Phonolite associated with organic compound and potassium solubilizing bacteria in tomato cultivation[1]

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ABSTRACT

The use of rock dust in Brazil has been studied and encouraged to reduce the Brazilian dependence on the foreign market for chemical fertilizers and as an alternative to organic agriculture. The objective of this research was to evaluate the effects of using phonolite rock powder and organic compound, associated with inoculation with the potassium solubilizing bacteria (KSB) of the genus *Burkholderia* on the chemical characteristics of the soil, macro and micronutrients in the aerial part, and tomato productivity. The experiment was installed in pots in a greenhouse, using a randomized block design in a 5 × 2 factorial arrangement, representing five doses of phonolite added in an organic compound, with the presence or absence of inoculation with the strain 100-13. The nutrient contents in the soil and the aerial part and the production of tomato were evaluated. Differences were observed between the doses of the organic compound and the phonolite associated with the inoculation. The treatments containing the highest doses of an organic compound provided better levels of nutrients in the soil and the aerial part, and in the tomato productivity. The inoculation with the bacterial strain improved the release of potassium from the powder of the phonolite rock.

Keywords: food safety; organic matter; rock powder; *Solanum lycopersicum* L.

INTRODUCTION

Given the perspectives related to the increase in demand for food production in the coming decades, new challenges associated with the sustainability of the agricultural sector arise, concerning environmental preservation, rational use of natural resources, food and work security, and the proper destination of waste (Macedo & Nishizaki Júnior, 2017). Of these, the use of natural resources is a concern since some are not renewable and are expected to run out in the next decades, for example, the mineral deposits used as a raw material in the manufacture of chemical fertilizers (Pantano *et al.*, 2016).

Brazil imports most of the chemical fertilizers used in agriculture, especially potassium, whose import exceeds 90% (Oliveira *et al.*, 2019). Regarding mineral fertilizers, phonolite is a rock found in the municipality of Poços de Caldas-MG, which has already been commercialized as a source of potassium for agriculture, being known as eKoSil®. Therefore, in recent years, researchers and the industrial sector have sought to reverse this scenario through the adoption of national mineral deposits as an alternative source of this nutrient (Teixeira *et al.*, 2012). Associated with the use of rock dust in agriculture, there is a process called biosolubilization that evaluates the contribution of soil microorganisms in the release of rock nutrients (Florentino *et al.*, 2017).

This biotechnology can also enable the use of rock

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dust in organic agriculture (Pereira Filho *et al.*, 2015), a sector that has shown significant growth in recent years (Costa *et al.*, 2018). Among the foods grown in the organic production system (Morales, 2019), the tomato (*Solanum lycopersicum* L.) stands out, representing one of the most consumed and cultivated vegetables in the world, including Brazil (CONAB, 2019).

In Brazil, the organic production of tomato is carried out mainly by large business groups, but it has the potential to expand for small farmers (Tivelli, 2015). However, according to Melo *et al.* (2017), it is necessary to improve organic farming techniques, involving plant genotypes, soil fertility, and pest and disease management.

Of the nutrients demanded by the tomato, potassium (K) is the most required element during the reproductive period (Fratoni *et al.*, 2016), being also important in the quality of the fruits, which is associated with the red color, resulting from the synthesis of carotenoids (Marschner, 2012). The demand for K in organic farming is mainly met by the use of organic compounds (Soldateli *et al.*, 2020), which, in addition to providing nutrients, increases the cation exchange capacity (CEC), water retention and favors the soil microbial communities (Zandonadi *et al.*, 2014). However, the content of nutrients in the organic compound can vary according to its origin (Valente *et al.*, 2009), which can influence the production and quality of the fruits.

Therefore, the use of organic compounds associated with rock dust and solubilizing microorganisms can guarantee the efficient supply of nutrients in the cultivation of organic tomatoes. Thus, the objective of this research was to evaluate the effects of using phonolite rock dust and organic compound, associated with inoculation with the potassium solubilizing bacteria (KSB) of the genus *Burkholderia* on the chemical characteristics of the soil, macro and micronutrients in the aerial part, and tomato productivity.

MATERIAL AND METHODS

Experiment installation

The experiment was installed in the olericulture and agricultural department of UNIFENAS in Alfenas-MG, from September to December 2018 in a greenhouse with the cultivar Campestre of the Santa Cruz group, using pots with a capacity of 11 dm^3 of soil. The experimental was a randomized blocks design in a 5 × 2 factorial arrangement, with five doses of organic compound (CO) and phonolite (PHON), in the following proportions: 1) 280 g CO e 0 g PHON; 2) 210 g CO e 12,5 g PHON; 3) 140 g CO e 25 g PHON; 4) 70 g CO e 37,5 g PHON; 5) 0 g CO e 50 g PHON, associated or not with the inoculation of the bacterial strain UNIFENAS 100-13, of the species *Burkholderia cenocepacia* (Andrade *et al.*, 2019; Paredes Filho *et al.*, 2020) and four repetitions.

The doses of the organic compound and phonolite were based on the requirements of the culture, being 280 g, 210 g, 140 g, 70 g, and 0 g, corresponding to 100, 75, 50, 25, and 0% of the recommended amount, respectively (Novais *et al.*, 1991). The same was adopted for phonolite, with 0 g, 12.5 g, 25 g, 37.5 g, and 50 g, equivalent, respectively, to 0, 25, 50, 75, and 100% of the recommended amount of K_2O for the tomato cultivation (Marques *et al.*, 2014).

The organic compound used, consisting of chicken manure and peat, presented the following chemical characteristics: Nitrogen = 0.5%; Moisture = 40%; Total Organic Carbon = 15%; Cation Exchange Capacity = 300 $mmol_c$ dm^{-3}; Carbon/Nitrogen ratio = 18; Potential hydrogen = 6.0, and Cation Exchange Capacity/Carbon = 20. Phonolite rock powder used in the experiment has 8% of K_2O and 25% of Si in its composition (eKoSil®). Assis *et al.* (2017) using the X-ray fluorescence technique (FRX) quantified the following elemental composition of the phonolite rock: 52,9% SiO_2; 22,4% Al_2O_3; 7,3% K_2O; 4,8% Fe_2O_3; 1,5% CaO and 0,16% MgO.

The bacterial strain UNIFENAS 100-13 isolated from the rhizosphere of Brachiaria brizantha has a proven ability to solubilize potassium in vitro (Florentino *et al.*, 2017) and the soil (Paredes Filho *et al.*, 2020). It is also capable of fix nitrogen (Silva *et al.*, 2019), solubilizes phosphorus, and produces indole acetic acid (Terra *et al.*, 2019) therefore is considered a plant growth-promoting bacterium.

For inoculation in tomato plants, this strain was grown in the FAM medium (Magalhães & Döbereiner, 1984) liquid for three days, long enough to reach the log growth phase, containing approximately 10^9 mL^{-1} cells.

The soil was classified as dystrophic red latosol (Oxisol) with clay texture, collected in the 0-20 cm layer, showing the following chemical characteristics (Santos *et al.*, 2018): pH (H_2O) = 5,2; P Mehlich = 3,7 mg dm^{-3}; K^+= 30,4 mg dm^{-3}; Ca^{2+}= 1,1 cmol dm^{-3}; Mg^{2+}= 0,2 cmol dm^{-3}; Al^{3+}= 0,2 cmol dm^{-3}; H+Al= 3,6 cmol dm^{-3}; Sum of Bases (SB) = 1,4 cmol dm^{-3}; T = 5,0 cmol dm^{-3}; V (Base saturation) (%) = 27,8; Organic Matter (O.M.) = 1,42 dag kg^{-1}; P residual =

18,5 mg L^{-1}; Zn= 1,0 mg dm^{-3}; Fe= 53,6 mg dm^{-3}; Mn= 7,6 mg dm^{-3}; Cu= 1,4 mg dm^{-3}; B= 0,07 mg dm^{-3}; S= 13,2 mg dm^{-3}.

The soil was homogenized and sieved with a 4 mm mesh. Then the soil was corrected with dolomitic lime according to the method of increasing the base saturation to 70% (Ribeiro *et al.*, 1999) and incubation for 60 days. Before planting the seedlings, 100 mg dm^{-3} of urea (45% N) and 300 mg dm^{-3} of simple superphosphate (18% P_2O_5) were applied per pot, according to the recommendations of Novais *et al.* (1991). Nitrogen was divided into three cover applications, every 15 days, totaling 45 days (227 mg dm^{-3} of urea for each application) (Van Genuchten, 1980).

Subsequently, according to the treatments described above, there was the addition of the organic compound and phonolite and the transplantation of a seedling per pot. The humidity was kept at 80% of the field capacity, and the openings at the bottom of the pots were sealed to avoid loss of nutrients after irrigation.

The inoculation with the bacterial strain was one week after the transplant, being inoculated two mL per plant. The plants were tutored and conducted on a single stem, thus carrying out all cultural and phytosanitary treatments. After 90 days, the fruits, the aerial part, and the soil were collected and taken to the laboratory for analysis and data collection.

The levels of nutrients in the soil and the aerial part of the plants were examined, as well as the parameters of development and production of the tomato in dry mass of the aerial part (DMAP), the number of fruits (NF), fresh fruit mass (FFM), and dry fruit mass (DFM). In the aerial part, the following nutrients were analyzed: nitrogen (N, g kg^{-1}), phosphorus (P, g kg^{-1}), potassium (K, g kg^{-1}), calcium (Ca, g kg^{-1}), magnesium (Mg, g kg^{-1}), sulfur (S, g kg^{-1}), boron (B, mg kg^{-1}), copper (Cu, mg kg^{-1}), iron (Fe, mg kg^{-1}), manganese (Mn, mg kg^{-1}) and zinc (Zn, mg kg^{-1}). The soil contained in each pot was collected and taken to the laboratory for fertility analysis, with the following parameters being analyzed: Organic Matter (O.M., g dm^{-3}), pH ($CaCl_2$), potencial acidity (H + Al, $mmol_c$ dm^{-3}), P (mg dm^{-3}); K (mg dm^{-3}), Ca ($mmol_c$ dm^{-3}), Mg ($mmol_c$ dm^{-3}), sum of bases (SB, $mmol_c$ dm^{-3}), cation exchange capacity (CEC, $mmol_c$ dm^{-3}), base saturation (V, %), B (mg dm^{-3}), Cu (mg dm^{-3}), Fe (mg dm^{-3}), Mn (mg dm^{-3}), Zn (mg dm^{-3}) and electric conductivity (EC, dS m^{-1}).

The data referring to the nutrient content in the soil and the aerial part, dry mass of the aerial part (DMAP), number of fruits (NF), fresh mass (FFM) and dry (DFM) of the fruits were submitted to analysis of variance, using the Sisvar statistical analysis program, version 5.3 (Ferreira, 2011) and the treatment averages were compared using the Scott-Knott test at 5% probability.

RESULTS AND DISCUSSION

According to the results, it was possible to observe a difference between the treatments containing the different doses of organic compound and phonolite associated with inoculation with the bacterial strain 100-13, for the nutrients contents in the soil, aerial part, development, and production parameters of the tomato.

The potassium contents in the soil and aerial part of the tomato are in Table 01. For both parameters, there was an interaction of the different doses of the organic compound and phonolite with the inoculation of strain 100-13.

In soil, potassium contents were influenced by the presence of the highest doses of the organic compound or phonolite and association of inoculation with strain 100-13. In the absence of inoculation, the highest levels of this nutrient were observed for treatments 1 and 2, containing the highest doses of organic compost, 280g CO and 0 PHON, and 210g CO and 12.5g PHON. When the bacteria were inoculated, the highest levels of this nutrient were observed in treatments containing the highest dose of the organic compound, treatment 1 (280g CO and 0g PHON), and highest dose of phonolite, treatment 5 (0g CO and 50g PHON), respectively. This result highlights the performance of the potassium solubilizing strain, 100-13, corroborating the results obtained by Paredes Filho *et al.* (2020).

The organic compound constitutes an important source of potassium in the soil, which, since it is not part of any stable organic compost, its release is independent of the performance of microorganisms (Souza & Alcântara, 2008), justifying the results obtained for treatments in which higher doses of the organic compound were applied.

In the aerial part, the highest K value was for treatment 3, with intermediate doses of the organic compound and phonolite (140 g CO + 25 g PHON). This response can be related to the balance in nutrient concentrations of the organic compound and phonolite, which, besides potassium, has other nutrients (Teixeira *et al.*, 2012).

Table 1: Potassium (K) contents in the soil and the aerial part of tomatoes grown under different doses of the organic compound (CO) and phonolite (PHON), associated with inoculation with bacterial strain 100-13

Treataments*	K in the soil (mg dm^{-3})		K in the aerial part (g Kg^{-1})	
	With inoculation	Without inoculation	With incoculation	Without inoculation
1	33,15 Aa	29,25 Ab	11,55 Cb	14,05 Ca
2	25,35 Ba	25,35 Aa	18,65 Bb	20,55 Ba
3	21,45 Ba	17,55 Ba	21,95 Aa	21,25 Aa
4	21,45 Ba	13,65 Bb	12,05 Ca	10,35 Db
5	29,25 Aa	21,45 Bb	11,95 Ca	10,85 Db

* 1 = 280g CO + 0g PHON; 2 = 210g CO + 12,5g PHON; 3 = 140g CO + 25g PHON; 4 = 70g CO + 37,5g PHON; 5 = 0g CO + 50g PHON. Means followed by different letters, uppercase in the column and lowercase in the row, for each variable, differ by the Scott Knott test at 5% probability.

Regarding the other nutrients evaluated in the soil and the aerial part of the tomato plants, it was found that the treatments constituted by the different doses of organic compound and phonolite associated with inoculation with bacterial strain 100-13 were significant for most parameters, except for the pH value in the soil. Tables 2 and 3 show the minimum and maximum values found for the parameters analyzed in the soil and the aerial part after tomato cultivation, respectively, as well as the occurrence of bacterial inoculation contribution in the highlighted treatment.

Table 2: Parameters analyzed, minimum and maximum contents, highlighted treatment, interaction between organic compound and phonolite (T) *versus* bacterial strain inoculation (I), and contribution of inoculation found in the soil after tomato cultivation using different doses of the organic compound and phonolite associated with the strain 100-13

Parameters analyzed	Minimum and maximum contents	T*I (Pr > Fc)	Highlighted treatment**	Inoculation contribution
Phosphorus	5,1–40,8 (g kg^{-1})	Yes	2	No
Potassium	13,65–33,15 (g kg^{-1})	Yes	1	Yes
Calcium	14,95–94,95 (g kg^{-1})	Yes	2	No
Magnesium	2,95–9,95 (g kg^{-1})	Yes	1	Yes
Iron	5,95–20,95 (mg kg^{-1})	Yes	5	Yes
Manganese	2,85–6,35 (mg kg^{-1})	Yes	2	No
Copper	1,15–2,95 (mg kg^{-1})	Yes	2	No
Zinc	4,25–13,45 (mg kg^{-1})	Yes	5	Yes
Boron	0,46–1,075 (mg kg^{-1})	Yes	5	Yes
O.M.	13,95–23,95 (g dm^{-3})	Yes	2	No
pH	4,85–5,35	No	1	-
H+Al	37,95–72,95 ($mmol_c$ dm^{-3})	Yes	2	No
SB	25,85–101,65 ($mmol_c$ dm^{-3})	Yes	2	No
CEC	63,85–174,65($mmol_c$ dm^{-3})	Yes	2	No
V	40,95–69,95 (%)	Yes	5	No
EC	0,45–2,15 (dS m^{-1})	Yes	5	No

* 1 = 280g CO + 0g PHON; 2 = 210g CO + 12,5g PHON; 3 = 140g CO + 25g PHON; 4 = 70g CO + 37,5g PHON; 5 = 0g CO + 50g PHON.
** Treatment that gave a higher content of nutrients due to the doses of CO e PHON.

Table 3: Minimum and maximum nutrient content, highlighted treatment, interaction between organic compound and phonolite (T) *versus* inoculation with bacterial strain (I) and contribution of inoculation in the aerial part of tomato plants grown with different doses of organic compound and phonolite associated with the bacterial strain 100-13

Parameters analyzed	Minimum and maximum nutrient content	T*I (Pr > Fc)	Highlighted treatment**	Inoculation contribution
Nitrogen	28,15 - 43,65 (g kg^{-1})	Yes	2	No
Phosphorus	0,95 - 6,85 (g kg^{-1})	Yes	3	No
Potassium	11,20 – 21,60 (g kg^{-1})	Yes	3	No
Calcium	1,85 – 13,76 (g kg^{-1})	Yes	1	Yes
Magnesium	0,95 – 5,25 (g kg^{-1})	Yes	1	Yes
Sulfur	1,25 - 4,75 (g kg^{-1})	Yes	1	Yes
Iron	44,05 – 286,45 (mg kg^{-1})	Yes	1	No
Manganese	60,25 – 144,25 (mg kg^{-1})	Yes	1	No
Copper	2,65 – 9,40 (mg kg^{-1})	Yes	2	No
Zinc	14,30 – 68-15 (mg kg^{-1})	No	1	-
Boron	6,25 – 43,95 (mg kg^{-1})	Yes	3	No

* 1 = 280g CO + 0g PHON; 2 = 210g CO + 12,5g PHON; 3 = 140g CO + 25g PHON; 4 = 70g CO + 37,5g PHON; 5 = 0g CO + 50g PHON.
** Treatment that gave a higher content of nutrients due to the doses of CO e PHON.

It is observed that the organic compost when used in higher doses, in treatments 1 and 2, contributed to the improvement of the chemical characteristics of the soil, except for the values of iron, zinc, boron, base saturation, and electrical conductivity, in which the highest values were obtained for treatments containing the highest dose of phonolite and the lowest dose of the organic compound, identified as treatment 5 (Table 2).

Regarding bacterial inoculation, its contribution to the parameters of potassium, magnesium, iron, zinc, and boron is verified, which can be attributed to the solubilization of the rock by strain 100-13, since most of these elements are phonolite's constituents (Teixeira *et al.*, 2012).

The treatments containing the highest doses of the organic compound stood out, providing the highest contents of nutrients in the aerial part, indicating the importance of using the organic compound for the supply of nutrients to the plant (Table 3).

Evaluating the data in Table 3, for some elements such as phosphorus (Nishimoto *et al.*, 1977; Silva *et al.*, 2001), magnesium (Furlani *et al.*, 1978; Silva *et al.*, 2001), and iron (Bataglia, 1988; Silva *et al.*, 2001), their contents are in the range recommended for tomato culture. While potassium (Furlani *et al.*, 1978), calcium, sulfur (Takahashi, 1989), copper, and zinc (Silva *et al.*, 2001), are below the recommended range for productive tomato plants. The analysis of the nutrient content of the aerial part of the crops is important since it indicates the productive potential (Kurihara *et al.*, 2013).

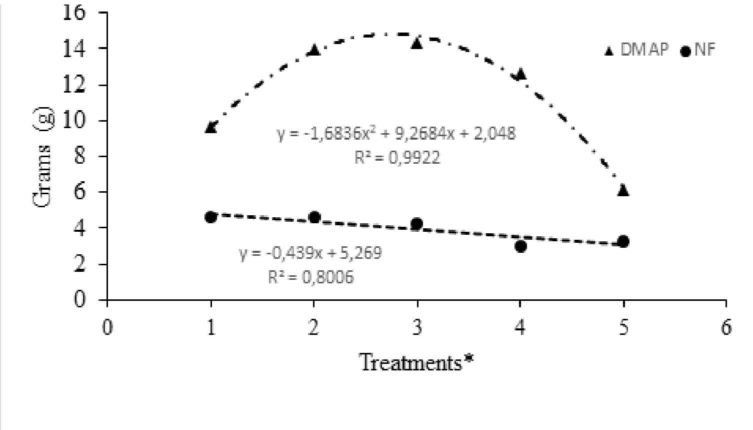
It can also be seen that, despite the occurrence of the interaction between the organic compound and phonolite versus inoculation, there was no contribution to the higher nutrient content of the aerial part. In a study developed by Golinski *et al.* (2014), the feasibility of using basaltic rock powder for carrot yield was reported. Fruet *et al.* (2019) did not observe any effect of the use of slate rock powder in the production of beet, requiring further studies on the interaction of the various types of rock powder in the cultivation of vegetables.

The production of organic acids has been considered the main mechanism of potassium solubilization by bacteria, as discussed in detail in the review by Meena *et al.* (2014).

The strain tested in this study was isolated from the rhizosphere of *Brachiaria brizantha* and had a proven ability to fix N_2 and to contribute to the plant development of Mombaça grass (Silva *et al.*, 2019). It also tolerates high salinity concentrations in the culture medium, produces indole-3-acetic acid, and solubilizes phosphorus (Terra *et al.*, 2019) and potassium (Florentino *et al.*, 2017). Therefore, these diverse functional characteristics of this strain may explain the good results promoted in the dry mass of fruits and levels of some nutrients in the plants.

Regarding the development and production of tomato grown with different doses of the organic compound and phonolite (CO and PHON) associated with the strain 100-13, a significant effect of the doses of CO and PHON was found for every parameter: the dry mass of the aerial part (DMAP), number of fruits (NF), fresh fruit mass (FFM), and dry fruit mass (DFM). The interaction between doses of CO and PHON and inoculation was verified for DFM.

Generally, for all these parameters, the best results are related to the presence of the highest doses of the organic compound. The DMAP (g) and NF values are shown in Figure 1.



* 1 = 280g CO + 0g PHON; 2 = 210g CO + 12,5g PHON; 3 = 140g CO + 25g PHON; 4 = 70g CO + 37,5g PHON; 5 = 0g CO + 50g PHON.

Figure 1: Values of dry mass of the aerial part (DMAP) (g) and number of fruits (NF) of the tomato when cultivated with different doses of organic compound associated to the phonolite.

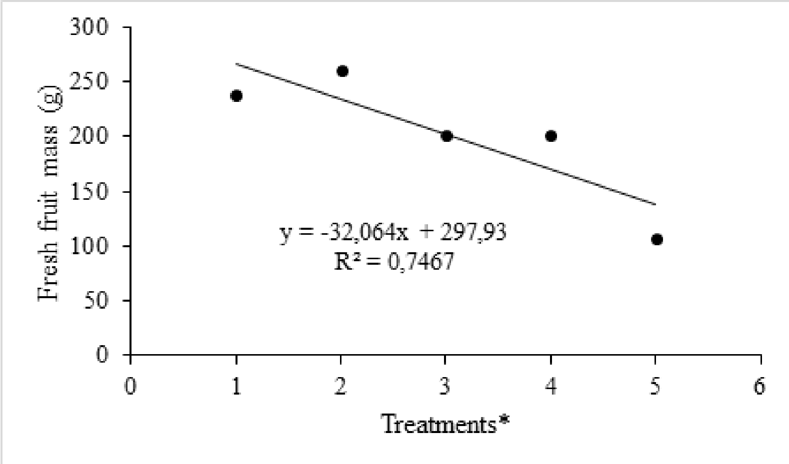
It is observed that the highest DMAP value was in treatment 3 with a dose of 140g CO + 25g PHON, indicating benefits in the association between these two compounds. Phonolite, besides potassium, has other nutrients (Teixeira *et al.*, 2012), which can also contribute to the vegetative development of tomatoes. However, for the NF, it was found that there was a decrease in the values of this parameter as it reduced the doses of organic compost, evidencing the importance of this input for tomato productivity (Costa *et al.*, 2018).

Regarding the fresh fruit mass (FFM), a behavior similar to that observed for the NF was verified, with a reduction in the FFM as the contents of organic compound decreased, and the contents of the phonolite rock dust increased (Figure 2).

In Table 4 are the values of dry fruit mass (DFM), showing the occurrence of interaction between the doses of the organic compound and phonolite and inoculation with strain 100-13.

It appears that the highest values of DFM were for the dose 210g CO + 12.5g PHON, and the inoculation was not efficient for this treatment. The results show that the strain 100-13 contributed to the highest values of DFM in treatments with doses of 140 and 70 g of the organic compound. These can happen because the survival of a given microbial group in the rhizosphere is dependent on the supply of nutrients to the plant, which directly influences the release of compounds that will be used by microbial metabolism (Souto *et al.*, 2008). Thus, plants with satisfactory nutrient availability can release different compounds, therefore not stimulating the performance of these microorganisms in their rhizosphere (Cardoso & Nogueira, 2007). Hence, it appears that high doses of organic matter, although beneficial for plant development and fresh fruits, can interfere with the performance of bacterial strain 100-13.

Bacteria of the *Burkholderia* genus have a great diversity of species, isolated from different habitats, such as soil, water, effluents, and rhizosphere, including human, animal, and plant pathogens. Recently, research has demonstrated the great potential of bacteria of this genus to act as plant growth promoters (Paganin *et al.*, 2011; You *et al.*, 2020; Jha & Popat, 2021). However, due to the pathogenicity of some species, the use in agriculture is still limited, requiring further studies.



* 1 = 280g CO + 0g PHON; 2 = 210g CO + 12,5g PHON; 3 = 140g CO + 25g PHON; 4 = 70g CO + 37,5g PHON; 5 = 0g CO + 50g PHON.

Figure 2: Values of fresh fruit mass (FFM) of tomato when cultivated with different doses of organic compound associated with phonolite.

Table 4: Values of dry fruit mass of tomato (DFM) grown with different doses of organic compound and phonolite associated or not with bacterial strain 100-13

Treatments*	With Inoculation	Without Inoculation
1	48,97 Ba	41,47 Bb
2	61,96 Ab	66,46 Aa
3	30,40 Ca	27,29 Cb
4	29,14 Ca	25,92 Db
5	16,45 Db	20,19 Ea

* 1 = 280g CO + 0g PHON; 2 = 210g CO + 12,5g PHON; 3 = 140g CO + 25g PHON; 4 = 70g CO + 37,5g PHON; 5 = 0g CO + 50g PHON. Means followed by different letters, uppercase in the column and lowercase in the row, for each variable, differ by the Scott Knott test at 5% probability.

You *et al.* (2020) agree that studies are needed to identify the risks to human health associated with *B. cenocepacia* inoculants in agriculture. These authors also emphasize that the soil naturally contains several potentially pathogenic microorganisms, which can be increased by commonly used agricultural techniques, such as manure and irrigation with wastewater. Therefore, it is necessary to compare the benefits and disadvantages of new technologies with practices that are known to impact soil biodiversity, for instance, the use of synthetic fertilizers.

Based on the growth and production parameters, the importance of using the organic compound associated with phonolite in tomato production is verified. Considering that the studies of this rock dust are still incipient for most cultures, these data have significant relevance, being able to direct new studies and works in different areas, such as in basic and applied research, and the extension of this information and practices to the producer.

CONCLUSIONS

Phonolite can be an alternative source of potassium for tomatoes, but the use of organic compound is essential for the supply of most nutrients.

Inoculation with bacterial strain 100-13 contributed to the greater release of potassium from the powder of the phonolite rock.

Treatment with 210g of the organic compound and 12.5g of phonolite conferred a higher fruit fresh and dry mass of tomato.

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