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Incorporation in soil and addition of enzyme inhibitor as a way to increase the efficiency of pig slurry and mineral fertilizer

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ABSTRACT: The incorporation of nitrogen (N) fertilizers in the soil and the use of enzyme inhibitors (EI) can improve the efficiency of N fertilization by reducing losses by ammonia volatilization and nitrate leaching. This study aimed to evaluate the efficiency of EI addition and fertilizer incorporation on both grain and dry mass yields of corn as well as on some soil chemical properties, with focus on N availability. A field experiment was carried out for three years in randomized blocks with four replications. The treatments consisted of a 2 × 5 factorial, including two forms of fertilizers application: superficial (SUP) and incorporated (INC), allocated in the plots; and five fertilizations: mineral fertilizer (NPK); NPK + EI; pig slurry (PS); PS+EI; and control (TEST), allocated in the subplots. The soil mineral N content was determined at 30, 60, and 90 days after fertilizer application (DAA) and these times were considered as sub-subplots. All fertilizers increased the dry corn matter and grain yields and the soil availability of N, P, and K, mainly in the upper layer (0.00-0.05 m). However, only the PS promoted higher productivity when incorporated into the soil relative to the soil superficial application. The incorporation of fertilizers increases soil available P but has little effect on soil mineral N. The EI addition to the fertilizers promotes higher soil mineral N contents in the soil until 30 and 60 DAA respectively when superficially applied and soil incorporated, although this does not increase the corn productivity.

Keywords: dicyandiamide, organic fertilizer, *Zea mays*.

INTRODUCTION

The Brazilian soils, in general, have some chemical limitations to agricultural production, requiring the supply of nutrients to promote high crop yields. However, in addition to increase the production costs, most fertilizers have low uptake efficiency by the plants, particularly nitrogen (N) and phosphorus (P) sources, which have average efficiency of only 51 % (Fan et al., 2004; Dobermann and Cassman, 2005) and 50 % (Roy et al., 2016), respectively. Thus, several products have been used to increase the N fertilizers efficiency, with emphasis on enzyme inhibitors (EI), such as N-(n-butyl) triphosphoric triamide (NBPT) and dicyandiamide (DCD). The NBPT inhibits urease by decreasing the urea hydrolysis, and thus the N loss by ammonia volatilization (Aita et al., 2014; Dall'Orsoletta et al., 2017). The DCD inhibits the ammonia monooxygenase enzyme, present in nitrifying bacteria, thus keeping nitrogen longer in the ammonium form (Gonzatto et al., 2016), which reduces the losses by leaching (Aita et al., 2013).

Some management techniques such as the incorporation into the soil can also improve fertilizers efficiency, especially for N (Dell et al., 2012; Damian et al., 2018) and P (Roberts and Johnston, 2015). Incorporation of ammonium compounds allows retention of ammonium in the soil electrical negative charges and also enables the ammonia, that forms inside the soil to react with hydrogen, returning to ammonium (Singh et al., 2008). Furthermore, when P fertilizers were incorporated into the soil in a concentrated way, microzones in soil with high P availability are created, even in soils with high adsorption capacity (Roberts and Johnston, 2015).

However, the technical feasibility of EI addition and organic fertilizer incorporation still need further evaluation. Withers et al. (2018) emphasized the importance of rational organic wastes use, as a source of nutrients for the sustainability of Brazilian agriculture. In part, this practice is already being used, such as the application of pig slurry (PS) in the south of Brazil (Cassol et al., 2012; Lourenzi et al., 2014; Moraes et al., 2014; Alves et al., 2017). However, techniques to improve the efficiency of PS and reduce its polluting environmental effect still require research efforts, especially to control losses of N through ammonia volatilization in areas conducted under the no-tillage system (Aita et al., 2014; Capoane et al., 2015). In addition, in the absence of soil plowing, nutrients with low mobility accumulate in the soil surface, like P (Boitt et al., 2018). In this case, P can be lost through erosion (Bertol et al., 2017) and cause eutrophication of surface water sources, especially at periods of high rainfall following application of PS (Cherobim et al., 2017).

Thus, our hypothesize is that the application of EI in addition to the incorporation of mineral and organic fertilizers reduces N and P losses at the same time that improve the efficiency of such fertilize, resulting in increases in corn yield. Therefore, this study aimed to evaluate the influence of EI addition and soil incorporation of mineral and organic fertilizers on grain and dry mass yields of corn, as well as on some soil chemical properties, with focus on N availability.

MATERIALS AND METHODS

The experiment was conducted in Lages - SC (27° 49' 00" S, 50° 35' 50" O), in a site located at 892 m a.s.l. The climate is mesothermal humid, with mild summer (type Cfb, according to Köppen classification system), with distributed rainfalls throughout the year and average annual precipitation and temperature of 1,550 mm and 15.6 °C, respectively.

The experiment site is located in a *Cambissolo Húmico Álico* (Santos et al., 2013), which corresponding to a Humic Cambisol (IUSS, 2015), with 8 % slope. Before the experiment, this area has been used for 20 years with native grass species. Also, six months prior to the experiment installation, the soil was limed with 8.0 Mg ha^{-1} of dolomitic limestone incorporated into the 0.00 to 0.20 m layer by plowing and disking. At the beginning of the experiment, the soil presented the following properties: pH(H_2O) = 5.4; clay = 455 g kg^{-1}, and organic matter = 46 g kg^{-1}; 1.5, 5.6, 1.9, and 12.7 $cmol_c$ dm^{-3} respectively for exchangeable Al^{3+}, Ca^{2+}, Mg^{2+}, and Cation Exchange Capacity (CEC); and 3.1 and 92 mg dm^{-3} for available P and K (Mehlich 1), respectively. All evaluations were performed according to Tedesco et al. (1995).

The treatments consisted of a 2 × 5 factorial scheme, including two forms of fertilizers application: superficial (SUP) and incorporated (INC); and five fertilizations systems: mineral fertilizer (NPK), NPK + enzymatic inhibitors (EI), pig slurry (PS), and PS+EI, in addition to the control (TEST), with no fertilizer. Treatments were allocated in a complete randomized blocks design with four replicates. The application form was located in the main plot and the fertilization system in the subplots. The soil mineral N was evaluated at 30, 60, and 90 days after fertilizer application (DAA), which was considered as a sub-subplots. The plot and subplot sizes were respectively 168 m^2 (8 × 21 m) and 33.6 m^2 (8 × 4.2 m), with a useful area of 22.4 m^2.

The experiment was carried out during three crop seasons: 2012/2013 (Alves et al., 2017), 2013/2014 and 2014/2015, with corn (*Zea mays*) in the summer and black oat (*Avena stringosa*) as a cover crop in the winter. Fertilizers were applied 15 days before corn sowing, which always occurred in the second half of October. The mineral fertilizers were urea, triple superphosphate, and potassium chloride applied at doses based on the official research committee for corn (SBCS - NRS, 2004). Thus, the total mineral fertilizer added for the three seasons was 440, 465, and 230 kg ha^{-1} for N, P_2O_5, and K_2O, respectively.

The doses of PS applied were respectively 40, 35, and 47 m^3 ha^{-1} PS for 2012/2013, 2013/2014, and 2014/2015 growing season, to apply the same amount of N provided by NPK treatment. Thus, in the three crop seasons, PS added a total of 435, 864, and 291 kg ha^{-1} of N, P_2O_5, and K_2O, respectively. The PS was collected from stabilized anaerobic lagoons that received the wastes of a commercial meat pork production. The chemical composition of the PS (Table 1) was determined according to Tedesco et al. (1995). The EI was mixed with the fertilizers, in a dose of 10 kg ha^{-1} few minutes before its application in the field. The EI was the Agrotain Plus® that has 81 % of dicyandiamide (DCD) which inhibits the nitrification process and is formulated with the urease inhibitor N-(n-butyl) triphosphoric triamide (NBPT).

The incorporation of PS was performed with a slurry spreader tank with 4,000 L of capacity comprised of a set of incorporation lines developed by the MEPEL® company. This set was adapted from a no-till sowing machine, which has a front cutting disc and a furrower stem, in which the posterior face is located the output of the jet of PS. Thus, incorporations were performed in furrows of 0.12 m deep and spaced 0.35 m, with average width soil mobilization of 0.10 m (Rech et al., 2018). The PS superficial applications were made with the same equipment, but with the incorporation set suspended over the ground. The

Table 1. Dry mass and total N, P, and K contents of pig slurry originated from swine in the growing phase

Crop year	Dry mass(1)	N(2)	P(2)	K(2)
	g kg^{-1}		kg m^{-3}	
2012/2013(3)	38	3.90	2.90	1.30
2013/2014	39	3.42	2.99	3.69
2014/2015	87	3.38	3.34	1.30
Means	54.7	3.57	3.08	2.10

(1) Values determined by the weight difference between the wet swine manure and swine manure oven-dried at 65°C until a constant mass. (2) Contents determined after sulphuric digestion (Tedesco et al., 1995). (3) Data from Alves et al. (2017).

NPK application was manual incorporated in furrows opened by the same incorporation set, and then covered with soil by hoe.

In all crop seasons, we used the 30F53 Pioneer® cultivar, cropped in no-tillage system, with sowing rows spaced 0.7 m, with an estimated population of 65,000 plants ha^{-1}. Weeds were controlled with herbicide glyphosate, applied 15 days before corn sowing, along with the dryness of the oat, and Nicossulfurom, approximately 20 days after oat germination.

Samples of soil from layers 0.00-0.05, 0.05-0.10, and 0.10-0.20 m were collected with a Dutch auger in November 2015, at 30 DAA, to determine the amount of soil available nutrients. To quantify mineral N, soil samples were collected at 30, 60, and 90 DAA. The determinations were performed according to the methodology described by Tedesco et al. (1995).

The aerial parts of corn dry mass were estimated by collecting 12 plants; grains yield was determined by mechanical harvesting of remaining plants in the useful area of each subplot. For dry mass quantification, grains were removed and subsequently the plants were dried at 65 °C until constant weight. The grain and dry mass yields from 2012/2013 were reported by Alves et al. (2017).

The data obtained were subjected to analysis of variance (ANOVA) at ($p<0.05$). The comparison of means was performed by the Tukey test ($p<0.05$), using the statistical software Sisvar 5.6 (Ferreira, 2011).

RESULTS

Corn grain and dry mass yields

The corn dry mass was influenced by fertilization, but not by the form of fertilizers application (Figure 1a). The average corn dry mass from fertilized treatments was approximately 50 % higher than without fertilization. However, there was no difference among fertilizations, regardless of the form (organic or mineral) and the presence of EI addition.

There was an interaction effect of application form and fertilization on corn grain yield (Figure 1b). The effect of application form was restricted to plots fertilized with PS; the incorporation, however, provided greater grain yield than the surface application. In the PS fertilizations, the average grain yields ranged from 6.3 Mg ha^{-1} yr^{-1}, when the PS was incorporated into the soil, to 4.9 Mg ha^{-1} yr^{-1}, when the application was on the soil surface. The fertilization increased grain yield compared to control, regardless of the application form of the fertilizer source (organic or mineral) and the presence or not of EI. The average productivity for the superficial application varied from 5.2 to 3.1 Mg ha^{-1} yr^{-1} for fertilized treatments and control, respectively. For the soil incorporated fertilizers, average yield varied from 5.2 to 3.3 Mg ha^{-1} yr^{-1} on fertilized treatments and control, respectively.

Soil chemical properties

The fertilizers application form did not affect the soil nutrients content, exception for available P in the 0.00-0.05 m layer, where the P on the incorporated treatments was higher than P on the surface application treatments (Figure 2).

Soil pH and the Al^{3+} contents were not affected by fertilization, regardless of the layers evaluated (Table 2). In general, Ca^{2+} and Mg^{2+} were higher than control only on PS treatments and in the first two upper soil layers. For available K^{+} content, the NPK without the addition of EI was the only treatment that gave higher values than the control in all soil layers. For the soil available P content, the effects differed among layers. In the layer of 0.00-0.05 m, the use of PS increased the P content in relation to

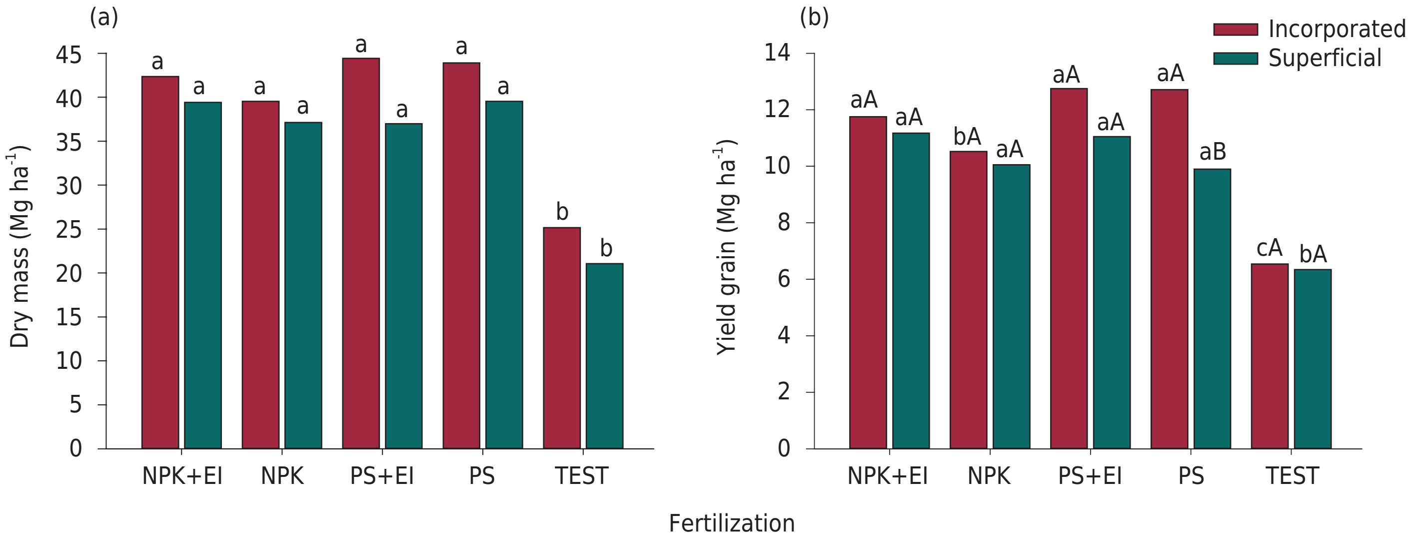

Figure 1. Yield of aerial part dry mass (a) and grains (b) of corn in the second and third crops in a *Cambissolo Húmico Álico* (Humic Cambisol) under fertilization with mineral fertilizer (NPK), mineral fertilizer plus enzyme inhibitors (NPK+EI), pig slurry (PS), pig slurry plus enzyme inhibitors (PS+EI), and control without fertilization (TEST), in application on soil surface and incorporated. Columns with the same lowercase letters among fertilizations within each application form and with same uppercase letters between application forms within each fertilization do not differ among themselves by the Tukey test ($p<0.05$).

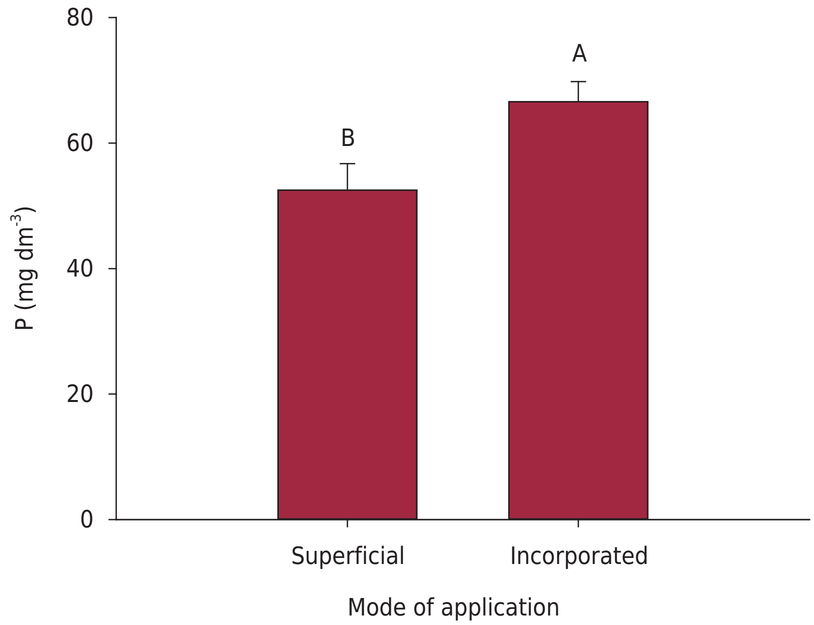

Figure 2. Available P (Mehlich-1) contents in the soil layer of 0.00-0.05 m in a *Cambissolo Húmico Álico* (Humic Cambisol) after three years of fertilization with applications on soil surface and incorporated into the soil. Means of contents from four reps and five tested fertilizations: mineral fertilizer (NPK), mineral fertilizer plus enzyme inhibitors (NPK+EI), pig slurry (PS), pig slurry plus enzyme inhibitors (PS+EI), and control without fertilization (TEST). Columns with the same letters do not differ among themselves by the Tukey test ($p<0.05$). Bars on the columns represent the standard error of the mean.

NPK, and these two fertilizers increased the P content relative to the control. For the 0.05-0.10 m layer, soil available P contents were similar among fertilization, which differed from the control.

Mineral nitrogen in the soil

For the soil mineral N content, a triple interaction was observed between the application time, application forms, and fertilizations (Table 3). In general, the highest contents were

Table 2. Chemical properties in the soil layers 0.00-0.05, 0.05-0.10, and 0.10-0.20 m in a *Cambissolo Húmico Álico* (Humic Cambisol) after three years of fertilizations with mineral fertilizer (NPK), mineral fertilizer plus enzyme inhibitors (NPK+EI), pig slurry (PS), pig slurry plus enzyme inhibitors (PS+EI), and control without fertilization (TEST). Means of the applications forms on the soil surface and incorporated into the soil and four reps

Treatments	$pH(H_2O)$[(1)]	Al^{3+}[(2)]	Ca^{2+}[(2)]	Mg^{2+}[(2)]	K^{+}[(3)]	P[(3)]
		cmol$_c$ dm^{-3}			mg dm^{-3}	
			0.00–0.05 m			
NPK	6.4^{NS}	0.3^{NS}	13.3 bc	11.1 ab	145 a	60 b
NPK+EI	6.4	0.2	13.6 abc	11.3 ab	133 ab	68 ab
PS	6.5	0.1	14.0 ab	12.1 a	135 ab	87 a
PS+EI	6.7	0.1	15.3 a	12.9 a	112 ab	71 ab
TEST	6.5	0.4	12.0 c	9.7 b	96 b	12.1 c
Mean	6.5	0.2	13.6	11.4	124	60
CV%	4.6	133	9.7	11.3	22.3	25.5
			0.05–0.10 m			
NPK	6.0^{NS}	0.6^{NS}	11.9 ab	10.7 ab	121 a	46 a
NPK+EI	6.0	0.8	12.6 ab	10.6 ab	106 ab	48 a
PS	6.0	0.5	13.1 ab	11.1 a	101 ab	54 a
PS+EI	6.1	0.5	14.0 a	12.0 a	97 ab	44 a
TEST	6.2	0.8	11.4 b	9.1 b	90 b	8.0 b
Mean	6.1	0.6	12.6	10.7	103	40
CV%	6.2	99.4	12.3	11.4	16.7	43.1
			0.10–0.20 m			
NPK	5.3^{NS}	1.7^{NS}	9.2^{NS}	10.4^{NS}	106 a	15.5^{NS}
NPK+EI	5.4	1.8	9.1	9.9	93 ab	23.1
PS	6.0	1.4	9.5	10.6	88 ab	24.8
PS+EI	6.1	1.4	9.8	11.9	85 b	17.9
TEST	5.4	1.6	8.5	10.1	81 b	8.3
Mean	5.6	1.6	9.2	10.6	90	17.7
CV%	5.4	68.8	14.9	13.3	15.6	73.1

[(1)] Values determined in samples diluted in water, according Tedesco et al. (1995). [(2)] Extracted from the soil with KCl (1 mol L^{-1}) solution and quantified by atomic absorption spectrophotometry, which methodology was adapted by Tedesco et al. (1995). [(3)] Extracted from the soil by double acid (Mehlich 1), and P quantified by colorimetry, and K, by flame spectrophotometry. CV: coefficient of variation; NS: not significant by the F test ($p<0.05$). Means followed by the same letters in the columns do not differ by the Tukey test ($p<0.05$).

observed at 30 DAA, regardless of the application form and type of fertilizer used. As an exception, the control presented constant contents of mineral N from 30 to 90 DAA, in the layers 0.05-0.10 and 0.10-0.20 m, in the superficial application form.

In relation to the application form, at 30 DAA was observed effect in the NPK treatment without EI, where the soil mineral N content was higher with incorporation relatively to the superficial application, in all soil layers. However, the NPK+EI superficially applied presented mineral N content higher than the incorporated form during this same period, although this effect was limited to 0.00-0.05 m layer. For PS, with or without the addition of IE, the application form showed no differences on the soil mineral N content in all times of evaluation. In general, the fertilizers with the addition of IE superficially applied maintained higher soil mineral N content up to the 60 DAA relatively to fertilizers without EI. This effect is pronounced when the EI were added to mineral fertilizer, maintaining mineral N content higher than the organic fertilizer. However, when the fertilizers were incorporated, the effect of the addition of EI in the soil mineral N content was only observed up to the 30 DAA.

Table 3. N-mineral content in the soil layers 0.00-0.05 m, 0.05-0.10 m, and 0.10-0.20 m in a *Cambissolo Húmico Álico* (Humic Cambisol) after three years of fertilizations with mineral fertilizer (NPK), mineral fertilizer plus enzyme inhibitors (NPK+EI), pig slurry (PS), pig slurry plus enzyme inhibitors (PS+EI), and control without fertilization (TEST) with applications forms on soil surface (SUP) and incorporated into the soil (INC), determined at the times 30, 60, and 90 days after the fertilizers application (DAA). Means of four reps

Form	DAA	NPK	NPK+EI	PS	PS+EI	TEST
		N-mineral[1]				
		mg dm^{-3}				
		0.00–0.05 m				
INC	30	87.8 aBa	116.7 aAb	73.1 aBa	107.3 aAa	28.0 aCa
	60	30.9 bAa	44.5 bAa	32.4 bAa	46.9 bAa	11.8 bBa
	90	18.5 bABa	30.1 cAa	25.2 bAa	28.7 cAa	5.6 bBa
SUP	30	68.7 aCb	133.9 aAa	64.3 aCa	99.3 aBa	25.7 aDa
	60	32.6 bBa	50.6 bAa	40.8 bABa	37.7 bABa	13.9 abCa
	90	19.5 bAa	25.3 cAa	24.5 cAa	23.7 cAa	9.6 cAa
		CV% = 14.98				
		0.05–0.10 m				
INC	30	83.7 aBa	105.3 aAa	69.6 aBa	107.8 aAa	23.1 aCa
	60	23.8 bBCa	42.2 bAa	30.6 bABa	44.5 bAa	8.1 bCa
	90	18.0 bABa	25.4 cAa	25.0 bAa	28.0 cAa	5.2 bBa
SUP	30	60.1 aCb	119.8 aAa	56.8 aCa	92.6 aBa	18.8 aDa
	60	29.8 bAa	46.8 bAa	36.0 bAa	35.4 bAa	12.4 aBa
	90	19.1 bAa	21.9 cAa	18.9 cAa	22.0 bAa	8.0 aAa
		CV% = 17.19				
		0.10–0.20 m				
INC	30	91.9 aAa	105.9 aAa	69.6 aBa	102.7 aAa	23.7 aCa
	60	19.2 bBCa	34.3 bABa	26.7 bABCa	41.1 bAa	7.9 abCa
	90	18.9 bABa	21.5 bABa	21.5 bABa	27.6 bAa	6.4 bBa
SUP	30	62.1 aCb	121.6 aAa	57.6 aCa	83.8 aBa	18.9 aDa
	60	23.7 bABa	38.1 bAa	34.2 bAa	29.0 bAa	9.3 aBa
	90	19.9 bAa	21.4 cAa	18.1 bAa	23.6 bAa	7.2 aAa
		CV% = 20.78				

[1] Mineral nitrogen (NH_4^+ + NO_3^-) was quantified by the macro-kjeldahl method adapted by Tedesco et al. (1995). CV: coefficient of variation. Means followed by the same first lowercase letters between times of determination within each fertilization and form, same uppercase letters between fertilization within each time and form, and last lowercase letters between forms within each fertilization and time do not differ by Tukey test ($p<0.05$).

DISCUSSION

Corn grain and dry mass yields

The lack of difference in grain and corn dry mass yield among the fertilized treatments may be due to nutrient availability that has remained at similar content among the fertilizations (Table 2). Furthermore, the soil nutrient contents are in the class of high availability (CQFS-RS/SC, 2004), regardless of the fertilizer used. The addition of IE did not affect the corn grain and dry mass yields, which agree with results published by Schirmann et al. (2013). Studying fertilization with NPK and split dose of PS (a total of 62 m^3 ha^{-1}), with and without the addition of EI, applied in pre-sowing and topdressing in corn, in a Red Acrisol, these authors also did not observe difference between the NPK and the PS, regardless EI addition. On the other hand, in a Red Ferrasol, Seidel et al. (2010) observed that the PS and mineral fertilizer did not differ on corn grain yield. In the first crop season of this current study, there was also no difference between soil fertilized with PS or NPK, and with EI addition in the corn grains and dry mass yields (Alves et al., 2017). Therefore, it is considered that the fertilization with PS is similar to NPK on the supply of nutrients necessary to corn dry mass and grain yield.

The incorporation of PS presented higher grains yield relatively to the superficial application, probably due to the greater availability of P on the upper soil layer promoted by this form of application. Also, in the study conducted by Damian et al. (2018), the incorporation of pig and bovine slurry provided higher productivity of grains of wheat relatively to the surface application. These authors believe that the increase in grain yield caused by the incorporation was associated with greater mineralization of soil organic matter, as well as intensification of microbial activities promoted by small mobilization of soil with the incorporation machine.

Soil chemical properties

The PS and NPK did not affect soil pH, regardless of the addition of EI and fertilizer application forms. The soil used has high organic matter content, which gives a high buffer power, reducing the variations in pH that could be caused by fertilization. The lack of effect of PS and NPK in pH is also due to the lack of alkalinizing compounds, such as carbonates or organic anions, or acidifying compounds reactions, like nitrification of NH_4^+, in enough amounts to cause significant impacts in the soil pH. Thus, the soil Al^{3+} content was also not affected following PS and NPK fertilization.

The NPK fertilization did not differ from control (TEST) in the soil Ca^{2+} and Mg^{2+} contents, which is explained by the fact that the amount of added calcium by the triple superphosphate is not relevant. However, larger amounts of Ca^{2+} and Mg^{2+} were applied with the PS, which explain the increase in soil Ca^{2+} and Mg^{2+} contents with PS fertilizer relatively to the control. The PS fertilizer added a higher quantity of K^+ than the NPK, besides the latter presented higher soil K^+ content. According to Ernani (2016), potassium has a weak strength of attraction in the negative soil charges in relation to the Ca^{2+} and Mg^{2+}. Thus, the greater addition of Ca^{2+} and Mg^{2+} with the PS may have promoted the leaching of K^+ added by the organic fertilizer, as observed by Sacomori et al. (2016). Maggi et al. (2011), applies PS once at the doses of 0, 100, 200, and 300 m^3 ha^{-1} and observed high concentrations of K^+ in leachate in the higher doses.

The doses of PS were based on the recommendation of nitrogen for the corn. Therefore, the quantities of P and K were higher than the crop needs. As a result, much of the quantity added was not absorbed by the plants and remained available in the soil, whereas in the mineral fertilization nutrients were added according to the crop recommendation. Therefore, on the first layer (0.00-0.05 m), the P content was higher for the PS treatment relative to the NPK. However, because P is little mobile in the soil, this effect was not observed in the 0.05-0.10 m layer, where the fertilizers effects were similar; no effect was observed in the 0.10-0.20 m layer. However, successive applications of high doses of PS for many years, in addition to increasing the levels of P on the superficial layers, can also increase in the subsurface layers (Lourenzi et al., 2015; Boitt et al., 2018).

The surface application increases the nutrients contents in the most superficial layers, mainly of those nutrients with low mobility, like P (Boitt et al., 2018). However, the most superficial layer is also more susceptible to losses of those nutrients by both erosion and runoff (Baker et al., 2017; Bertol et al., 2017). Thus, the slurry applied superficially favors the occurrence of losses of P by surface runoff (Bierer et al., 2017). Comparing the superficial and incorporated applications of fertilizers, higher levels of P are observed in the first layer in the incorporated application, indicating that the incorporation of fertilizers prevents the P lost at the same rate as when applied superficially (Kleinman et al., 2011; Cherobim et al., 2017). Thus, the incorporation of fertilizers in the soil mitigates potential environmental problems, such as the water eutrophication.

Despite the benefits of organic fertilizers incorporation, the adoption of this technique has not been high due to the higher energy consumption and cost of the equipment (Maguire et al., 2011). Chen et al. (2014) studied the cost of different fertilizer application techniques and found that the incorporation has a higher initial cost. Nevertheless, as over the years of use, the economic result of incorporation may be greater than the surface application due to higher fertilizer efficiency and a consequent decrease in fertilizers purchase. Rotz et al. (2011) also compared different application techniques and concluded that the additional costs of the incorporation of organic fertilizers are compensated with the long-term economic return.

Mineral N in the soil

The decrease in the soil mineral N contents over time occurred in all the evaluated conditions. At 90 DAA all treatments without the soil mobilization presented levels equivalent to the control, which did not receive N during the three years. This behavior was expected, because the increase in mineral N content resulting from the fertilizers is not permanent (Gonzatto et al., 2016), this is due to N absorption by the plants, and above all, to losses processes that usually occur with this nutrient, especially by nitrate leaching and denitrification (Bayer et al., 2015; Rauber et al., 2017).

When the N fertilizers are incorporated in the soil, the N losses by ammonia volatilization decrease sharply due to the exposure of ammonia in the air is reduced and its retention by soil particles increases (Dell et al., 2012; Aita et al., 2014; Rauber et al., 2017). In addition, incorporation increases the chances of the ammonia react with acidic soil zones, incorporating proton and returning the form of ammonium. Thus, higher mineral N contents would be expected in the treatments with fertilizers incorporation and this was observed for the NPK at 30 DAA. However, where the PS fertilizer was incorporated there were higher mineral N contents than the control at 90 DAA, indicating that the soil mobilization by the incorporation process of fertilizers, even being small, is able to stimulate N mineralization from organic matter by micro-organisms (Raiesi and Kabiri, 2017).

The EI DCD acts on the enzyme ammonia monooxygenase, delaying the nitrification (Gonzatto et al., 2016), while the NBPT inhibits the enzyme urease and acts by lengthening the urea hydrolysis period (Manunza et al., 1999). The duration of the effect of inhibition is also distinct, the DCD acts in the soil for up to 30 days, depending on the conditions of temperature and humidity (Kelliher et al., 2008; Rauber et al., 2017, 2018), while the NBPT has effect not more than 15 days after its application (Dall'Orsoletta et al., 2017). Thus, the treatments with the addition of EI maintained higher soil mineral N content up to the 30 DAA and after this time, with the reduction of the effect of EI, this content tended to be similar among the treatments.

Considering the effect of EI, it is remarkable that the maintenance of high mineral N content occurs when these products were added to the urea (Turner et al., 2010). However, when applied to PS fertilizer, the EI does not have the same efficiency in maintaining the levels of mineral N, probably because the greater part of the N in this waste are ammonium (Gonzatto et al., 2016), and therefore is not affected by the NBPT. This difference observed between organic and mineral fertilizers must be considered in order to choose the use of EI (Aita et al., 2014; Gonzatto et al., 2016; Rauber et al., 2018).

Another important issue to be highlighted is that the evaluated EI has as main objective to keep the N in the form of ammonia longer in the soil, being common to find this effect until approximately 25 DAA (Gonzatto et al., 2016; Rauber et al., 2018). Thus, with decreasing the nitrification process, the added N is maintained in the ammonium form for a longer time, allowing soil mineral N content up to around 30 DAA, regardless of the application form. This increase in the availability of mineral N of the soil, however, despite having presented a significant correlation with yield (R 0.731***), was not enough to increase the yield of corn grains. Even in the fertilized treatments where EI was not applied, the contents of mineral N in the soil were sufficient to sustain the grains yield allowed by the edaphic-climatic conditions of the experiment at levels similar to those treatments with addition of EI, corroborating with results of other authors (Schirmann et al., 2013; Gonzatto et al., 2016).

The relatively low grain yield obtained in the experiment also favored the lack of response to the addition of EI in the two crop seasons evaluated, especially because the soil has a high organic matter content, which can provide much of the N absorbed by plants. In addition, under the studied conditions, the amount of N required by crops was supplied in a single, but high dose despite the N exposure to processes of loss. However, the EI could positively influence the corn grains yield when it reaches higher values, where the difference found in the soil mineral N content could result in increased yield. Thus, future studies to investigate the effect of the EI (DCD and NBPT) addition to N fertilizers should be conducted under high productivity conditions and with the use of moderate doses to enable yield differences due to the maintenance of higher soil mineral N content throughout the growing season.

CONCLUSION

The enzyme inhibitors addition to N fertilizer keeps high soil mineral N content long after the fertilization, but this may not provide increases on corn dry mass and grain yields.

The pig slurry incorporation increases its fertilizer efficiency as it provides an increase in corn grain yield, but this was not observed to mineral NPK fertilizers.

The soil fertilization, regardless of the tested fertilizer source or the of enzyme inhibitors addition increases the soil N, P, and K availability, which favors the corn yield.

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