup> (Soroatto et al., 2021; Almeida Júnior et al., 2022; Luchese et al., 2023). Therefore, basalt and phyllite dusts were sieved through a 0.35 mm mesh and applied to the soil of each treatment considering an equivalent dose of 2.5 t ha-1. The dusts had: 7,6 and 8,8 pH; 0,96 and 1,60% of P<sub>2</sub>O<sub>5</sub> [neutral ammonium citrate (NAC) + water]; 0,00 and 0,06% K<sub>2</sub>O in water; 7,37 and 2,32% of Ca; 1,03 and 1,37% of Mg; 0,17 and 0,17% of N; 0,010 and 0,019% of Cu; 0,010 and 0,004% of Zn; 0,042 e 0,017% of Mn, respectively, basalt and phyllite. The dusts used were intended to condition the soil and not replace fertilization.

The oat cultivar Fapa 43 was used in the experiment. Plant thinning occurred 15 days after sowing (DAS), leaving with a population of six plants per pot, and inoculation was carried out via seeds at a dose of  $2 \text{ mL kg}^{-1}$ , using a commercial inoculant (2 x  $10^8$  colony-forming units) composed of the Abv5 and Abv6 strains of the bacterium *Azospirillum brasilense*.

## **Determination of Plant Variables**

The first cut was carried out 15 days after sowing and the other evaluations were performed whenever the control plants reached 20 cm in height, totaling three cuts with measurements of the response variables. At each cut, 10 cm of plant was left for regrowth, simulating the height for field grazing, and made with the application of nitrogen fertilizer. Emergence speed index (ESI) was measured daily by counting the seedlings from the moment when the first leaf emerged, for eleven days, to determine the number of plants emerged per day as proposed by Maguire (1962).

Fresh matter (FM) was determined at each cut, preserving 10 cm of stubble and dehydrating the samples in a forced circulation oven at 105 °C until reaching a constant weight for the determination of dry mass (DM). Root length (RL) was determined by measuring the roots with a graduated ruler at the end of the crop cycle. Root dry mass (RDM) was determined after dehydrating the samples in a forced circulation oven at 105 °C at the end of the experiment. Plant height (H) was determined measuring the plants with a graduated ruler at the end of all the cuts.

### Determination of soil chemical attributes

The soil was collected at the end of the crop cycle and sieved through a 2-mm mesh for the analysis of the following variables: phosphorus (P), potassium (K), calcium (Ca), magnesium (Mg), potential soil acidity (PSA), base saturation (BS), cation exchange capacity (CEC) and aluminum saturation (Al) in CEC. P content was determined in a visible light spectrophotometer after extraction with Mehlich-1 solution (0.05 mol L<sup>-1</sup> hydrochloric acid and 0.0125 mol L<sup>-1</sup> sulfuric acid); K content was determined by flame photometry after extraction with Mehlich-1 solution; Ca, Mg and Al contents were determined in a flame atomic absorption spectrophotometer after extraction in 1 mol L<sup>-1</sup> potassium chloride solution.

PSA was estimated with buffered calcium acetate (pH 7.0) with volumetric determination in sodium hydroxide (NaOH) solution in the presence of phenolphthalein indicator; CEC was obtained by the sum of  $Ca^{+2}$ ,  $Mg^{+2}$  and  $K^+$  cations and PSA; BS was calculated by the ratio between  $Ca^{+2}$ ,  $Mg^{+2}$  and  $K^+$  cations and CEC. All soil variables mentioned were determined based on the Manual of Methods and Analyses of the Official Network of Laboratories (*Rede Oficial de Laboratórios* - ROLAS) (Silva *et al.*, 2019).

#### Data processing and statistical analysis

The data set was subjected to normality and homoscedasticity evaluation by the Shapiro-Wilk and Levene tests, and were transformed by approximation using the Box-Cox method when necessary. When normal, the data were subjected to analysis of variance according to the design described, and when significant F, the means were compared by the LSD test at 5% probability level (P < 0.05). Multivariate exploration was carried out by initially identifying the multicollinearity of the variables tested with the Variance Inflation Factor and application of Forward Selection to identify significant variables, followed by principal component analysis (PCA) to check the spatial distribution of the treatments along with the plant and soil variables. All statistical procedures were performed in the RStudio software and the figures in the Origin software.

## **RESULTS AND DISCUSSION**

#### Vegetative characteristics of plants

Plant variables were affected by the treatments tested. Figure 1 shows that the emergence speed index

(ESI) had a significant difference, showing that the treatments FBA, FP and control achieved better ESI compared to the treatments that received inoculation with *Azospirillum brasilense* (FA) and application of basalt (FB).

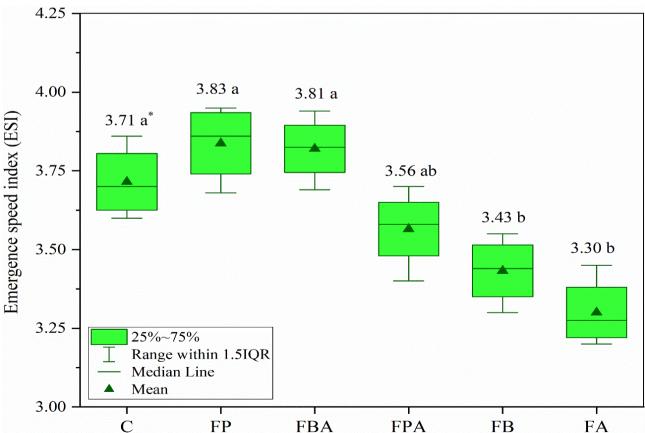
The association of *Azospirillum brasilense* with basalt dust indicates a positive effect of the FBA treatment, increasing the ESI of oats when compared to single application. Germination and seedling emergence speed is a variable that is related to the qualitative characteristics of seeds (Freitas *et al.*, 2022). However, the use of basalt and phyllite dusts seems to affect the emergence speed of white oat seedlings, especially when associated with inoculation.

The ESI found in treatment FA indicates that there is no individual effect of bacterial action for this variable. This result corroborates that reported by Worma *et al.* (2019), who evaluated the physiological quality of maize seeds subjected to different treatments with *Azospirillum* spp. and found no effects on seedling emergence. However, the use of *Azospirillum*  *brasilense* together with basalt dust in the present study accelerated the emergence of white oat seedlings.

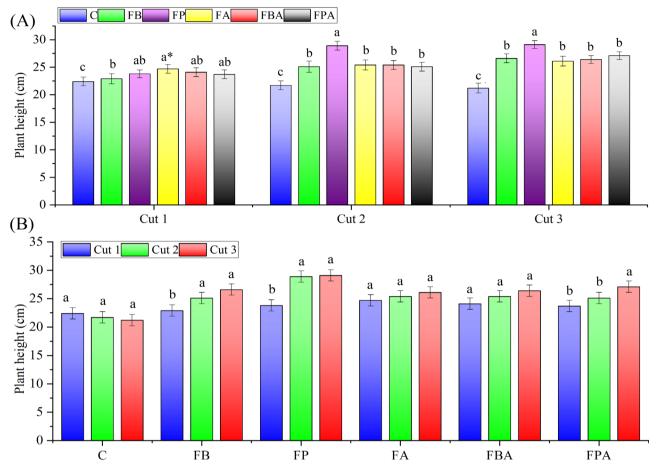
Wheat seeds coated with basalt and granodiorite showed superior performance in terms of seedling emergence, emergence speed, and dry mass (Eberhardt *et al.*, 2019), indicating that the use of remineralizers can improve traits related to germination and seedling emergence in specific crops. The presence of remineralizers in the soil can affect the water retention capacity, reducing evaporation losses, as smaller particles increase the surface contact area (Rudmin *et al.*, 2020). In this context, the soil of the treatments that received rock dust would have a higher water retention capacity, allowing better conditions of germination and emergence for seeds and seedlings, which justifies the average ESI found in the treatment that received only phyllite dust (FP).

The data presented in Figure 2 show statistical difference (P = 0.021; F = 3.583) for the variable plant height (H) between treatments (Figure 2A) and between cuts (Figure 2B) in the aerial part of white oat.

**Figure 1** - Emergence speed index (ESI) of white oat seedlings in treatments: C, control; FB, fertilizer + basalt; FP, fertilizer + phyllite; FA, fertilizer + *Azospirillum brasilense*; FBA, fertilizer + basalt + *Azospirillum brasilense*; FPA, fertilizer + phyllite + *Azospirillum brasilense*; \* Different letters show statistical difference between treatments using the LSD test (P < 0.05)



**Figure 2** - Plant height (H) of the treatments control (C), fertilizer + basalt (FB), fertilizer + phyllite (FP), fertilizer + *Azospirillum brasilense* (FA), fertilizer + basalt + *Azospirillum brasilense* (FBA), fertilizer + phyllite + *Azospirillum brasilense* (FPA) in the three cuts of the shoot part of the white oat crop. \*Different letters indicate statistical difference between treatments for each cut (A) and between cuts for each treatment (B) by LSD test (P < 0.05)



In the first cut, plant height was higher in treatment FA (24.7 cm), whose values were statistically higher than those found in treatments FB (22.9 cm) and C (22.4 cm). In the second and third cuts, treatment FP showed superiority in plant height, as can be seen in Figure 2A, while the lowest values were found in the control.

When evaluating the response of maize to seed inoculation with *Azospirillum brasilense*, Oliveira *et al.* (2024) obtained higher plant height in the inoculated treatments due to the production of growth-promoting substances (phytohormones, extracellular substances, mucilages) by the inoculated bacterial strains. Concari *et al.* (2023) also reported an increase in the height of white oat plants in treatments that involved seed inoculation with *Azospirillum brasilense*.

In this context, the best result obtained for the treatments inoculated with *Azospirillum brasilense*, during

the evaluation in the first cut, occurred as a direct response to the inoculation of oat seeds. In the initial stages of germination and emergence, the seedling has part of its growth process guaranteed by seed reserves, with low demand for nutrient absorption (Fabiani *et al.*, 2019). However, when the first cut occurs, the action of bacteria is important to promote vegetative growth, affecting the development of the aerial part of white oat plants.

The data presented in Figure 2B show that the application of remineralizers, especially phyllite dust, tends to promote long-term effects, because, as can be seen, the treatments that did not receive the application of remineralizers (C and FA) showed no differences between the cuts. This effect occurs because, when rocks are ground and applied to the soil, they have a low nutrient release rate compared to soluble fertilizers, hence contributing to a residual effect on the soil for a longer period of

time (Manning; Theodoro, 2020). Thus, there were later responses to the application of basalt and phyllite dusts, justifying the differences found from the second cut.

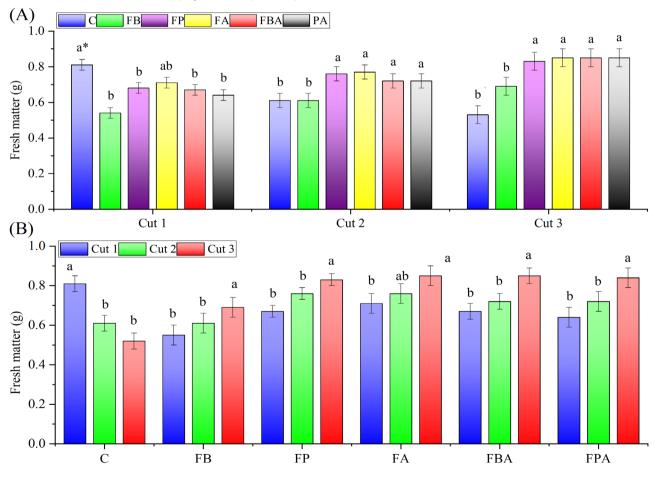
Phyllite dust contains in its composition mainly  $Al_2O_3$  and  $SiO_2$ , besides elements in the form of oxides such as  $K_2O$ ,  $Fe_2O_3$  and MgO, as well as other metals in a smaller proportion (Melo; Thaumaturgo, 2012). The mineral elements present in rocks are essential for plant growth and can be released in the form of nutrients in the soil through the solubilization of rock powders (Theodoro *et al.*, 2021). In this case, phyllite dust seems to be important during the vegetative growth of white oats, especially after the second cut.

White oat is not a crop used for grazing, but the production of fresh matter (FM) is an important aspect from the forage point of view, as its cultivation for silage production is a practice widely employed in dairy and beef cattle farming in southern Brazil (Santos *et al.*, 2021).

The data presented in Figure 3 revealed a statistical difference (P = 0.032; F = 6.574), indicating that the first cut is not adequate to evaluate the difference between the treatments due to the short time (Figure 3A). However, from the second cut onwards, the application of phyllite dust and inoculation with *Azospirillum brasilense* led to the highest values for FM. Figure 3A shows that, in the third cut, the production obtained with the application of basalt dust associated with inoculation (0.85 g) equals the production of the treatments FP (0.83 g), FPA (0.85 g) and FA (0.85 g).

The difference found between the cuts for the treatments that received the application of remineralizers and *Azospirillum brasilense*, as well as their association, reveals a positive long-term effect on forage production. Figure 3B shows that the FM production found in the control decreases, with higher production in the first cut and about 34% reduction in the forage potential in the third cut. On the other hand, for the other treatments, FM production increases the forage production potential by up to 20% in the third cut (Figure 3B).

**Figure 3** - Fresh matter (FM) of the treatments control (C), fertilizer + basalt (FB), fertilizer + phyllite (FP), fertilizer + *Azospirillum brasilense* (FA), fertilizer + basalt + *Azospirillum brasilense* (FBA), fertilizer + phyllite + *Azospirillum brasilense* (FPA) in the three cuts of the shoot of the white oat crop. \*Different letters indicate statistical difference between treatments for each cut (A) and between cuts for each treatment (B) by LSD test (P < 0.05)



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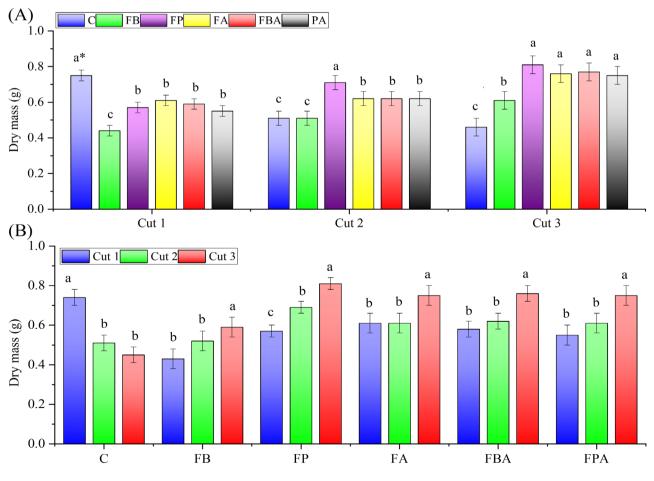
Figure 4 shows that there was a significant difference (P = 0.015; F = 9.720) in dry mass production (DM) from the first to the third cut in treatments that received the application of phyllite and inoculation (Figure 4A). In the first cut, the highest DM values were found in the control (0.75 g), while FB (0.44 g) had the lowest value. In the second cut, the use of phyllite (0.71 g) promoted higher dry mass production compared to FA (0.62 g), FBA (0.61 g) and FPA (0.62 g), with the lowest values found in C (0.51 g) and B (0.51 g). In the third cut, *Azospirillum brasilense* and its association with remineralizers [FPA (0.75 g); FBA (0.77 g)] promoted better results, differing from the use of basalt (0.61 g) and control (0.46 g), which had the lowest values.

Figure 4B shows that there was a statistical difference between the cuts for the treatments tested, and the control showed higher DM production only in

the first cut. It can be observed that dry mass production was higher in the third cut for treatments FB, FP, FA, FPA and FBA. This result can be related to FM production (Figure 3), but dry mass is more reliable when it reveals a difference between the production of the first and third cut in treatments that received the application of phyllite and basalt associated (FPA and FBA) or not with inoculation (FP and FB).

Chemical fertilizers are highly soluble sources and rapidly release large amounts of nutrients when applied to soils (Brito *et al.*, 2022), promoting high yields in the short term (Reetz, 2016). The release of nutrients from remineralizers is slow, as the weathering of the minerals that make up the rocks is a complex, heterogeneous and gradual process (White *et al.*, 2017). In view of this, the increase in FM and DM production observed in the third cut in treatments that received the application of phyllite and basalt is justified.

**Figure 4** - Dry mass (DM) of the treatments control (C), fertilizer + basalt (FB), fertilizer + phyllite (FP), fertilizer + *Azospirillum brasilense* (FA), fertilizer + basalt + *Azospirillum brasilense* (FBA), fertilizer + phyllite + *Azospirillum brasilense* (FPA) in the three cuts of the shoot of the white oat crop. \*Different letters indicate statistical difference between treatments for each cut (A) and between cuts for each treatment (B) by LSD test (P < 0.05)



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Remineralizers have a low rate of nutrient release into the soil, and reveal a low capacity for immediate responses (Manning; Theodoro, 2020). Phyllite, when compared to basalt, shows greater solubilization potential, due to its mineralogical composition (Melo; Thaumaturgo, 2012), releasing more quickly nutrients that can be assimilated by plants, reflecting on some vegetative parameters. The use of PGPR through inoculation proves to be an appropriate practice to increase the response of the use of remineralizers, validating the hypothesis that inoculation accelerates the effect of basalt and phyllite. In addition, the proposed practice promoted an increase in plant growth and phytomass production, indicating that it is an indirect response to the effect of substances produced in the rhizosphere or to the solubilization of phyllite and basalt by bacteria.

Microorganisms are able to act on minerals, through surface attacks (Sharma *et al.*, 2023), release of extracellular polymeric substances (LIU *et al.*, 2024), biofilm formation and delamination (Mustoe, 2018), solubilizing nutrients to forms that can be readily absorbed by plants (Ribeiro *et al.*, 2023). Studies have already pointed to increased yield in Poaceae such as maize (*Zea mays* L.) through inoculation with *Azospirillum brasilense* (Hungria *et al.*, 2022; Sandini *et al.*, 2024; Santos *et al.*, 2021). This shows that, despite the restricted solubility of the rocks, it is possible to obtain adequate levels of nutrition using soil remineralization as a complementary practice to soluble chemical fertilization.

The data presented so far suggest that the growth and development of white oat is favored by the use of the phyllite remineralizer and inoculation with *Azospirillum brasilense*, when compared only to the use of conventional chemical fertilization (control). In addition, the association of inoculation with the application of basalt shows more positive effects when compared to the use associated with phyllite. The statistical test revealed a significant difference in root length (RL), with treatments FB, FA and FBA promoting the highest growth of the root system of white oat, and the lowest values were obtained in treatments FP and C. For the root dry mass (RDM) variable, inoculation with *Azospirillum brasilense* associated with basalt dust (FBA) promoted the highest value, differing from FA, FB, FP, and FPA, with the lowest means found in the control (Table 2).

The highest RL values were found in treatments that received the association of remineralizers with inoculation (FBA and FPA) and in treatment FA, indicating that it was a response of inoculation with *Azospirillum brasilense*. Oliveira *et al.* (2024), when evaluating the effect of inoculation with *Azospirillum brasilense* in maize (*Zea mays* L.) seeds, obtained higher values for root length when this bacterium was present, corroborating the results obtained in the present study. *Azospirillum brasilense* is related to several plant growth benefits (Hungria *et al.*, 2022), with emphasis on the synthesis of phytohormones such as auxins, cytokinins, and gibberellins (Cassán *et al.*, 2020; Porto *et al.*, 2020). Therefore, the fact that the treatments that used inoculation showed the highest root growth is justified by a well-known characteristic of the inoculated bacterial strains.

An individual effect was observed for the RL variable in the treatment that received the application of basalt dust (FB). Basalt rocks improve the physicalchemical properties of the soil, as they increase water retention, favoring nutrient absorption and stimulating root growth (Rudmin *et al.*, 2020). The probable stimulus in the release of nutrients from this rock promoted by bacterial activity, associated with the release of root exudates with a solubilizing effect, may have released a greater amount of nutrients to the soil. In addition, basalt, when compared to phyllite, has a higher concentration of nutrients to plants, promoting greater plant nutrition (Theodoro; Almeida, 2013).

Table 2 - Root length (RL) and root dry mass (RDM) of white oat plants after three cuts

Treatments	RL	RDM
Treatments	cm	mg
Control (C)	$8.46\pm0.82~cd\texttt{*}$	$40 \pm 12.21 \text{ c*}$
Fertilizer + Basalt (FB)	$11.96 \pm 0.70$ a	$80\pm10.09~b$
Fertilizer + Phyllite (FP)	$8.18\pm0.91\ d$	$80\pm9.87~b$
Fertilizer + Azospirillum brasilense (FA)	$11.52 \pm 0.62$ a	$70 \pm 11.55b$
Fertilizer + Basalt + Azospirillum brasilense (FBA)	$11.98 \pm 0.75 \text{ ab}$	$120 \pm 15.68$ a
Fertilizer + Phyllite + Azospirillum brasilense (FPA)	$9.82 \pm 1.05 \text{ bc}$	$80\pm10.50\ b$
P-value	0.001	0.002
F-value	8.712	5.448

\* Different letters show statistical difference between treatments using the LSD test (P < 0.05)

The occurrence of shorter root length in treatments that used phyllite can be explained by two probable effects: densification of phyllite due to its smaller particle size, causing compaction and consequently less root growth; or compensation for the stress condition with greater growth of the aerial part. The RDM values indicate that the association of inoculation with phyllite has the capacity to promote benefits related to plant nutrition, since root mass is an important characteristic for increasing water and nutrient absorption. However, a more in-depth study is needed to prove the effects related to the application of phyllite dust on root growth.

#### Soil chemical attributes

The chemical attributes of the soil presented in Table 3 show that there is a statistical difference between the treatments tested for the application of remineralizers and the inoculation, as well as the single application of these inputs. It is observed that the FP contents in the soil of the FBA and FPA treatments are below the value observed in the FB treatment, indicating that the interaction of the bacterium *Azospirillum brasilense* is not positive to optimize the release of P when associated with basalt dust; however, the FP content was the same for treatments FB, C, FP and FA. On the other hand, the K contents found in the soil of the FP and FPA treatments differed significantly and reveals that the inoculation may have optimized the solubilization of this nutrient when associated with the phyllite dust.

The data presented in Table 3 indicate that there is an interaction of the bacterium *Azospirillum brasilense* with the remineralizers under study; however, some characteristics that may be related to the composition of the phyllite and basalt dusts seem to promote some questionable effects, especially in the reduction of P, Ca and Mg contents of the soil in the FBA and FPA treatments. Phyllite is characterized by being of metamorphic origin and has high levels of  $Al_2O_3$  and  $SiO_2$  in its mineralogical composition (Costa, 2021). There is a great variation in the composition of phyllites; however, in general their composition has 55 to 75% of SiO<sub>2</sub>, 2 to 6% of Fe<sub>2</sub>O<sub>3</sub>, 12 to 23% of  $Al_2O_3$ , 0.5 to 4.5% of CaO, 1 to 4% of MgO, 2 to 6% of K<sub>2</sub>O, and the P<sub>2</sub>O<sub>5</sub> content generally does not exceed 0.5% (Grimm; Behrens, 2010; Monteiro *et al.*, 2022).

Bacteria of the genus Azospirillum spp. have several mechanisms of action under different patterns and can act on the solubilization of potassium oxides ( $K_2O$ ) (Fukami *et al.*, 2016). Andrade *et al.* (2022) found increments in nutrient contents in the soil and leaf tissue of Tamani grass (*Panicum maximum* L.) in the treatments that used inoculation with *Azospirillum* spp. Therefore, the inoculated bacterial strains may have acted on the solubilization of K from the phyllite dust, which explains the effect observed in the soil of FPA.

From another perspective, basalts are igneous rocks (Costa, 2021) and generally have approximately 45 to 65% of SiO<sub>2</sub>, 8 to 15% of Fe<sub>2</sub>O<sub>3</sub>, 10 to 15% of Al<sub>2</sub>O<sub>3</sub>, 4 to 12% of CaO, 2 to 6% of MgO, and K<sub>2</sub>O and P<sub>2</sub>O<sub>5</sub> contents generally do not exceed 1 to 2%, respectively (Conceição *et al.*, 2022; Dalmora *et al.*, 2016; Nunes *et al.*, 2014).

In some specific cases with predominance of micas and feldspars in the mineralogical composition, some specific types of basalts may have higher contents of K<sub>2</sub>O, exceeding 4% (Manning *et al.*, 2017). In this context, the difference observed in the K content of treatment FP (439.77 mg dm<sup>3</sup>) compared to FB (478.40 mg dm<sup>-3</sup>) and FBA (482.10 mg dm<sup>-3</sup>) can be explained by the mineralogical composition of the rocks.

Treatments <sup>1</sup> -	Р	Κ	Ca	Mg
	mg dm <sup>-3</sup>	mg dm <sup>-3</sup>	cmol <sub>c</sub> dm <sup>-3</sup>	cmol <sub>c</sub> dm <sup>3</sup>
С	$26.21\pm0.50\ ab*$	461,80 ± 9.15 ab*	$19.40 \pm 2.10 \text{ a*}$	5.20 ± 0.21 a*
FB	$28.64\pm0.59\;a$	$478.41 \pm 10.02 \ a$	$17.91 \pm 1.85 \text{ ab}$	$5.10\pm0.15\;ab$
FP	$25.90\pm0.42\ ab$	$439.70\pm8.74\ b$	$17.75\pm2.55$ ab	$5.20\pm0.10\;ab$
FA	$26.53\pm0.36\ ab$	$462.76 \pm 11.09 \text{ ab}$	$17.70\pm1.57~ab$	$5.09\pm0.18\ b$
FBA	$24.60\pm0.48\ b$	$482.10 \pm 10.01$ a	$14.63 \pm 1.21 \text{ c}$	$4.60\pm0.10\ c$
FPA	$24.02\pm0.63\ b$	$470.11 \pm 7.70$ a	$16.81\pm2.48\ b$	$5.11\pm0.14\ ab$
F-value	8.095	3.844	6.918	3.620
P-value	0.037	0.042	0.001	0.002

Table 3 - Phosphorus (P), Potassium (K), Calcium (Ca), Magnesium (Mg) contents in the soil at the end of the experiment

<sup>1</sup>Treatments: C, control; FB, fertilizer + basalt; FP, fertilizer + phyllite; FA, fertilizer + *Azospirillum brasilense*; FBA, fertilizer + basalt + *Azospirillum brasilense*; FPA, fertilizer + phyllite + *Azospirillum brasilense*. \* Different letters show statistical difference between treatments using the LSD test (P < 0.05)

It was observed that the lowest contents of Ca and Mg were found in the soil of the FBA treatment (14.67 and 4.65 cmol<sub>a</sub> dm<sup>-3</sup>), which may be related to the lower solubilization of the basalts compared to the phyllite in short periods of evaluation. Some studies reveal significant increases in Ca and Mg contents in soils that have received the application of basalt rock dust in various situations (Aguilera et al., 2020; Almeida Junior et al., 2022; Seidel et al., 2022). However, in this case, the incubation period is insufficient to release cations in non-ionized phases, as is the case with calcium (CaO) and magnesium (MgO) oxides that may be retained in low-solubility mineral phases. In addition, the reduction of cations in the soil may be a consequence of weathering processes due to precipitation and fixation of cations in other mineral phases (Batista et al., 2018).

The K<sup>+</sup>, Ca<sup>+2</sup> and Mg<sup>+2</sup> cations are used to calculate the base saturation of the soil (BS) (Ronquim, 2010), so Ca and Mg contents may have influenced the BS values found, especially in the FBA treatment (Table 4). According to Ronquim (2010), low BS values indicate that small amounts of cations are saturating the negative charges of CEC and that most of them are being neutralized by H<sup>+</sup> and Al<sup>+3</sup>.

Table 4 shows that the Al saturation observed in the soil of the FBA treatment was higher, directly interfering with the PSA value, which is higher than that found in the soil of the FB and FA treatments. In addition, the FBA treatment had the lowest BS value, except when compared to the FPA treatment. These effects may have influenced the determination of CEC of the treatments under study, since the sum of bases is used as a multiplication factor in the calculation of CEC.

The difference in PSA between the treatments FBA, FA and FB may be related to the  $Al_2O_3$  content of basalt and phyllite dusts; however, it is not possible to state this fact due to the lack of information on the mineralogical and chemical composition of the rocks used as raw material for the remineralizers employed in this study. In their studies with bentonite, Chaves *et al.* (2019) reported an increase in potential soil acidity due to the  $Al_2O_3$  content (13.34%) present in the composition of the mineral. However, this effect seems to be related only to the initial phases of the reaction of rock dust in the soil solution due to the release of  $Al^{+3}$  by the solubilization of silicates, which over time reverse the process, reducing the potential acidity and the  $Al^{+3}$  content (Melo *et al.*, 2012).

The data related to PSA and Al indicate that the application of rock dusts with high  $Al_2O_3$  content in the composition can increase potential soil acidity in the initial phases of the reaction in the soil solution. When studying doses of basalt rock dust in association with bovine manure, Gotz *et al.* (2019) found an increase in potential acidity at the lowest doses (3 tha<sup>-1</sup>) and a reduction in this indicator with the increase in the dose (9 t ha<sup>-1</sup>), which is related to the effect of weathering on silicates suggested by Melo *et al.* (2012). This corroborates the results of the present study and explains the effects observed on PSA in the treatments that received the application of basalt powder.

In view of the results presented so far, there is a need to evaluate the dispersion of the chemical soil variables together with the plant variables, to verify their associations. In Figure 5, the principal component analysis (PCA) shows the spatial distribution of plant variables along with the chemical attributes and treatments evaluated in the cultivation of white oat. It is observed

1	Al	PSA	BS	CEC
Treatments <sup>1</sup>	cmol <sub>c</sub> dm <sup>-3</sup>	cmol <sub>c</sub> dm <sup>-3</sup>	%	cmol <sub>c</sub> dm <sup>-3</sup>
С	3.83 ± 0.38 b*	8.74 ± 0.89 ab*	74.91 ± 3.54 a*	34.60 ± 1.22 a*
FB	$4.03\pm0.40\ b$	$7.61\pm1.12~b$	$76.39 \pm 2.98 \text{ a}$	$31.93\pm0.98\ ab$
FP	$4.22\pm0.35\ b$	$8.37 \pm 1.05 \text{ ab}$	$74.16 \pm 3.50 \text{ a}$	$32.42\pm1.50~ab$
FA	$4.55\pm0.30\ b$	$7.48\pm0.84\ b$	$76.24 \pm 2.55$ a	$31.36\pm1.25~b$
FBA	$6.04 \pm 0.41$ a	$10.42\pm0.90~a$	$66.40\pm3.70\ b$	$30.98\pm1.12\ b$
FPA	$4.57\pm0.45\ b$	$9.39\pm0.75 \text{ ab}$	$71.15 \pm 3.81 \text{ ab}$	$32.57 \pm 1.09 \text{ ab}$
P-value	20.624	17.351	4.943	6.406
F-value	0.041	0.045	0.008	0.044

Table 4 - Aluminum (Al), H + Al (PSA), base saturation (BS) and cation exchange capacity at pH7.0 (CEC) in the soil of the treatments at the end of the experiment

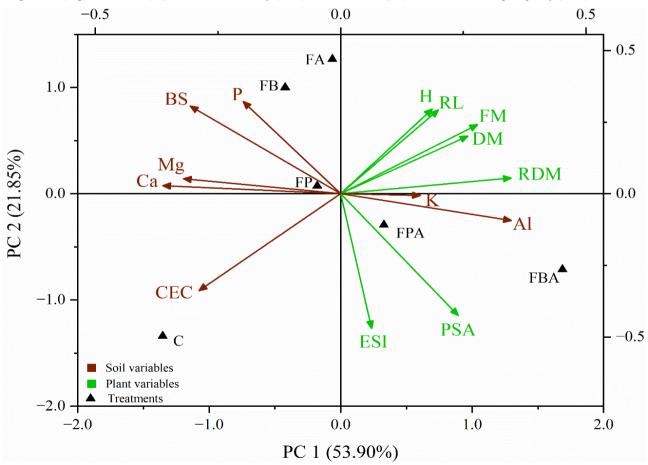
<sup>1</sup>Treatments: C, control; FB, fertilizer + basalt; FP, fertilizer + phyllite; FA, fertilizer + Azospirillum brasilense; FBA, fertilizer + basalt + Azospirillum brasilense; FPA, fertilizer + phyllite + Azospirillum brasilense. \*Different letters show statistical difference between treatments using the LSD test (P<0.05). PSA: potential soil acidity

that PC 1 explained 53.90% and PC 2 explained 21.85%, totaling 75.40% of the data variability. The variables P, BS, Mg and Ca are associated with treatments FP, FB and FA, whereas the variables H, RL, FM, DM, RDM, K, Al, PSA and ESI are related to FPA and FBA treatments.

It is worth pointing out that K and Al are the only soil variables that show correlation with the vegetative characteristics of plants through the use of phyllite and basalt associated with inoculation of *Azospirillum brasilense*. The dispersion of the data indicates that, although there was an increase in Al in CEC (Table 4), this was not a limiting factor for the vegetative development of oat plants. In addition, the approximation of the FBA and FPA treatments with K and Al reveals that inoculation with the bacterial strains Abv5 and Abv6 of *Azospirillum brasilense* can influence the solubilization of silicates and initially release K and Al for occupation of charges in the soil CEC. Therefore, PSA increases because the solubilization of silicates releases Al, and the release of bases such as Ca and Mg occurs later, as proposed by Melo *et al.* (2012). The results presented in this study corroborate what has been reported by the scientific community, pointing out the tendency of the beneficial effects of the application of remineralizers to occur in the long term, especially due to the difficulties involved in the search for alternatives that accelerate the solubilization of this input when applied to soils. In view of the above, it is worth highlighting that the dose and incubation time may have inhibited the results that could be achieved in the present study, in the evaluation of potential acidity and Al.

Another important factor from the scientific and agronomic point of view is the selection of remineralizers with known chemical and mineralogical composition or duly registered with the Ministry of Agriculture, Livestock and Food Supply (MAPA) through the current legislation. In addition, we recommend that studies with remineralizers be preferably carried out in the field with evaluation periods longer than two years, as carried out by Galina *et al.* (2024), allowing the analysis of effects on the nutritional condition of plants.

**Figure 5** - Analysis of principal components of oat cultivation through the variables Emergence speed index (ESI), Plant height (H), Fresh matter (FM), Dry mass (DM), Root length (RL), root dry mass (RDM), Phosphorus (P), Potassium (K), Calcium (Ca), Magnesium (Mg), Aluminum (Al), Potential soil acidity (PSA), base saturation (BS) and Cation exchange capacity (CEC)



## **CONCLUSIONS**

- 1. Association of phyllite and basalt dust with *Azospirillum brasilense* promotes benefits to the cultivation of white oat, improving the vegetative development of plants;
- 2. Inoculation with *Azospirillum brasilense* improves the efficiency in the use of basalt and phyllite dusts, promoting greater release of nutrients to the soil;
- 3. Phyllite dust shows advantages in improving the vegetative characteristics of white oat plants in early stages, surpassing basalt in this aspect.

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