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# FRACTIONATION OF INORGANIC PHOSPHORUS AS A FUNCTION OF THE APPLICATION METHOD OF PHOSPHATE AND BIOMAPHOS® SOLUBILIZER

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**ABSTRACT** - The mode of application, the source of P applied and the solubilizers alter the dynamics of P. The fractionation of P can help in understanding. The objective of this work was to evaluate the response of cultures and alterations in the forms of P as a result of the modes of P application and use of the Biomaphos® solubilizer. The design was randomized blocks in split plots: plot-application mode (100% P broadcast on the surface, 100% in the sowing furrow, ½ broadcast and ½ in the furrow, without P); subplot—with and without Biomaphos®), with seven replicates. The soil under study was the Dystroferric Latosol Red. For soybean and bean crops, productivity (kg ha<sup>-1</sup>), P content in the grain and leaf (g kg<sup>-1</sup>) and, in the soil, the fractionation of inorganic P - Pi<sub>H2O</sub>, Pi<sub>Al</sub>, Pi<sub>Fe</sub>, Pi<sub>Ca</sub> - (mg kg<sup>-1</sup>), were evaluated. Only the application method of phosphate fertilizer changed productivity. The use of Biomaphos® promoted a positive effect on P content in leaves, for 100% of phosphate fertilizer in the row in both crops and, ½ the row + ½ broadcast for beans. The lowest averages of P content in grains occurred in the treatment without P. In beans, without addition of P, there was an increase in the Pi<sub>Fe</sub> fraction and a decrease in Pi<sub>Ca</sub>, influenced by the use of Biomaphos®. The application methods and the use of Biomaphos® increased productivity, by one bag per hectare in soybean and two in beans.

Keywords: inorganic phosphorus, labile phosphorus, phosphorus fractions.

# FRACIONAMENTO DE FÓSFORO INORGÂNICO EM FUNÇÃO DO MODO DE APLICAÇÃO DE FOSFATADO E SOLUBILIZADOR BIOMAPHOS®

RESUMO - O modo de aplicação, o fracionamento, a fonte de fósforo (P) aplicada e os solubilizadores podem alterar a dinâmica do P. O objetivo do trabalho foi avaliar a resposta das culturas e alterações nas formas de P inorgânico, em decorrência dos modos de aplicação do P e uso do solubilizador Biomaphos®. O delineamento experimental foi blocos casualizados, em parcelas subdivididas, sendo a parcela, o modo de aplicação do P (100% a lanço, em superfície, 100% no sulco de semeadura, ½ a lanço e ½ no sulco e a testemunha, sem aplicação de P) e a subparcela, com Biomaphos® e sem), contendo sete repetições. O solo em estudo foi o Latossolo Vermelho Distroférrico. Para as culturas de soja e feijão, avaliaram-se a produtividade (kg ha<sup>-1</sup>), teor de P no grão e na folha (g kg<sup>-1</sup>) e, no solo, o fracionamento de P inorgânico - Pi<sub>H2O</sub>, Pi<sub>Al</sub>, Pi<sub>Fe</sub> e Pi<sub>Ca</sub>. (mg kg<sup>-1</sup>). Apenas o modo de aplicação do fertilizante fosfatado alterou a produtividade. O uso do Biomaphos® promoveu efeito positivo no teor de P nas folhas, para 100% do fertilizante fosfatado no sulco em ambas as culturas e, ½ sulco + ½ à lanço para o feijão. As menores médias dos teores de P nos grãos ocorreram no tratamento sem P. No feijão, sem adição de P, houve aumento da fraçãoPi<sub>Fe</sub> e diminuição da Pi<sub>Ca</sub>, influenciadas pelo Biomaphos®. Os modos de aplicação e o uso do Biomaphos® incrementaram a produtividade, em uma saca por hectare na soja e duas no feijão.

Palavras-chave: fósforo inorgânico, fósforo lábil, frações de fósforo.

### INTRODUCTION

Phosphorus (P) is an important macronutrient, essential for the development and growth of plant species (REED et al., 2015). In the soil, it occurs in organic (Po) and inorganic (Pi) forms, with different degrees of lability (SANTOS et al., 2022). Pi is present in primary minerals and/or adsorbed in silicate clays, in the hydroxyls of aluminum and iron oxyhydroxides and in soil organic matter by cation bridges (SANTOS et al., 2008). Po is strongly bound to organic molecules, and both can be in labile, moderately labile and non-labile forms (PAVINATO et al., 2009).

To better understand the dynamics of this nutrient in the soil, it is necessary to know the concentrations in the different fractions, using a methodology with sequential extractors (CHANG; JACKSON, 1957), using a soil sample subjected to several selective reagents for the P forms, based on the binding energy and nature of the compound. This methodology can provide important information about the availability of the nutrient in the soil (GATIBONI et al., 2013).

Several factors can be linked to the availability of P and changes in the fractions, such as the cultivation

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system (PAVINATO et al., 2009), addition of phosphate fertilizer, soil type (SANTOS et al., 2022) and biological activity of the soil. In the case of the no-tillage system, there is an increase in labile forms of P (ANDRADE et al., 2003), due to the increase in Po, through the increase in organic matter and addition of fertilizers, helping to replenish P (REDDY et al., 2000). Santos et al. (2008) and Nunes et al. (2020) reported that different methods of applying phosphate fertilizers caused changes in the P fractions in the soil.

Microorganisms play a fundamental role in the transformation of P, either by releasing organic acids, solubilizing or mineralizing P, thus increasing the superficial root area (OGUT et al., 2011) or by the P content in the microbial biomass. Positive results were found with the use of P solubilizers, which convert unavailable P into soluble P, as is the case with Biomaphos®, based on bacteria of the genus *Bacillus* (*B. subtilis* and *B. megaterium*), capable of increasing the levels of soluble P and its absorption by the crop (EMBRAPA, 2019).

In view of the above, the objective was to evaluate changes in the fractions of inorganic P in the soil adhered to the roots of soybeans and beans, depending on the method of P application, in a direct planting system in a sequence of crops.

## MATERIAL AND METHODS

The experiment was conducted at Fazenda Santa Maria, in the municipality of Santa Terezinha de Itaipu (Paraná), under geographic coordinates 25°28'09" S and 54°22'41" W and altitude of 292 me. The climate of the region is classified as subtropical, with an average annual temperature ranging from 21°C to 22°C with a minimum of 17°C and a maximum of 28°C, summers considered hot, infrequent frosts, with summer rains and average annual precipitation ranging from 1600 to 1800 mm, with no defined dry season and average relative humidity between 75 and 80%. The soil of the region under study is classified as Dystroferric Red Latosol (SANTOS et al., 2014), with 746 g kg<sup>-1</sup> of clay, 199 g kg<sup>-1</sup> of silt and 55 g kg<sup>-1</sup> of sand.

After fallowing and correction and addition of gypsum, a new soil analysis was carried out in the 0-0,20 m layer for chemical analysis, which showed pH ( $H_2O$ ) = 6,23; organic matter = 27,52 g kg<sup>-1</sup>; P = 10,10 mg dm<sup>-3</sup>, S = 2,18 mg dm<sup>-3</sup>; Ca, Mg, K, Al and H+Al, respectively, 6,95; 1,82; 0,85; 0,0 and 4,28 cmolc dm<sup>-3</sup>, sum of bases = 9,62 cmolc dm<sup>-3</sup>, CTC = 13,90 cmolc dm<sup>-3</sup>, V% = 69,21; %Ca, %Mg and %K, respectively, 50; 13 and 6,12. The soil has a high P

content, measured by the Mehlich<sup>-1</sup> extractor (SBSC/NEPAR, 2017).

The conduction area was in a Direct Planting System with Crop-Livestock Integration for 14 years, and in the last 5 years soybean and corn were sown, both in winter and summer. In 2015, the P content in the soil was 5,4 mg dm<sup>-3</sup>, considered low, by the Mehlich-1 extractor (SBSC, 2017). The plots were installed with the application methods, correcting the P content in the soil and exporting it by the crops inserted from October 2015 to March 2017, with soybean and corn in the summer, increasing the soil content to 10,10 mg dm<sup>-3</sup>, considered very high for the same extractor (SBSC/NEPAR, 2017), using a source based on simple superphosphate and monoammonium phosphate.

In April 2017, 4 Mg ha<sup>-1</sup> of calcitic limestone (PRNT 90%) was applied and after plastering, at a dose of 0,25 Mg ha<sup>-1</sup>. Brachiaria (*Brachiaria brizantha* cv. marandu) was cultivated from November/2017 to August/2019), soybean (from November/2019 to March/2020), winter crop mix (Japanese turnip, forage turnip, white oats, black oats and rye, from May/2020 to September/2020), beans (from October/2020 to February/2021) and millet (from October/2021 to March/2022).

The experimental design used was randomized blocks, in subdivided plots, where the plot consisted of 4 P application modes (100% broadcast, on the surface, 100% in the sowing furrow, ½ broadcast, on the surface + ½ in the furrow and the control, without P application) and the subplot, consisting of Biomaphos® and without), with 7 replicates and 50 m² plots. Figure 1 shows the meteorological data from 2017 to 2022, according to INMET.

Table shows the data on fertilizers/conditioners, doses and formulations of N, P<sub>2</sub>O<sub>5</sub>, K<sub>2</sub>O, S and CaO applied in each growing season, two years before the experiment was set up and during it. Brachiaria seeds (33 kg ha<sup>-1</sup>) were manually planted in the plots, incorporating them with a harrow at 2 opening points to minimize soil disturbance. Afterwards, 4 cuts of its green biomass were made, using a blinding machine. For soybeans and beans, a seeder/fertilizer with 11 rows and 0,45 m spacing between rows was used, dividing each side into 5 rows, with 1 central zero, in order to analyze the treatments with and without Biomaphos<sup>®</sup>. The direction of the seeder was changed according to the subplot draw. For soybeans, the cultivar M5947 IPRO was used, with 12 seeds per meter, and for beans, the cultivar IPR Sabiá, with 16 seeds per meter.

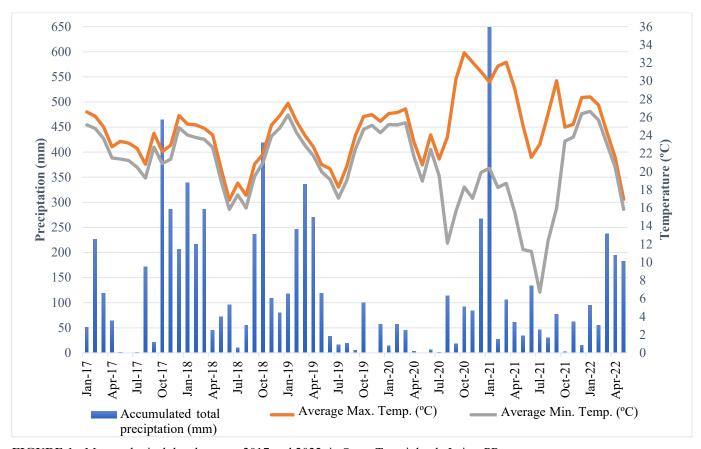


FIGURE 1 - Meteorological data between 2017 and 2022, in Santa Terezinha de Itaipu-PR.

**TABLE 1** - Summary of fertilizers and correctives, doses and formulations of N, P<sub>2</sub>O<sub>5</sub>, K<sub>2</sub>O, S and CaO, applied in each growing period.

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	System/culture	Dose (kg ha <sup>-1</sup> ) N-P <sub>2</sub> O <sub>5</sub> -K <sub>2</sub> O-S-CaO	Formulations N-P <sub>2</sub> O <sub>5</sub> -K <sub>2</sub> O-S-CaO	Information
2000-2014	Crop/livestock integration			
2015	Soybean	12-86-107-00	03-21-00	MAP+SSP
2013	Soybean	12-80-107-00	00-00-60	KCl
			20-00-00-24	NH <sub>4</sub> SO <sub>4</sub>
2016	Corn (summer)	236-86-223-110-126	00-19-00-12-28	SSP
	, in the second of the second		00-00-60-00	KCl
		4 Mg ha <sup>-1</sup>	Calcitic limestone	45% CaO
		•		5%MgO
2017	Fallow			90% PRNT
				95%PN
		0.25 Mg ha <sup>-1</sup>	Plaster	10% S 8%CaO
2017/18	Brachiaria			
2018/19	Brachiaria			
2019/20	Soybean	10-80-26-00	4-30-10-00	
2020	Winter mix			
2020/21	D	11-81-23-00	4-28-08-00	
	Beans	59-00-00-68	21-00-00-24	$(NH_4)_2SO_4$
2021	Millet	-	-	-

Biomaphos® inoculation was 150 mL ha<sup>-1</sup> in seed treatment at the time of sowing, according to the manufacturer's recommendations. Inoculation and fertilization occurred only for soybean and bean crops, at the recommended dose for P and K export (SBCS, 2017).

Broadcast fertilization, both partial and total, was performed manually in order to avoid heterogeneity in distribution. 268 kg ha<sup>-1</sup> of 4-30-10 (N-P<sub>2</sub>O<sub>5</sub>-K<sub>2</sub>O) was used in soybean and, for beans, 289 kg ha<sup>-1</sup> of 04-28-08 (N-P<sub>2</sub>O<sub>5</sub>-K<sub>2</sub>O), with 285 kg ha<sup>-1</sup> of ammonium sulfate, in

the formulation with 21% N, as topdressing, in a single plot at 40 DAP.

For the winter mix, a wheat/cover seeder was used, using 50 kg ha<sup>-1</sup> of the RX 210 mix. When the maximum dry matter peak was reached, in pre-flowering, desiccation was carried out, with 4 L ha<sup>-1</sup> of glyphosate (48%). After 20 days, cutting occurred using a knife roller. For millet, 25 kg ha<sup>-1</sup> of the ADR300 variety was sown according to the Brachiaria crop, with cutting using the blind machine in pre-flowering. The necessary cultural treatments were carried out in all crops, and presowing desiccation was also performed to ensure that this occurred without weeds.

The productivity (kg ha<sup>-1</sup>) of the crops was evaluated, with a harvest of 5,4 m<sup>2</sup> of useful area. For soybeans and beans, they were harvested at the R.8 stage and the moisture content was corrected to 13% (SOUZA et al., 2013), removing the effect of the number of plants, taking 5 measurements of 1 linear m, obtaining the average of the stand. For all crops, the residue from the harvest and cutting straw was spread in the experimental subplots (BARBOSA et al., 2015).

For the analysis of inorganic phosphorus fractionation (Pi), samples of soil adhered to the roots were collected from 5 random plants per subplot, in soybean and bean crops, at the R.1 stage, in order to evaluate the changes in Pi caused by use of Biomaphos® and method of applying phosphate fertilizer. After collection, the samples were sent to the Soil Fertility and Plant Mineral Nutrition Laboratory at Unioeste, where

they were dried in an oven for 24 h at 40°C and, after drying, they were ground using a mill with a 2 mm mesh sieve (ADFS - air-dried fine soil).

The methodology used to quantify  $PiH_2O$  (NH<sub>4</sub>Cl), PiAI (NH<sub>4</sub>F), PiFe (NaOH), PiCa (H<sub>2</sub>SO<sub>4</sub>) and total Pi (sum of the others) was according to Chang and Jackson (1957) and in the supernatants, by UV/Vis spectrophotometry (BRAGA; DEFELIPO, 1974), with results expressed in mg kg<sup>-1</sup>.

The data were tabulated and the normality and homogeneity of variances test was applied. When normal, statistical analysis was performed in a split-plot scheme. The F test was performed and, in case of significance, the Tukey test was performed, at a 5% probability of error, using the Genes computational application (CRUZ, 2013).

#### RESULTS AND DISCUSSION

The averages for soybean and bean productivity are presented in Table 2, verifying that there was no interaction between the method of application of phosphate fertilizer and the use of Biomaphos<sup>®</sup>. Only for the mode of application, the treatment without P obtained a significant difference, where those that received P obtained a minimum difference of 22 and 9,6 bags per hectare more, for soybeans and beans, respectively. This reinforces the importance of P, essential for the growth and development of crops (REED et al., 2015), for root formation and consequently, for increasing productivity (ROSSI et al., 2018).

TABLE 2 - Soybean and bean productivity, according to application methods and BiomaPhos® solubilizer.

P application modes	Soybean productivity (kg ha <sup>-1</sup> )	Bean productivity (kg ha <sup>-1</sup> )
100% broadcast on the surface	4188.14 a*	2333.32 a
$\frac{1}{2}$ broadcast + $\frac{1}{2}$ in the furrow	4209.00 a	2496.03 a
100% in the sowing furrow	4149.00 a	2541.66 a
Witness (without P)	2812.28 b	1756.94 b
BiomaPhos®		
With application	3870.75 a	2341.43 a
No application	3808.46 b	2222.55 b
CV1(%)	10.18	15.28
CV2(%)	2.27	7.67

<sup>\*</sup>Means followed by the same lowercase letter in the column do not differ from each other by the Tukey test, at a 5% probability of error.

For the application methods that received P, no significant difference was observed in productivity. In soybean cultivation in 2017, a recurrence of the results was noted for the treatments that received P, that is, a higher average obtained with fertilization with P½ in the broadcast on the surface +½ in te furrow and a lower average with 100% in the sowing furrow, with no statistical difference between the application methods. This same result was verified by other authors (NUNES et al., 2011; FIORIN et al., 2016), with the use of phosphate fertilizer broadcast on the surface, in the sowing furrow or in split form, using soluble forms of P. This reiterates that the application method does not influence productivity, when there is a residual effect of

P from previous crops and/or when the available P content in the soil is above the critical level (NUNES et al., 2011) and higher than the levels suitable for each soil (SOUSA et al., 2016).

For P export and extraction, in the 2015 and 2016 harvests (Table 1), high doses were added, with a critical level for the soil of 9 mg dm<sup>-3</sup> (NEPAR/SBCS, 2017), with the same value being 10,10 mg dm<sup>-3</sup>. Regarding the use of Biomaphos<sup>®</sup>, this was positive in crop productivity, with an increase of 1 and 2 bags per hectare in soybeans and beans, respectively. In general, the application method had a more incisive influence on the increase in productivity of both crops, when compared to the use of Biomaphos<sup>®</sup>. The lower influence

may be linked to the great diversity of microorganisms existing in cultivated soils. For each gram of soil, the existence of approximately 10.000 bacterial species is estimated (TORSVIK et al., 2002).

Another relevant factor that may have resulted in a smaller difference in productive increase, regarding the use of Biomaphos® and a greater difference in increase in the application methods for soybean crops, when compared to beans, was possibly related to the low rainfall that occurred in November 2019, the month of soybean sowing, lower than that which occurred in the months of bean cultivation. One of the factors that regulates life in the soil is water, and its deficit inhibits and/or impairs microbial growth, development, activity, dynamics (MOREIRA; SIQUEIRA, 2006). Therefore, there was no water limitation for bean crops, which also helped maintain soil microbial life. Among these microorganisms, bacteria of the Bacillus genus are capable of solubilizing P (MAHDI et al., 2011; WANG et al., 2020). Bacillus megaterium and Bacillus subtilis strains are capable of solubilizing Ca<sub>3</sub>(PO<sub>4</sub>)<sub>2</sub> from the source rock (PAIVA et al., 2021a). Other studies corroborate the productivity result found (ARAÚJO et al., 2021).

Paiva et al. (2021a), conducting research in 415 units, 212 municipalities and 9 Brazilian states, using soybean seeds inoculated with 100 mL ha<sup>-1</sup> of Biomaphos<sup>®</sup>, concluded that there was an increase in productivity in all locations, with variations between 1 to

15,8 bags per hectare and an average value of 4,8 bags per hectare.

When using Biomaphos®, the greater buffering power of this soil, assuming a P draining role (KOME et al., 2019), especially when applied on the soil surface, favored the absorption of P applied 100% in the sowing furrow, resulting in higher averages for the P content in the soybean leaf (Table 3), reaching reference values of 2,8 to 3,6 g for each kg of soybean plant material, in the R.1 phase (NEPAR/SBSC, 2017). The same was verified for the P content in the bean leaf, with 100% broadcast on the surface application, similar to that without P application (Witness).

The lower foliar P content in soybeans, in the application of 100% phosphate fertilizer by broadcast on the surface, with Biomaphos® inoculation, may be linked to a higher biomass, since the cultivated plants have greater height and dry biomass of the aerial part, promoting a dilution of the absorbed nutrient and decreasing the foliar P concentration, when compared to the absence of the inoculant. In view of the above, it was found that this difference did not impact the content in the grain, generating only significant differences for the application method, in the treatment without the addition of P. In the P content in the soybean grain, for the treatments that received P, there was no significant difference, favored by its high mobility in the plant, redistributed quickly (MALAVOLTA et al., 1997), with the reference value for P export in soybeans being 4,5 g kg<sup>-1</sup> (SBCS/NEPAR, 2017).

**TABLE 3 -** P content in soybean and bean leaves and grains, depending on the method of application of P and BiomaPhos<sup>®</sup> solubilizer.

	P content in soybean leaves (g kg <sup>-1</sup> )		P content in bean leaves (g kg <sup>-1</sup> )		
D1:4:	BiomaPhos®				
P application modes	No application With application		No application	With application	
100% broadcast on the surface	2.89 Aa*	2.57 Bb	2.02 Aa	1.90 Abc	
$\frac{1}{2}$ broadcast + $\frac{1}{2}$ in the furrow	2.80 Aa	2.67 Ab	2.03 Ba	2.38 Aa	
100% in the sowing furrow	2.85 Aa	3.00 Aa	2.36 Aa	2.36 Aab	
Witness (without P)	2.43 Ab	2.42 Ab	1.98 Aa	1.67 Bc	
CV1(%)	7.83		19	19.60	
CV2(%)	7.39		10.13		
	P content in	soybean grains (g kg <sup>-1</sup> )	P content in bean grains (g kg <sup>-1</sup> )		
	BiomaPhos®				
			No application	With application	
100% broadcast on the surface		3.90 a	•••		
$\frac{1}{2}$ broadcast + $\frac{1}{2}$ in the furrow	4.20 a		3.76 Aa	3.81 Ab	
100% in the sowing furrow	e sowing furrow 4.28 a		3.66 Ba	4.15 Aa	
Witness (without P)	ess (without P) 3.27 b		3.33 Ab	3.16 Ac	
CV1(%)	11.97		7.00		
CV2(%)			5	.20	

<sup>\*</sup>Means followed by the same lowercase letter in the column and uppercase letter in the row do not differ from each other according to the Tukey test, at a 5% probability of error.

The use of Biomaphos® positively influenced the P content in bean leaves (stage R.1), in fertilization ½ broadcast and ½ in the furrow. This is because the deposition location of the phosphate fertilizer, added to the

use of Biomaphos®, solubilizing phosphates, producing phytases, IAA and siderophores, allowed greater root development, mainly of thinner roots and consequently greater P uptake (VELLOSO et al., 2020). Neither the

application method nor the use of Biomaphos® were sufficient to increase the P content in bean leaves, to the reference values in the range of 3,5-8,0 g for each kg of plant material in R.1 (SBSC/NEPAR, 2017), all remaining below.

The use of Biomaphos® was not sufficient to positively alter the P content in the leaves and grains of the bean crop when phosphate fertilizer was not added. Unlike N-fixing microorganisms, P-solubilizing microorganisms do not meet the total P requirement of the crop, but rather associate with each other, improving the use of existing P (PAIVA et al., 2021b). Therefore, there was no associative effect in the soil that did not receive P, given the low levels of the nutrient.

The application method without P differed statistically from the other methods of applying phosphate fertilizer for the P content in the bean grain when using Biomaphos<sup>®</sup>. For the study in question, when 100% was applied in the sowing furrow, we obtained an increase of up to 13,32% in the export of P to the grains. Paiva et al. (2020b) obtained increases in the export of P to the grains

of close to 19%, which consequently influenced productivity. The reference value for P export in beans is  $4 \text{ g kg}^{-1}$  (SBCS/NEPAR, 2017).

For the inorganic fractionation of P in soybean (Table 4), there was no interaction between the application modes and Biomaphos®. Only the application modes resulted in a significant difference. The highest Pi values for all fractions were verified when 100% phosphate fertilizer was applied in the sowing furrow. This fact may be due to the buffering capacity of the Latosol, which requires higher doses of P added via fertilization to increase 1 mg dm<sup>-3</sup> of P in the soil (SOUSA et al., 2016) when compared to sandy soils. For fertilization in the sowing furrow, there is saturation of the adsorption sites of the clay minerals of the Latosol, replacing the P in the soil solution and making it available to the crop (KURIHARA et al., 2014). No inorganic P was detected in any treatment for the PiAl fraction. This is because this fraction occurs when Al<sup>+3</sup> is present in the soil, being zero for the soil in question (0 cmolc dm<sup>-3</sup>).

**TABLE 4** - Fractionation of inorganic and total inorganic P in the soil attached to soybean roots, according to the P application method

Fractionation of inorganic P (mg kg <sup>-1</sup> ) and total inorganic matter from the soil attached to the soybean roots					
P application modes	Pi <sub>H2O</sub>	$Pi_{Fe}$	$Pi_{Ca}$	Pi <sub>Al</sub>	Pi-total
100% broadcast on the surface	5.39 b	95.07 b	29.43 ab	ND	129.91 bc
$\frac{1}{2}$ broadcast + $\frac{1}{2}$ in the furrow	7.10 ab	129.04 a	41.34 ab	ND	177.50 ab
100% in the sowing furrow	7.93 a	137.19 a	48.01 a	ND	193.14 a
Witness (without P)	2.90 с	65.73 b	26.14 b	ND	94.79 c
CV(%)	35.74	25.77	52.96		30.29

<sup>\*</sup>Means followed by the same lowercase letter in the column and uppercase letter in the row do not differ from each other by the Tukey test, at 5% probability of error. ND = not detected.

The lowest Pi contents were found in the PiH2O fraction. This result may occur in cultivated soils, since it is easily soluble (MELESE et al., 2015), and also due to the Cl<sup>-</sup> anion, from the NH<sub>4</sub>Cl extractant, not exchanging with the phosphate of covalent bonds (SOUZA JUNIOR et al., 2012). There was a significant difference for the Pi<sub>H2O</sub> contents that received some broadcast fertilization and without P, which can be explained by the lower availability of P in the soil solution due to the larger soil/fertilizer contact area of the broadcast application (MALAVOLTA, 1981) and the non-addition of phosphate fertilizer for fertilization without P. The highest inorganic P contents are found in the PiFe fraction. This is because, in the formation of Latosols, iron oxides/hydroxides are present in the clay fractions, with basaltic origin and present in minerals such as goethite and hematite (MARCOLIN; CALEGARI, 2020).

Table 5 shows the P fractionation for the soil adhered to the roots of the bean crop. There was interaction

between the treatments and Biomaphos® in the  $Pi_{Fe}$  and  $Pi_{Ca}$  fractions. When there was no addition of phosphate fertilizer (without P), an increase in the  $P_{Fe}$  fraction and a decrease in  $P_{Ca}$  were obtained in the presence of Biomaphos®. This is because microorganisms that solubilize P release metabolites capable of acidifying the environment, through the production of organic acids (YOUNG et al., 2013), with  $P_{Fe}$  being favored by the decrease in pH and  $P_{Ca}$  by the increase in pH (PEREIRA et al., 2021). Batista et al. (2018), using different strains of bacteria, including *Bacillus subtilis* and *Bacillus megaterium*, concluded that there was a decrease in the pH of the medium and an increase in P bound to Iron.

The results of the  $Pi_{H_2O}$ ,  $Pi_{Al}$  and total Pi contents were similar to those of the soybean crop regarding the application method, but the concentrations of  $Pi_{Fe}$ ,  $Pi_{Ca}$  and  $Pi_{H_2O}$  were higher than the values obtained for the soybean crop.

**TABLE 5** - Fractionation of inorganic and total inorganic P, of the soil adhered to the bean roots, according to the method of application of P and Biomaphos<sup>®</sup> solubilizer.

	Fractionation of inorganic P (mg kg <sup>-1</sup> ) and total inorganic P in the soil attached to bean r				
P application modes	$Pi_{Fe}$		F	Pi <sub>Ca</sub>	
100% broadcast on the surface	137.46 Aab*	143.13 Aa	38.04 Aa	40.61 Aab	
½ broadcast + ½ in the furrow	186.91 Aa	169.94 Aa	48.85 Aa	40.58 Aab	
100% in the sowing furrow	180.66 Aa	183.28 Aa	51.64 Aa	57.99 Aa	
Witness (without P)	77.02 Bb	193.43 Aa	60.86 Aa	24.94 Bb	
CV1(%)	26.89		41.14		
CV2(%)	26.22		42.21		

	BiomaPhos®			
	No application	With application	No application	With application
	$Pi_{ m H2O}$	$Pi_{Al}$	Pi-total	
100% broadcast on the surface	8.83 b	ND	188.46 b	
½ broadcast + ½ in the furrow	26.06 a	ND	246.70 ab	
100% in the sowing furrow	34.39 a	ND	271.18 a	
Witness (without P)	4.19 b	ND	182.32 b	
CV1(%)	86.93 27.45			7.45

<sup>\*</sup>Means followed by the same lowercase letter in the column and uppercase letter in the row do not differ from each other according to the Tukey test, at a 5% probability of error.

### **CONCLUSIONS**

Only the application methods of phosphate fertilizer changed productivity, P content in soybean grains, fractionation of soil P adhering to soybean roots, and fractionation of soil P adhering to bean roots for  $PiH_{2}O$  and total Pi, with emphasis on those that received some form of P in the sowing line (100% or  $\frac{1}{2}$ ).

The use of Biomaphos® had a positive effect on the P content of soybean and bean leaves, for 100% of the fertilizer in the sowing furrow in soybean and 100% in the sowing furrow or  $\frac{1}{2}$  sowing furrow +  $\frac{1}{2}$  broadcast on the surface, for beans.

For the P content in the grain of both crops, with and without the use of Biomaphos®, the lowest averages were obtained in the treatment without P, reflecting on productivity.

The use of P in different application methods with the addition of Biomaphos® provided an increase in productivity of 1 bag per hectare in soybeans and 2 bags per hectare in beans.

The addition of Biomaphos® to the bean crop provided an increase in the  $Pi_{Fe}$  fraction and a decrease in  $Pi_{Ca}$  for the treatment without P fertilization.

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## REFERENCES

ANDRADE, F.V.; MENDONÇA, E.S.; ALVAREZ, V.; NOVAIS, R.F. Addition of organic and humic acids in Oxisols and phosphate adsorption. **Brazilian Journal of Soil Science**, v.27, [s.n.], p.1003-1011, 2003.

ARAÚJO, F.F.; BONIFÁCIO, A.; BAVARESCO, L.G.; MENDES, L.W.; ARAÚJO, AS. F. *Bacillus subtilis* changes the root architecture of soybean grown on nutrient-poor substrate. **Rhizosphere**, v.18, [s.n.], [s.p.], 2021.

BARBOSA, N.C.; ARRUDA, E.M.; BROD, E.; PEREIRA, H.S. Vertical distribution of phosphorus in the soil according to application methods. **Bioscience Journal**, v.31, n.1, p.87-95, 2015.

BATISTA, F.C.; FERNANDES, T.A.; ABREU, C.S.; OLIVEIRA, M.C.; RIBEIRO, V.P.; GOMES, E.A.; LANA, U.G.P.; MARRIEL, I.E.; PAIVA, C.A.O. Potential of corn rhizospheric and endophytic microorganisms to solubilize iron phosphate and produce siderophores. Research and development bulletin. 1ª Ed. Sete Lagoas: Embrapa milho e sorgo, 2018. 23p.

BRAGA, J.M.; DEFELIPO, B.V. Spectrophotometric determination of phosphorus in soil extracts and plant material. **Ceres Journal**, v.21, [s.n.], p.133-144, 1957.

CHANG, S.C.; JACKSON, M.L. Fractionation of soil phosphorus. **Soil Science**, v.84, [s.n.], p.133-144, 1957.

CRUZ, C.D. GENES - a software package for analysis in experimental statistics and quantitative genetics. **Acta Scientiarum**, v.35, n.3, p.271-276, 2013.

DAMODAR REDDY, D.; SUBBA RAO, A.; RUPA, T.R. Effects of continuous use of cattle manure and fertilizer phosphorus on crop yields and soil organic phosphorus in a Vertisol. **Bioresource Technology**, v.75, [s.n.], p.113-118, 2000.

EMBRAPA. BRAZILIAN AGRICULTURAL RESEARCH COMPANY. **Product with Brazilian technology can reverse external dependence on phosphate fertilizers.** 2019. Available at: <a href="https://www.embrapa.br/busca-de-noticias/">https://www.embrapa.br/busca-de-noticias/</a>

/noticia/45773416/produto-com-tecnologia-brasileira-pode-reverter-dependencia-externa-por-adubos-fosfatados>. Access on: 12 mai. 2023.

FIORIN, J.E.; VOGEL, P.T.; BORTOLOTTO, R.P. Application methods and sources of fertilizers for soybean crops. **Brazilian Journal of Agricultural Sciences**, v.11, n.2, p.92-97, 2016.

GATIBONI, L.; BRUNETTO, C.; SANTOS, D. R.; KAMINSKI, J. Chemical fractionation of soil phosphorus forms: uses and limitations. In: ARAÚJO, A.P.; AVELAR, B.J.R. (Eds.). Topics in Soil Science, Viçosa: Brazilian Society of Soil Science, v.8, p.141-187, 2013.

KOME, G.K.; ENANG, R.K.; TABI, F.O.; YERIMA, B.P.K. Influence of Clay minerals on some soil fertility attributes: a review. **Open Journal of Soil Science**, v.9, [s.n.], p.155-188, 2019.

KURIHARA, C.H.; SILVA, W.M.; TSUJIGUSHI, B.P.; SILVA, J.V.S.; DIAS, M.M. **Phosphate fertilization in the direct planting system.** Technical Circular. 26. Dourados: Soybean Embrapa, 4p., 2014.

MAHDI, S.S.; HASSAN, G.H.; FAISUL-UR-RASOOL, A.H. Phosphorus availability issue- its fixation and role of phosphate solubilizing bacteria in phosphate solubilization. **Research Journal of Agricultural Sciences**, v.2, n.1, p.174-179, 2011.

MALAVOLTA, E. **Agricultural Chemistry Handbook.** 3. ed. São Paulo: Ceres Agronomics, 319p. 1981.

MALAVOLTA, E.; VITTI, G.C.; OLIVEIRA, S.A. **Assessment of the nutritional status of plants:** principles and applications. 2. ed. Piracicaba: Potafós, 319 p., 1997. MARCOLIN, L.; CALEGARI, M. R. Chemical and physical attributes and mineralogy in Latossols (Oxisols) and their relation with the landscape in the west of Paraná. **Revista do Departamento de Geografia**, v.39, [s.n.], p.1-14, 2020.

MELESE, A.; GEBREKIDAN, H.; YLI-HALLA, M.; YITAFERU, B. Phosphorus status, inorganic phosphorus forms, and other physicochemical properties of acid soils of farta district, northwestern highlands of Ethiopia. **Applied and Environmental Soil Science**, v.2015, [s.n.], p.1-11, 2015.

MOREIRA, F.M.S.; SIQUEIRA, J.O. Soil microbiology and biochemistry. Lavras: UFLA, 2002. 625p.

NUNES, R.D.S.; SOUSA, D.M.G.; GOEDERT, W.J.; OLIVEIRA, L.E.Z. PAVINATO, P.S.; PINHEIRO, T.D. Distribution of soil phosphorus fractions as a function of long-term soil tillage and phosphate fertilization management. **Frontiers in Earth Science**, v.8, n.350, p.1-12, 2020.

NUNES, R.D.S.; SOUSA, D.M.G.; GOEDERT, W.J.; VIVALDI, L.J. Distribution of phosphorus in soil due to cultivation system and management of phosphate fertilization. **Brazilian Journal of Soil Science**, v.35, n.3, p.877-888, 2011.

OGUT, M.; ER, F.; NEUMANN, G. Increased proton extrusion of wheat roots by inoculation with phosphorus solubilising microorganims. **Plant and Soil**, v.339, n.1, p.285-297, 2011.

PAIVA, C.A.O.; BINI, D.; MARRIEL, I.E.; GOMES, E.A.; SANTOS, F.C.; COTA, L.V.; SOUSA, S.M.; ALVES, V.M.C.; LANA, U.G.P.; SOUZA, F.F. Inoculant based on phosphate-solubilizing bacteria in corn and soybean crops (BiomaPhos®): frequently asked questions and good inoculation practices. Sete Lagoas: Embrapa Corn and Sorghum, 2021b, 18p. (Embrapa Corn and Sorghum. Research and Development Bulletin, 252).

PAIVA, C.A.O.; COTA, L.V.; MARRIEL, I.E.; ALVES, V.M.C.; GOMES, E.A.; SOUSA, S.M.; SANTOS, F.C; SOUZA, F.F.; LANDAU, E.C.; PINTO JÚNIOR, A.S.; LANA, U.G.P. Validation of the recommendation for the use of the Biomaphos<sup>®</sup> inoculant (Bacillus subtilis CNPMS B2084 and Bacillus megaterium CNPMS B119) in soybean crops. Sete Lagoas: Embrapa Corn and Sorghum, 2021a, 19p. (Embrapa Corn and Sorghum. Research and Development Bulletin, 279).

PAIVA, C.A.O.; COTA, L.V.; MARRIEL, I.E.; GOMES, E.A.; SOUSA, S.M.; LANA, U.G.P.; SANTOS, F.C.; PINTO JÚNIOR, A.S.; ALVES, V.M.C. Technical and economic viability of Biomaphos® (*Bacillus subtilis* CNPMS B2084 and *Bacillus megaterium* CNPMS B119) in corn and soybean crops. Sete Lagoas: Embrapa Corn and Sorghum, 2020a. 21p. (Embrapa Corn and Sorghum. Research and Development Bulletin, 210).

PAIVA, C.A.O.; MARRIEL, I.E.; GOMES, E.A.; COTA, L.V.; SANTOS, F.C.; SOUSA, S.M.; LANA, U.G.P.; OLIVEIRA, M.C.; MATTOS, B.B.; ALVES, V.M.C.; RIBEIRO, V.P.; VASCO JÚNIOR, R. Agronomic recommendation of *Bacillus subtilis* (CNPMS B2084) and *Bacillus megaterium* (CNPMS B119) strains in corn crops. Sete Lagoas: Embrapa Corn and Sorghum, 2020b. 18 p. (Embrapa Corn and Sorghum. Technical Circular, 260). PAVINATO, P.S.; MERLIN, A.; ROSOLEM, C.A. Phosphorus fractions in Brazilian Cerrado soils as affected by tillage. Soil and Tillage Research, v.105, n.1, p.149-155, 2009.

PEREIRA, D.S.; COSTA, Y.K.S.; CARVALHO, L.B. The P element: forms and dynamics in tropical soils. **Brazilian Agronomy Journal**, v.5, [s.n.], p.1-4, 2021.

REED, S.C.; YANG, X.; THORNTON, P.E. Incorporating phosphorus cycling into global modeling efforts: a worthwhile, tractable endeavor. **New Phytologist, Tennesse**, v.208, n.2, p.324-329, 2015.

ROSSI, N.G.; VILAR, C.C.; USHIWATA, S.Y.; REIS, R.; NABEIRO, J.C.X. Influence of phosphate fertilizer application method on soybean production in no-till and conventional planting systems in the Cerrado. **Global Science and Technology**, v.11, n.2, p.101-111, 2018.

SANTOS, D.R.; GATIBONI, L.C.; KAMINSKI, J. Factors affecting phosphorus availability and phosphate fertilization management in soils under no-tillage system. **Rural Science**, v.38, n.2, p.576-586, 2008.

SANTOS, E.D.A.; TNG, D.Y.P.; APGAUA, D.M.G.; ASSUNÇÃO, R.R.; MANZI, A. O. Soil phosphorus fractions and their relation to leaf litterfall in a central Amazonian terra firme rainforest. **Acta Amazonica**, v.52, n.2, p.104-113, 2022.

SANTOS, H.G.; JACOMINE, P.K.T.; ANJOS, L.H.C.; OLIVEIRA, V.A.; LUMBRERAS, J.F.; COELHO, M.R.; ALMEIDA, J.A.; CUNHA, T.J.F.; OLIVEIRA, J.B. **Brazilian soil classification system.** 4.ed. Brasília: Embrapa, 2014. 376p.

SBCS. SOCIEDADE BRASILEIRA DE CIÊNCIA DO SOLO. Fertilization and liming manual for the state of Paraná. Curitiba: SBCS/NEPAR. 182p. 2017.

SOUSA, D.M.G.; NUNES, R.S.; REIN, T.A.; SANTOS JUNIOR, J.D.G. **Phosphorus management in the Cerrado region.** In: FLORES, R.A.; CUNHA, P.P. (Eds.). Soil management practices for adequate plant nutrition in the Cerrado. Goiânia: UFG, 2016. p.291-358.

SOUZA, C.A.; FIGUEIREDO, B.P.; COELHO, C.M.M.; CASA, R.T.; SANGOI, L. Plant architecture and soybean productivity resulting from the use of growth reducers. **Bioscience Journal**, v.29, n.3, p.634-643, 2013.

SOUZA JÚNIOR, R.F.; OLIVEIRA, F.H.T.; SANTOS, H.C.; FREIRE, F.J.; ARRUDA, J.A. Fractions of inorganic phosphorus in soil and their correlations with phosphorus quantified by extractors and corn. **Brazilian Journal of Soil Science**, v.36, [s.n.], p.159-169, 2012.

TORSVIK, V.; GOKSOYR, J.; DAAE, F.L. High diversity in DNA of soil bacteria. **Applied in Environmental Microbiology**, v.56, [s.n.], p.782-787, 2009.

VELLOSO, C.C.V.; PAIVA, C. A.O.; GOMES, E.A.; LANA, U.G.P.; CARVALHO, C.G.; GUIMARÃES, L.J.M.; PASTINA, M.M.; SOUSA, S.M. Genome-guided insights of tropical *Bacillus* strain efficient in maize growth promotion. **FEMS Microbiology Ecology**, v.96, n.9, p.157, 2020.

WANG, Y.; LI, P.; ZHANG, B.; WANG, Y.; MENG, J.; GAO, Y.; HE, X.; HU, X. Identification of phosphate-solubilizing microorganisms and determination of their phosphate-solubilizing activity and growth-promoting capability. **BioResources**, v.15, n.2, p.2560-2578, 2020. YOUNG, L.S.; HAMEED, A.; PENG, S.Y.; SHAN, Y.H.; WU, S.P. Endophytic establishment of the soil isolate *Burkholderia* sp. CC-Al74 enhances growth and P-

utilization rate in maize (Zea mays L.). Applied Soil

Ecology, v.66, [s.n.], p.40-47, 2013.