Humic substances and phosphorous sources in maize crops

Lucas Morais LÔBO¹*, Eva de Melo FERREIRA², Camilla Oliveira MUNIZ³, Carlos Alberto da Silva FERREIRA³, Eliana Paula Fernandes BRASIL³

¹Fortaleza Agrícola Company, Edéia, GO, Brazil.

²Post-Graduation in Sanitation, Environment and Water Resources, Federal University of Minas Gerais, Belo Horizonte, MG, Brazil.

³School of Agronomy and Food Engineering, Federal University of Goiás, Goiânia, GO, Brazil.

*E-mail: lucasloboagro@gmail.com

Received in March/2018; Accepted in March/2019.

ABSTRACT: The objective of this study was to verify the effect of the application of different rates of a commercial product based on humic substances (HS) on the dry matter contents, and P accumulation in maize plants and the agronomic efficiency of phosphorus sources in two soils with different adsorption capacities. The experiment was conducted in a greenhouse. The treatments consisted of four rates of humic substances (0, 100, 200, and 400 kg ha⁻¹) and two phosphorus sources (triple superphosphate – TSP; and reactive natural phosphate – RNP) in two types of soils (Rhodic Hapludox – RH; and Typic Quartzipsamment – TQ). The response of the shoot dry weight (SDW), root dry weight (RDW), shoot accumulated P, and root accumulated P to the application of HS was statistically significant (P <0.05). In the RH, the SDW, RDW, shoot and root accumulated P, and relative agronomic efficiency of the P sources increased with increasing HS rates; thus, the best results were found with the use of the highest rate (400 kg ha⁻¹). In the TQ, the best responses were found with the application of 100 kg ha⁻¹ of HS. These results indicate that the use of humic substances is efficient to increase dry matter and accumulated P contents in maize plants.

Keywords: accumulated P, relative agronomic efficiency, P adsorption.

Substâncias húmicas e fontes de fósforo na cultura do milho

RESUMO: O objetivo do estudo é verificar o efeito da adição de doses de um produto comercial a base de substâncias húmicas (SH), na produção de massa seca de plantas de milho, no acúmulo de P nas plantas e a eficiência agronômica de fontes de fósforo em dois tipos de solos com diferentes capacidades de adsorção. O trabalho foi conduzido em casa de vegetação. Os tratamentos compreendem quatro doses de substâncias húmicas (0, 100, 200 e 400 kg ha⁻¹) e duas fontes de fósforo (superfosfato triplo – SFT e fosfato natural reativo – FNR), em dois tipos de solos (Latossolo Vermelho – LV e Neossolo Quartzarênico – NQ). As fontes de P foram fornecidas em dose constante de 75 mg.dm-3 de P. Houve resposta estatisticamente significativa (P<0,05) da produção de massa seca da parte aérea (MSPA), massa seca das raízes (MSR), P acumulado na parte aérea e P acumulado nas raízes à adição de SH. No LV os parâmetros MSPA, MSR, P acumulado na parte aérea, P acumulado nas raízes e a Eficiência agronômica relativa das fontes de P aumentaram com a adição crescente de SH, obtendo os melhores resultados com uso da maior dose (400 kg ha⁻¹). No NQ as melhores respostas foram obtidas com a adição de 100 kg ha⁻¹ da SH. Estes resultados indicam que a utilização de substâncias húmicas é eficiente para o aumento da produção de massa seca das plantas de milho e no teor de P acumulado.

Palavras-chave: P acumulado, eficiência agronômica relativa, adsorção de fósforo.

1. INTRODUCTION

Humic substances are divided in three categories: humin, fulvic acids consisted of solubilized polymers, and humic acids. Fulvic acids have grater solubility by presenting greater polarity and smaller molecular sizes; they form the main responsible fraction for cation transport mechanisms in the soil. Humic acids present low solubility under the acidity commonly found in tropical soils and are the most studied; they are responsible for the greater part of the cation exchange capacity (CEC) of the organic matter in the soil surface layers (PFLEGER et al., 2017).

The formation of humic substances is one of the less understood and most intriguing humus chemical characteristics. Studies involving the application of these substances in maize crops are essential to understand the behavior of these compounds when they are applied to agricultural crops. The initial understanding on the formation of humic substances is based in four theories: lignin modification, quinone-amino acid interaction, aromatic-acid synthesis by microorganisms, and the Mallard reaction (COLLADO et al., 2018).

ISSN: 2318-7670

Humus is the main component of soil organic matter and affects properties and forms of soils. Humic substances are organic compounds that are distributed in different ecosystems and are valuable for agriculture; they enrich the soil and enable a better development of crops (ZHANG et al., 2017). These substances are the organic matter compartment of more reactivity and are found involved in several chemical reactions. However, little information on the effects of soil cover crops on humic fractions of the organic matter are found, and a better understand of factors that govern the stabilization of humic substances is necessary for searching management

practices that contribute to soil preservation, such as no-tillage systems, and understand other effects of humic fractions (ROSA et al., 2017).

Organic matter is important for soil fertility and environment protection and sustainability, and is essential for agricultural crops. It is also important for the formation of fossil fuels by plant and animal tissue degradation, such as petrol. Humic substances are important for the mobility of environmental contaminants— such as Cd—in rice crops, water retention capacity by promoting mineral particle aggregation, nutrient sequestration and transport, biological activity of microorganisms, and reduction of erosive processes in soils naturally poor in organic matter or degraded by human action through non appropriate management practices (YU et al., 2018). Soils with low organic matter availability require managements that promote an enrichment; thus, the application of organic matter can reduce costs from fertilizer losses caused by leaching.

2. MATERIAL AND METHODS

The experiments were conducted in a greenhouse at the experimental area of the School of Agronomy of the Federal University of Goiás, Samambaia campus, in Goiânia, Goiás (GO), Brazil (16°35'S, 49°16'W, and 722 m of altitude). The climate of the region is Aw (tropical rainy), according to the Köppen classification.

The phosphorus (P) sources used were triple superphosphate (TSP) and reactive natural phosphate (RNP), which present different characteristics of solubility in water. The characterization of the P sources consisted of determination of total P_2O_5 contents, water-soluble P_2O_5 , neutral ammonium citrate + water (NAC + H_2O), and citric acid 2%, according to Brazil (2006) (Table 1).

Table 1. Total P_2O_5 , water-soluble P_2O_5 , neutral ammonium citrate + water (NAC + H_2O), and citric acid 2% of phosphorus sources—triple superphosphate (TSP) and reactive natural phosphate (RNP).

Tabela 1. Teores de P2O5 total, solúvel em água, citrato neutro de amônio + água (CNA + H2O) e ácido cítrico 2% das fontes de fósforo, superfosfato triplo (SFT) e fosfato natural reativo (FNR).

P ₂ O ₅ contents	TSP	RNP
Total (%)	48.3	31.1
Water soluble (%)	40.5	3.3
$NAC + H_2O$ (%)	45.0	5.9
Citric acid (%)	46.0	17.0

Samples of the soil surface layer (0-20 cm) were used for the experiments. The soils used were classified as Rhodic Hapludox (clayey texture) and Typic Quartzipsamment (sandy texture); they were collected in the region of Paraúna, GO, Brazil. The granulometry of the soil samples was characterized by the densimeter method (EMBRAPA, 1999), and the chemical analysis was performed according to the methods described by Raij et al. (2001) (Table 2).

A solid water-soluble organomineral fertilizer based on humic substances (HS) obtained from leonardite was used; the characteristics of this product are described in Table 3. Before the experiment implementation, soil samples were air dried, passed through a 2-mm mesh sieve, and placed in plastic bags for acidity analysis. Liming were conducted to increase the soil base saturation to 60%; the amount of lime used was calculate by the base saturation method (SOUZA; LOBATO, 2004).

Subsequently, the soil moisture was maintained at 80% of the soil retention capacity, during the experiment.

Table 2. Physical and chemical characteristics of samples of the soils Rhodic Hapludox (RH) and Typic Quartzipsamment (TQ). Tabela 2. Características físicas e químicas de amostras do Latossolo Vermelho (LV) e do Neossolo Quartzarênico (NQ).

Characteristic	RH	TQ
Clay (g kg ⁻¹)	650.0	110.0
Silt (g kg ⁻¹)	100.0	40.0
Sand (g kg ⁻¹)	250.0	850.0
Organic matter (g dm ⁻³)	27.0	15.0
Organic Carbon (g dm ⁻	15.6	8.7
CEC (cmol _c dm ⁻³)	4.6	3.1
pH in CaCl ₂	4.3	4.2
$H + Al (cmol_c dm^{-3})$	4.1	2.5
Al^{3+} (cmol _c dm ⁻³)	0.3	0.4
Ca ²⁺ (cmol _c dm ⁻³)	0.2	0.3
Mg ²⁺ (cmol _c dm ⁻³)	0.1	0.2
K^+ (mg dm ⁻³)	70.0	31.0
$P (mg dm^{-3})^{1}$	0.8	2.4
P-resin (mg dm ⁻³) ²	2.0	5.0

CEC = cation exchange capacity; ¹Mehlich-1 extractor; ² Extracted in ion exchange resin.

Table 3. Chemical characteristics of the commercial product—based on humic substances—used.

Tabela 3. Características químicas do produto comercial a base de substância húmica.

Nutrient	%	mg kg ⁻¹
P ₂ O ₅ (Total)	0,70	-
P ₂ O ₅ (NAC)	0,22	-
N	0,10	-
K_2O	8,50	-
Ca	1,62	-
Mg	0,17	-
S	0,20	-
В	-	26
Cu	-	48
Mn	-	34
Zn	-	48
Organic matter	50,10	-
Organic carbon	29,10	-
Moisture	13,90	-

Thirty days after incubation, the soils were placed in 6-liter plastic pots. The treatments applied to the two soils (Rhodic Hapludox and Typic Quartzipsamment) consisted of two P sources (TSP and RNP), and four rates of the commercial product based on humic substances at equal composition, corresponding to 0, 100, 200, and 400 kg ha⁻¹ applied in two crops.

The amounts of nutrients applied and homogenized in the two soils were 75 mg dm⁻³ of P, 100 mg dm⁻³ of K (potassium chloride), 100 mg dm⁻³ of N (urea), 1 mg dm⁻³ of B (borax), and 3 mg dm⁻³ of Zn (zinc sulfate). The pots were arranged in a completely randomized design with five replications.

Five maize (*Zea mays* L.) seeds (Limagrain 1060 hybid) were seeded in each pot. After emergence, the seedlings were thinned, leaving two plants per pot. The plants were removed at 30 days after emergence; their shoots and roots were placed separately in paper bags and dried in a forced-air circulation oven at 60 °C.

Their shoot dry weight (SDW) and root dry weight (RDW) were determined in a digital scale. Their P contents were extracted by nitroperchloric digestion and colorimetrically determined by ascorbic acid reaction (BRAGA; DEFELIPO, 1974). The evaluated parameters were SDW, accumulated P, and relative agronomical efficiency (RAE) of the P sources.

The results of the SDW and accumulated P were subjected to analysis of variance to evaluate the effects of the P sources and humic substances (HS) rates in the SDW and accumulated P contents. The means were compared by the Tukey's test (P<0.05), using the Sisvar program (FERREIRA, 2000).

The RAE of the P sources with and without application of HS was calculated using Equation 1:

RAEi (%) =
$$(\frac{Y \ source \ i-Y \ control}{Y \ source \ s-Y \ control}) \ x \ 100$$
 (Equation 1)

where: RAEi is the relative agronomical efficiency of the P source i; Y source i is the response obtained of dry weight or accumulated P for the source i; Y source s is the response obtained for the P standard source without application of HS; and Y control is the response obtained without application of P and HS.

3. RESULTS

The application of humic substances (HS) affect the shoot dry weight (SDW), root dry weight (RDW), and shoot accumulated P contents of maize plants (Table 4 and 5).

Table 4. Effect of P sources and humic substance rates on shoot and root dry weights of maize plants grown on a Rhodic Hapludox soil. Tabela 4. Efeito de fontes de P e doses de substância húmica na produção de massa seca de plantas de milho cultivadas em Latossolo Vermelho.

Shoot dry weight (g pot ⁻¹)					
P source	Humic substance rates (kg ha ⁻¹)				
P source	0	100	200	400	
TSP	15.26 Aa	17.03 ABa	17.30 Ba	17.97 Ba	
RNP	3.03 Ab	3.09 Ab	3.59 ABb	4.90 Bb	
Root dry weight (g pot ⁻¹)					
P source	Huı	mic substance	e rates (kg ha	1)	
P source	0	100	200	400	
TSP	10.17 Aa	11.30 ABa	12.74 BCa	13.38 Ca	
RNP	5.62 Ab	7.19 ABb	7.76 Bb	12.28 Ca	

TSP = triple superphosphate; RNP = reactive natural phosphate; means followed by the same uppercase letter in the rows (comparing the rates of humic substances for each P source) or lowercase letter in the columns (comparing P sources) are similar by the Tukey's test ($P \le 0.05$).

Table 5. Effect of P sources and humic substance rates on accumulated P contents in maize plants grown in a Rhodic Hapludox soil.

Tabela 5. Efeito de fontes de P e doses de substância húmica no teor de P acumulado nas plantas de milho cultivadas em Latossolo Vermelho.

Shoot accumulated P contents (mg pot ⁻¹)					
P	I	Humic substanc	e rates (kg ha ⁻¹)		
Source	0	400			
TSP	3.32 Aa	3.69 Aa	4.02 ABa	4.33 Ba	
RNP	0.40 Ab	0.52 Ab	0.62 Ab	0.84 Ab	
	Root accumulated P contents (mg pot ⁻¹)				
P	Humic substance rates (kg ha ⁻¹)				
Source	0	100	200	400	
TSP	1.11 Aa	1.26 Aa	1.93 Ba	2.36 Ba	
RNP	0.61 Ab	0.88 Ab	1.12 Bb	1.98 Cb	

TSP = triple superphosphate; RNP = reactive natural phosphate; means followed by the same uppercase letter in the rows (comparing the rates of humic substances for each P source) or lowercase letter in the columns (comparing P sources) are similar by the Tukey's test ($P \le 0.05$).

The SDW and RDW of maize plants grown in the Rhodic Hapludox (RH) soil were higher when using the P source of higher solubility in water (TSP) compared to the RNP source (Table 4). The same result was found for accumulated P contents (Table 5).

The results showed that the SDW, RDW, and accumulated P have significant responses to increasing rates of humic substances up to the rate of 100 kg ha⁻¹ (Tables 6 and 7).

Table 6. Effect of P sources and humic substance rates on shoot and root dry weights of maize plants grown in a Typic Quartzipsamment soil.

Table 6. Effect of P sources and doses of humic substance on the production of dry mass of maize plants grown in Quartzarenic Neosol.

Shoot dry weight (g pot ⁻¹)				
P	Humic substance rates (kg ha ⁻¹)			
source	0	100	200	400
TSP	24.82 Aa	27.90 Ba	27.97 Ba	27.33 Ba
RNP	4.14 Ab	6.11 Bb	6.04 Bb	4.98 ABb

Root dry weight (g pot ⁻¹)					
P	Humic substance rates (kg ha ⁻¹)				
source	0 100 200 400				
TSP	15.82 Aa	18.97 Ba	18.76 Ba	18.12 Ba	
RNP	7.46 Ab	13.17 Bb	12.20 Bb	8.06 Ab	

TSP = triple superphosphate; RNP = reactive natural phosphate; means followed by the same uppercase letter in the rows (comparing the rates of humic substances for each P source) or lowercase letter in the columns (comparing P sources) are similar by the Tukey's test ($P \le 0.05$).

According to the results of SDW and RDW using the P source of high solubility (TSP) (Table 6), the treatments with humic substances presented no significant differences. However, these treatments presented higher results than that without application of humic substances. The treatments with the highest rate of the P source of low solubility (RNP) (400 kg ha⁻¹) did not differ statistically from the control. The treatments with 100 kg ha⁻¹ of humic substances was the most efficient for the maize shoot and root P accumulation, in both P sources. These results indicate that the rate of this humic-substance-based commercial product that present the highest efficiency for Typic Quartzipsamments is 100 kg ha⁻¹.

Table 7. Effect of P sources and humic substance rates on accumulated P contents in maize plants grown in a Typic Quartzipsamment soil.

Table 7. Effect of P sources and humic substance doses on P content accumulated in corn plants grown in Quartzarenic Neosol.

Shoot accumulated P contents (mg pot ⁻¹)				
P	Humic substance rates (kg ha ⁻¹)			
source	0	100	200	400
TSP	7.25 Aa	8.83 Ba	8.91 Ba	8.81 Ba
RNP	1.44 Ab	2.14 Bb	1. 61 Ab	1.59 Ab

Root accumulated P contents (mg pot ⁻¹)					
P	Humic substance rates (kg ha ⁻¹)				
source	0	100	200	400	
TSP	1.33 Aa	2.06 Ba	1.87 ABa	1.79 Aba	
RNP	1.11 Aa	3.05 Cb	2.12 Ba	1.26 Ab	

TSP = triple superphosphate; RNP = reactive natural phosphate; means followed by the same uppercase letter in the rows (comparing the rates of humic substances for each P source) or lowercase letter in the columns (comparing P sources) are similar by the Tukey's test ($P \le 0.05$).

The results of relative agronomical efficiency (RAE) of the P sources as a function of humic substance rates is presented

in Table 8. The application of humic substances increased the RAE of the P sources (TSP and RNP) in both evaluated soils.

The RAE of the P sources in the clayey soil (RH) increased as the humic substances rates were increased. This result shows that the P adsorption capacity of this soil is high, requiring a higher humic substance rate to fill the adsorption sites, resulting in an increase in the evaluated maize variables. The RAE of the P sources increased with the application of 100 kg ha⁻¹ of humic substances to the sandy soil (TQ), and decreased when using higher rates. The formation of metal bonds between humic substances and P are probably responsible for the decreases in the evaluated maize variables.

The comparison of P sources (TSP and RNP) showed that the RAE, in general, is higher with the use of TSP. This can be explained by its higher solubility when compared to the RNP.

Table 8. Relative agronomic efficiency (RAE) of P sources within each humic substance rate and soil, based on shoot dry weight, root dry weight, and accumulated P contents in maize plants.

Tabela 8. Eficiência agronômica relativa (EAR) das fontes de P dentro de cada dose de substância húmica e solo, com base na produção de massa seca da parte aérea, massa seca das raízes e

quantidade de P acumulados nas plantas de milho. TQ / P sources RH / P sources HS rates TSP RNP **TSP** RNP Shoot dry weight (g pot-1) 0 100 13 100 12 19 100 113 13 113 200 115 17 113 19 15 400 120 25 111 Root dry weight (g pot-1) 43 0 100 50 100 100 82 67 122 112 200 129 73 120 75 47 400 137 124 116 Shoot accumulated P (mg pot-1 0 17 100 8 100 27 100 112 11 123 200 122 14 124 19 19 400 132 2.1 122 Root accumulated P (mg pot-1) 0 100 100 82 53 100 115 79 155 230

TSP = triple superphosphate; RNP = reactive natural phosphate; RH = Rhodic Hapludox (clayey soil); TQ = Typic Quartzipsamment (sandy soil).

141

134

159

95

102

225

4. DISCUSSION

180

205

200

400

According to Andrade (2005), the filling of P adsorption sites by humic and fulvic acids in the soil increases phosphate concentrations in the soil solution and, thus, its availability to plants. This high P availability in the soil promoted the absorption of this nutrient by the plants, resulting in a higher P RAIJ, B. van.; ANDRADE, J. C.; CANTARELLA, H.; concentration and dry matter contents in maize plants.

Silva et al. (1999) used humic substances extracted from charcoal in maize crops and found increases in dry matter contents in culms (161.55%), leaves (96.73%), and roots ROSA, D. M.; NÓBREGA, L. H. P.; MAULI, M. M.; LIMA, (162.86%), and in the length and surface area of the root system.

The clay amount and mineralogy that give high P adsorption capacity affect the efficiency of the humic acids in compete with P for adsorption sites (VIOLANTE et al., 1996).

Two situations were observed in the present work: the behavior of the humic substances (HS) in a soil with high clay SILVA, R. M. da; JABLONSKI. A.; SIEWERDT, L.; content (Rhodic Hapludox), in which the highest P availability and accumulated P and dry matter contents in the plants were

found with the use of the highest HS-based product rate (400 kg ha-1). Thus, the highest rate of this product was more efficient in filling the P adsorption sites in this soil. Contrastingly, in the soil with low clay content (Typic Quartzipsamment) and, consequently, lower quantity of adsorption sites, the high HS availability formed metal bonds between P and organic compounds, which can explain the decreases in the evaluated variables when using the two highest rates (200 and 400 kg ha

5. CONCLUSION

The application of humic substances (HS) increases the shoot dry weight (SDW), root dry weight (RDW), and accumulated P content in maize plants grown in a clayey (Rhodic Hapludox) and sandy (Typic Quartzipsamment) soils. The highest values of the evaluated parameters (SDW, RDW, and accumulated P) in the Rhodic Hapludox were obtained with the use of the HS-based commercial product at the rate of 400 kg ha⁻¹. In the Typic Quartzipsamment, the highest values of the evaluated parameters were obtained with the use of the HS-based commercial product at the rate of 100 kg ha⁻¹. The relative agronomical efficiency of the P sources increases after application of HS in Rhodic Hapludox and Typic Quartzipsamment soils and, thus, the application of these substances in these soils is recommended.

6. REFERENCES

ANDRADE, F. V. Ácidos orgânicos e sua relação com adsorção, fluxo difusivo e disponibilidade de fósforo em solos para plantas. 2005. 123 f. Tese (Doutorado em Agronomia: Solos e Nutrição de Plantas) – Universidade Federal de Viçosa, 2005.

BRAGA, J. M.; DEFELIPO, B. V. Determinação espectrofotométrica de fósforo em extratos de solo e material vegetal. Revista Ceres, v. 21, p. 73-85, 1974.

COLLADO, S.; OULEGO, P.; SUÁREZ-IGLESIAS, O.; DÍAZ, M. Biodegradation of dissolved humic substances by fungi. Applied Microbiology and Biotechnology, Germany, 2018, v.102 (8), pp.3497-3511. DOI: https://doi.org/10.1007/s00253-018-8851-6

FERREIRA, D. F. Análise estatística por meio do SISVAR para Windows versão 4.0. In: REUNIÃO ANUAL DA **SOCIEDADE** REGIÃO **BRASILEIRA** DA INTERNACIONAL DE BIOMETRIA, 45., 2000. São Carlos. Anais... São Carlos: UFSCar, 2000. p. 255-258.

PFLEGER, P.; CASSOL, P. C.; MAFRA, A. L. Substâncias húmicas em cambissolo sob vegetação natural e plantios de pinus em diferentes idades. Ciência Florestal, Santa Maria, v. 27, n. 3, p. 807-817, jul.-set., 2017. DOI: http://dx.doi.org/10.5902/1980509828631

QUAGGIO, J. A. Análise química para avaliação da fertilidade de solos tropicais. Campinas: Instituto Agronômico de Campinas, 2001. 284 p.

G. P.; PACHECO, F. P. Substâncias húmicas do solo cultivado com plantas de cobertura em rotação com milho e soja. Revista Ciência Agronômica. 2017, vol. 48, n. 2, 0045-6888. pp.221-230. **ISSN** http://dx.doi.org/10.5935/1806-6690.20170026

SILVEIRA JUNIOR, P. Crescimento da parte aérea e do sistema radicular do milho cultivado em solução nutritiva

- adicionada de substâncias húmicas. **Revista Brasileira de Agrociência**, Pelotas, v. 5, n. 2, p. 101-110, 1999.
- VIOLANTE, A.; RAO, M. A.; De CHIARA, A.; GIANFREDA, L. Sorption of phosphate and oxalate by a synthetic aluminum hydroxysulphate complex. **Europen Journal of Soil Science**, Bedfordshire, v. 47, p. 241-247, 1996. DOI: https://dx.doi.org/10.1111/j.1365-2389.1996.tb01395.x
- YU, Y.; WAN, Y.; CAMARA, A. Y.; LI, H. Effects of the addition and aging of humic acid-based amendments on the solubility of Cd in soil solution and its accumulation in rice. **Chemosphere**, USA, v. 196, p. 303-310, 2018. DOI: https://doi.org/10.1016/j.chemosphere.2018.01.002
- ZHANG, J.; WANG, J.; AN, T.; WEI, D.; CHI, F.; ZHOU, B. effects of long-term fertilization on soil humic acid composition and structure in black soil. **PLoS ONE**, USA, v. 12, p. 1-14, 2017. DOI: https://doi.org/10.1371/journal.pone.0186918