

**University of São Paulo
“Luiz de Queiroz” College of Agriculture**

**Legacy phosphorus accumulation under long-term soil and fertilizer
management in crop production**

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Thesis presented to obtain the degree of Doctor in
Science. Area: Soil and Plant Nutrition

**Piracicaba
2019**

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**Legacy phosphorus accumulation under long-term soil and fertilizer management
in crop production**

versão revisada de acordo com a resolução CoPGr 6018 de 2011

Advisor:

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DEDICATION

To Verediana, my great grandmother, and Décio, my grandfather, who passed away during my time in this research, which broke my heart - and which remain present in my mind and inspire me to go on.

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EPIGRAPH

*“Once you have tasted flight,
You will forever walk the earth with your eyes turned skyward,
For there you have been,
And there you will always long to return”.*

Leonardo da Vinci

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RESUMO

Legado do fósforo sob manejo a longo prazo do solo e fertilizantes fosfatados na produção agrícola

O fósforo (P) é o segundo macronutriente que mais limita a produção agrícola devido seu baixo nível de disponibilidade nos solos, exigindo alta demanda para obter alta eficiência das culturas em curto-prazo, porém, é um recurso não renovável. Em longo-prazo, diferentes adições desse fertilizante podem afetar as formas e o legado do P no solo e sua contribuição para aumentar a eficiência do seu uso pelas culturas ao longo do tempo. Neste contexto, **no capítulo dois**, avaliamos as mudanças nos pools de P do solo (lábeis, moderadamente lábeis e não lábeis) e o acúmulo do legado do P no solo após a fertilização P a longo prazo com o aumento do P inicial do solo com o uso de doses iniciais de P e aplicações anuais de P. As frações de P do solo mostraram um efeito significativo à aplicação de P nas frações lábeis de P com a adoção das doses anuais de 60, 90 e 120 kg de P_2O_5 ha⁻¹, enquanto os efeitos da aplicação inicial de P foram menos significativas para este solo tropical altamente intemperizado. Assim, o acúmulo de P nas frações lábeis promovidas pelas doses anuais de P poderia ajudar a melhorar a disponibilidade e legado do P no solo e representar uma estratégia lucrativa para reduzir grandes entradas de fertilizantes fosfatados inorgânicos nesses sistemas tropicais de cultivo. **No capítulo três**, avaliou-se a influência da adubação fosfatada a longo-prazo nas frações e legado do P do solo, sob rotação milho-soja. Foram observadas mudanças significativas nos frações de P do solo pelo efeito da aplicação de fertilizantes fosfatados em longo-prazo. A colocação do fertilizante fosfatado a lâzo aumentou o P lábil e moderadamente lábil na camada superficial do solo (0-7,5 cm) e teve maior eficiência de uso deste fertilizante em comparação à aplicação profunda. O uso de estratégias de aplicações combinadas contribuirá para manter as frações de P disponíveis maximizando a eficiência da fertilização fosfatada nos sistemas de cultivo reduzido. **No capítulo quatro**, avaliamos os efeitos da adubação fosfatada a longo-prazo (aplicação inicial e doses e aplicações anuais de P), na transição do pasto para cultivo de soja e milho safrinha sob plantio direto, nas mudanças e acumulação das frações e legado do P no solo. As frações de P no solo mostraram um efeito significativo à aplicação de P nas frações lábeis na superfície do solo com a adoção das doses e aplicações anuais de 100B e 100SP kg P_2O_5 ha⁻¹ em todas as aplicações iniciais de P, e 100B para todas as outras frações de P e profundidades do solo, enquanto os efeitos da aplicação inicial de P foram menos significativos para este solo tropical altamente intemperizado. Assim, o acúmulo de P da aplicação inicial (efeito residual do P no solo após cinco anos) e as aplicações anuais do fertilizante fosfatado nas frações de P lábil podem ajudar (pelo menos em parte) a reduzir a alta dependência atual às grandes entradas de fertilizante fosfatos inorgânico nos sistema de plantio direto.

Palavras-chave: Fertilização inicial; Fertilização anual; Aplicação de P; Cultivo convencional; Cultivo reduzido; Plantio direto; Fracionamento de Hedley; Frações de P; Formas de P; Labilidade de P

ABSTRACT

Legacy phosphorus under long-term soil and fertilizer management in crop production

Phosphorus (P) is the second macronutrient that most limits the agricultural production due to its low level of availability in the soils, thus requires high demand to obtain high crops efficiency in the short-term, however, it is a non-renewable resource. In long-term, different P fertilizer placement can effect, particularly the preferential forms of P retained and legacy in the soil pool and their contribution to increasing P use efficiency by crops over time. In this context, in the present thesis, **in the chapter two**, we aimed to assess changes in soil P pools (labile, moderately labile and non-labile) and legacy soil P accumulation after long-term P fertilization with an initial soil P buildup and annual P applications. P pools in the soil showed a significant effect of P application on labile P fractions with the adoption of annual rates of 60, 90 and 120 kg P₂O₅ ha⁻¹, while effects of the initial P application were less significant for this tropical high weathered soil. Thus, P accumulation from annual rates in the labile fractions of P could help to improve the soil legacy P availability and represent a profitable strategy to reduce large inputs of inorganic P fertilizer in tropical crop production systems. **In the chapter three**, we evaluated the influence of long-term P fertilizer placement on soil P pools and legacy soil P accumulation under a corn-soybean long-term rotation. Significant changes in soil P pools were observed by the long-term effect of P fertilizer placement. Broadcast P fertilizer placement increased the labile P and moderately labile P in the topsoil (0-7.5 cm), and had a greater P fertilizer use efficiency compared to deep band placement. The use of combined placement strategies will contribute to maintain the available P pools maximizing P fertilization efficiency in reduced tillage systems. **In the chapter four**, we evaluated the effects of long-term P fertilizer placement (initial placement and annual rates and placement of P) in the transition of pasture to double-crop of soybean and corn under no-till crop production on changes in the soil P pools and legacy soil P accumulation. Results of P pools in the soil showed a significant effect of P application on labile P fractions in soil surface with the adoption of annual rates and placement of 100B and 100SP kg P₂O₅ ha⁻¹ under all initial P application, and 100B for the all others P fractions and soil depths, while effects of the initial P application were less significant for this tropical high weathered soil. Thus, P accumulation from the initial (P remains in the soil after five years) and annual P fertilizer application in the labile P pools could help (at least in part) to reduce current high dependency and large inputs of inorganic P fertilizer in tropical no-till systems.

Keywords: Initial P fertilization; Annual P fertilization; P placement; Conventional tillage; Reduced tillage; No-till tillage; Hedley's fractionation; P fractions; P forms; P lability

1. GENERAL INTRODUCTION

Phosphorus (P) is an indispensable element for all life on Earth (Kruse et al., 2015). For crop production is a major issue since P is the most immobile of the soil macronutrients, and the availability or effectiveness of fertilizer sources for plant nutrition is limited. Phosphorus deficiency is a universal constraint to high crop yield and constitutes the second most important soil fertility problem throughout the world (Rashid et al., 2005). In nature, P occurs predominantly in the form of phosphate (four oxygen [O] atoms surrounding the central P atom; PO_4^{3-}). In plants, is mandatory in the maintenance of the organic cell: it is the key component of nucleotides that guarantee the helical structure of deoxyribonucleic acid (DNA) and ribonucleic acid (RNA) molecules; it makes up adenosine triphosphate (ATP), an indispensable source of energy for numerous biochemical processes (photosynthesis); and participates in the formation of phospholipids, which guarantee the structure of biological membranes (Theodorou and Plaxton, 1993; Childers et al., 2011).

Phosphorus is the twelfth most abundant chemical element in the earth's crust. Total P concentrations in soils range from 10^1 to 10^3 mg P kg^{-1} , depending on soil horizon with lower in the subsoil than in the soil surface; substrate is lower for sandy than loamy; pedogenesis is lower for older than younger; land-use is lower for forest than pasture and agriculture; and its land-use intensity is lower for extensive than intensive (Kruse et al., 2015). However, P deficiency is the most common nutritional stress in many regions of the world, affecting 42% of the cultivated land over the world (Liu et al., 1994). Although the total amount of P in the soil may be high, only a small proportion (< 6%) is available for plants (Yang et al., 2013).

In soil solution, P is found in the form of primary orthophosphate (H_2PO_4^-) in acid soils and secondary orthophosphate (HPO_4^{2-}) in alkaline soils. The movement of these ionic forms of P for taking up by plants and microorganisms occurs mainly by diffusion about 95% (Abdala et al., 2012). The concentration in soil solution is very small, principally H_2PO_4^- (typically 10^{-5} M), so there must be a supply of readily-available P in the soil to maintain this concentration as P for root absorption (Roberts and Johnston, 2015). Variations in yield potential and uptake of P from the soil is expected among plant varieties, due to the differences in the strategies developed by roots for enhancing nutrient acquisition. Under a low P availability soil situation, an extensive application of P fertilizers is required to supply crop demands.

The phosphate fertilizers are produced from phosphate rock (PR); and the practice of using PR as a source of P in agriculture is ancient The PR deposits of interest for mining

generally occur only under special conditions in some specific areas as a result of the P cycle (Filippelli, 2011). About 60% of the phosphate fertilizer applied to agricultural crops comes from PR resource (Cordell, 2010) and it has fundamental role to sustain high crop yields in modern agriculture (Cooper et al., 2011). Around 95% of this P is found as a calcium-phosphate mineral, known as apatite (Smil, 2000). Most sedimentary rocks contain very small concentrations of P, typically around 0.1%, but under certain conditions, which very rarely occur in the same place at the same time, and over millions of years, this concentration can increase to produce PR, like as phosphorites with 10-20% of P, represented about 100x more than the concentration of typical sedimentary rocks (Filippelli, 2011). Some studies claim that, at current rates of extraction, global commercial phosphate reserves will be depleted in the next two decades (Sattari et al., 2012). Van Kauwenbergh et al. (2013), who estimated recently that the world has over 300 years of P reserves for exploration and 1400 years of phosphate resources. Thus, the reserve estimates for PR are dynamic and can change for a number of reasons like the discovered of know reserves.

The geopolitical issues about PR will be increasingly influence in future P production because most of the known reserves of PR are present in Morocco (74%) while Europe has virtually no phosphate reserves remaining (Menezes-Blackburn et al., 2017). Also, China is the second and United States is the third largest PR producer in the world (Jasinski, 2018), however, their domestic phosphate reserves will deplete in fifty years (Cooper et al., 2011). Morocco will obtain a much greater share of worldwide production, from around 15% in 2010 to around 80% by 2100, which implies more control over market prices (Cooper et al., 2011). While there is no immediate concern of running out PR, the increasing demand for P fertilizers, the need to maintain and improve P status of many agricultural soils, especially in developing countries, and the adverse impacts of the transfer of soil P to surface waters and resulting detrimental effects of eutrophication, use P efficiently is vitally important and commonly acknowledged (Roberts and Johnston, 2015).

However, P recovery by crops in the year of the application rarely exceeds 25% and more often is only 10-15% of the P applied (Roberts and Johnston, 2015), 10-20% (Cornish, 2009) or 15- 25% (Li, 1999). While with optimal management, the long-term recovery of applied P from some soils can reach 80% (Wang et al., 2009; Song et al., 2011); the recovery of applied P by crops is often much lower, due to sorption reactions of P with Al- and Fe-oxides (acidic soils) or reactions with Ca to form less soluble or insoluble compounds (alkaline soils) (Wang et al., 2014). The main consequences are the low efficiency of P use and the competition between plants and soil by this element, causing a mismatch between the

crop demand and the quantity applied to provide it. Thus, the knowledge about soil P forms and dynamics is extremely important for the decision-making in the management of P fertilization in the crop production systems.

Soil P can be found in inorganic (Pi) and organic (Po) compounds that range from ions in solution to very stable inorganic compounds (Turner et al., 2007); this division into two groups is to facilitate the understanding of soil P dynamics. However, the identification of P forms within these two groups is difficult due to the infinity of reactions that this element faces up to and its resulting compounds (Gatiboni, 2003), and that differ by the degree of stability or solubility, with distinct availability to plant absorption (Souza et al., 2007).

The first group is Pi, which usually represents the greatest part of soil total P and often the main limiting factor for plant growth in terrestrial ecosystems (Devau et al., 2009). The concentration of Pi varies between 35 and 70% of the total P in the soil and the minerals formed can be separated into two groups: P of the primary minerals, or structural P; and P adsorbed or precipitated to the colloidal mineral fraction of the soil (Oelkers & Valsami-jones, 2008). The P of primary minerals, such as apatites and strengites, has very slow release. Despite the constant action of weathering agents, the amount of P released in a synchronized way with crops demands is insufficient. The P adsorbed to secondary minerals, such as Ca, Fe and Al phosphates, may present different patterns of availability because of their dependency on the mineral size and soil pH (Oelkers & Valsami-jones, 2008). Due to the high affinity of phosphate ion to forming complexes with high binding energy, the adsorbed or precipitated Pi to the mineral colloidal fraction can occur in all minerals in the soil. Therefore, Pi can be found in soil bound to Fe, Al and Ca, among others, and adsorbed to Fe and Al oxy-hydroxides (Parfitt, 1978).

Oxy-hydroxides of Al and Fe are the most abundant form of Al and Fe in soils, and therefore sorb more P than layer silicates and other crystalline forms of Al and Fe (McLaughlin et al., 2011). Adsorption to Fe and Al oxy-hydroxides occurs mainly in the forms of low crystallinity and with high unbalance of charges. The magnitude of the adsorption depends on the number of ligand sites capable of adsorbing neutral or electrically charged molecules. In acid soils, such as in tropical and subtropical regions, the principal Fe oxy-hydroxides are hematite, goethite, maghemite, magnetite and ilmenite and Al hydroxide is the gibbsite (Ferreira et al., 2007). Thus, the low crystallized Fe oxides, the amount of Al replacing Fe and, in smaller quantities, high crystallized oxides and kaolinite, are the main responsible for P adsorption (Rheinheimer et al., 2008). In addition, Fe and Al oxy-hydroxides have large specific surface area, responsible for large number of adsorption sites. In addition, these

colloids have numerous nanopores capable of promoting P occlusion, which also leads to the unavailability of the element (Aray and Sparks, 2007).

Adsorption of P to the inorganic colloids has an initial phase which electrostatic attraction, following a specific adsorption or chemisorption, through the exchange of ligands. Also, the adsorption reactions can be classified according of the number of functional groups, resulting in the formation of monodentate or bidentate internal sphere-complex, with one or two phosphate oxygens shared to one metal of the colloid surface, respectively; and binucleate internal sphere-complex, with two phosphate oxygen shared to two metal of the colloid surface (Novais et al. 2007; Rheinheimer et al., 2008). The binding energy increases from monodentate to bidentate and binucleate, and reversibility or desorption of phosphate decreases in the same order.

In general, P presents higher availability when the soil pH is maintained at 6.5 or within the range of 5.5 to 7.0 for plant uptake. In basic soils, such as temperate regions, the solubility of P is predominantly determined by Ca content (Bertrand et al., 2003) through P precipitation reactions, especially when in contact with the surface of Ca carbonates and clay minerals (Devau et al., 2010). In addition, the free CaCO_3 in calcareous soils can absorb P ions at low solution P concentrations (Freeman and Rowell, 1981). In this condition, phosphates react with Ca, generating dicalcium phosphate (DCP), which is unavailable to plants. However, DCP can be transformed into even more stable forms, such as octocalcium phosphate or hydroxyapatite, which are very little available for plants (Shen et al., 2011). Otherwise, in acidic soils precipitation reactions in soil solution usually form Al-P or Fe-P mineral phases (Hedley and McLaughlin, 2005) like variscite ($\text{AlPO}_4 \cdot 2\text{H}_2\text{O}$) and strengite ($\text{FePO}_4 \cdot 2\text{H}_2\text{O}$) (Sato et al., 2005). At low pH these hydroxyl-phosphates are stable, however, with increasing pH these phosphates have their solubility increased.

The second group, Po, which can constitute 5 to 80% of the total P in the soil (Sharpley, 1985), with 5 to 35% to the fertilizers topsoil; 70 to 80% to the grassland and forest soil (Achat et al., 2010) and to agriculture in general between 25 and high than 50% (Nash et al., 2014). In tropical soils, Po is an important P source for plants and must be taken into account in studies involving its dynamics and bioavailability (Rheinheimer & Anghinoni, 2001). Also, Po can be a dominant fraction in temperate soils and exists in several different chemical species (Stutter et al., 2015). The Po originates from plant residues added to soil, microbial tissue and products of its decomposition (Rheinheimer et al., 2000, Conte et al., 2002 and 2003; Martinazzo et al., 2007). The P contained in the microbial biomass, which represents a small fraction of the Po, usually presents less than 10% of the total, including the

P of the residues and OM. The wide variety of organic compounds in the soil justify why more than a half of the forms of organic P have not been identified yet (Weihrauch and Opp 2018).

The Po is subdivided in two ester group ($\equiv\text{C-OPO}(\text{OH})_2$): monoester and diester, with one or two ester groups per phosphate ion, respectively (Turner et al., 2005). The main forms already identified are the monoester with phytate or inositol phosphates, which comprise 10 to 80% of the total Po and is slightly soluble (Dalal, 1977; McLaren et al., 2017). The diester consist less than 10% and include phospholipids (0.5 to 7%), nucleic acids (~ 3%) and other phosphate esters (> 5%) (Dalal, 1977; Kruse et al., 2015). The amount of Po fractions founded in the soil will depend on the mineralization of these forms (Weihrauch and Opp, 2018) which the microbial biomass acting as both a source and a sink of soil P, being crucial for P cycling (Zhu et al., 2018). Still, the stability of these Po compounds depends on their nature and their interaction with the mineral fraction, since they are used as a source of carbon and electrons by microorganisms, whose result is their mineralization and availability of P in solution (Gatiboni et al., 2007). Thus, the transformation of Po plays a crucial role in recycling and bioavailability of this nutrient, what is influenced by a number of factors such as soil moisture, temperature, soil pH and redox potential (Shen et al., 2011).

As long term effects, soils with adequate inputs of phosphate fertilizers for plant growth and development will be able to provide inorganic and organic forms of P in similar amounts for plants. In soils with low or no P fertilizer addition, the organic forms are mainly responsible for supplying this nutrient for plants (Gatiboni et al., 2007). On the other hand, higher amount of inorganic P fertilizer added can be transformed to Po (Condrón et al., 2005). Therefore, a better understanding of the role and dynamics of P in the soil is essential for a correct use decision of phosphate fertilization placement and rates. Thus, the management of fertilization should favor absorption and decrease soil fixation processes and, consequently, increase the P utilization by plants (Novais & Smyth, 1999).

To improve nutrient availability and increase fertilizer use efficiency different placements for distinct tillage systems have been studied (Farmaha et al., 2012). The most commonly P fertilizers placements are broadcast, band and deep band, the last one with increasing use in the temperate soils. Broadcast fertilizer placement is a relatively less costly and popular method that is ideal for high-speed operations and high application rates (Khatiwada et al., 2014). This management makes practically 100% of the phosphate fertilizer come into contact with the soil colloidal phase, allowing high adsorption of P and reducing the use of this element by the plant. Therefore, under conditions of low P availability, banded

P is usually more effective. Banded P reduces nutrient contact with the soil colloids and results in less fixation (Grant, 2001). However, small part of the plants roots is exposed to the P from fertilizer (Malavolta, 1981). The positive effect of this localized P placement in a restricted volume of soil will be greater as much as the higher P sorption capacity (Anghinoni and Barber, 1980; Novais and Smyth, 1999). The deep band have been used in recent years as an alternative for low soil test P levels and to reduce the potential for P runoff and possible environmental degradation (Randall and Vetsch, 2008). Previous research evaluated extensively the differences between broadcast and band or deep band P application placement in short-term crop response under distinct soils, available P levels, tillage and cropping systems. Broadcast was more efficient in corn production than band in an Ultisol (Barreto and Fernandes, 2002); The deep-band placement increased early growth and P uptake above levels attained by the broadcast placement in some fields but not in others under no-till system in a Mollisols (Mallarino et al., 1999). Available P extracted by exchange anion resin was lower for broadcast treatments compared with deep-band treatments under a reduced-till system (Khatiwada et al., 2012). Deep band increased P availability in subsoil (15-25 cm) and broadcast in the soil surface (0-5 cm) under no till system in an Oxisol and Ultisol (Hansel et al., 2017a). Under natural precipitation conditions and no-till system there was not difference between deep band, broadcast, and band in soybean yield, however under a drought stress deep band P showed greater yields due to an increase on plants tolerance promoted by deeper root system growth (Hansel et al., 2017b).

Initial P fertilization correction (phosphating), little used in temperate soils, but well known and used in tropical soils is another management strategy to increase P availability for good crop production. Thus, the initial rates in an agricultural practice that aims to increase the P content available in the arable layer in soils with below available-critical levels, mostly used in tropical regions. This practice assumes that the most of P added from fertilizers is adsorbed on the soil colloids, especially on Fe and Al oxy-hydroxides, and that annual fertilizations with soluble phosphate will be subsequently more absorbed by the crops. Miranda & Miranda (2003) point out that the appropriate combination of initial corrective rates in total area and maintenance of annual rates has a great importance to promote crop productivity. However, the combination of P management in this correction/maintenance is still lacking in research, especially considering the rates and P placement methods.

The continuous phosphate fertilizer application over the cultivation time in amounts exceeding offtake in many soil of the world will promote accumulation of P in soils due to its higher adsorption capacity through the numerous forms already mentioned (Neset et al., 2008;

Sattari et al., 2012), building-up P levels (Pi and Po) in the soil, called residual P or legacy P (Condrón et al., 2013). Legacy soil P is considered the amount accumulated from the P inputs minus outputs over the time (Sattari et al., 2012, Rowe et al., 2016). Previous studies in the tropical soil (Coelho et al., 2017; Rodrigues et al., 2016; Withers et al. 2018) and temperate regions (Sattari et al., 2012; Rowe et al., 2016), suggest the great amount of legacy P might be more plant available than usually expected. However, few studies have evaluated the long-term effect of different P fertilizer placement and/or rates in the conventional tillage, no-till and strip-tillage, particularly the preferential forms of P retained in each soil pool and their contribution to increase P use efficiency by crops over time. To better understand soil P dynamics, it is necessary to know the different fractions of the element in the soil, throughout the sequential use of different extractive solutions, or other methodology if available. The knowledge of the nature and distribution of these fractions can provide important insight for comprehension of soil P availability (Rocha et al., 2005).

Chemical sequential extraction procedures have been and are still widely used to split extractable soil P into Pi and Po forms (Chang and Jackson, 1957; Hedley et al., 1982; Cross and Schlesinger, 1995). Sequential fractionation (Hedley et al., 1982) was developed to follow P transformation, to separate these various pools along a geochemical gradient, based on the sequential extraction of Pi and Po with different extraction forces, with a combination of alkali and acid reagents, resulting in the fragmentation of P in the soil, taking into account its different degree of lability. It has been applied to both tropical and temperate soils with considerable success (Cross and Schlesinger, 1995). In addition, it shows the distribution of P pools in labile, moderately and non-labile forms of Pi and Po in the soil (Condrón et al., 1985). Thus, this technique allows the observation of the dynamics of soil P pools and its cycling, provides information on the quantity and availability for plants, helping in the definition of better fertilizer management and appropriate soil use for each region/soil type.

For the above, the analysis of the interaction between P placement methods, P rates and soil components, as well as the verification of the transformations into soil P forms of distinct availability, allow us to optimize the use of phosphate fertilization and improve its agronomic efficiency in agricultural systems under distinct soil management (no-tillage, conventional, etc.).

Better understand the effects of P fertilizer placements and its impacts of long-term P accumulation in soils under different soils and management systems is necessary for establishing more sustainable P-use management strategies in agriculture. Thus, the general

hypothesis of this work is: phosphate fertilizer placement in long-term agricultural systems play a dominant role in P pools and change the soil P dynamics over the time.

The general objective of this study was to evaluate the effect of long-term P fertilizer placement (phosphating, band, deep-band, and broadcast) in three different management systems (conventional, reduced-tillage, and no-tillage) on changes in soil P pools (labile, moderately labile, and non-labile) and soil P legacy accumulation.

REFERENCES

- Abdala, D.B., Ghosh, A.K., da Silva, I.R., Novais, R.F., Alvarez Venegas, V.H., 2012. Phosphorus saturation of a tropical soil and related P leaching caused by poultry litter addition. *Agric. Ecosyst. Environ.* 162, 15–23.
- Achat, D.L., Bakker, M.R., Saur, E., Pellerin, S., Augusto, L., Morel, C., 2010. Quantifying gross mineralization of P in dead soil organic matter: testing an isotopic dilution method. *Geoderma* 158, 163–172.
- Adee, E., Hansel, F.D., Ruiz Diaz, D.A., Janssen, K., 2016. Corn response as affected by planting distance from the center of strip-till fertilized rows. *Front Plant Sci.* 7, 1232-1241.
- Ahmad, M., Ahmad, M., El-Naggar, A.H., Usman, A.R.A., Abduljabbar, A., Vithanage, M., Elfaki, J., Al-Faraj, A., Al-Wabel, M.I., 2017. Aging effect of organic and inorganic fertilizers on phosphorus fractionation in a calcareous sandy loam soil. *Pedosphere*. In press.
- Anghinoni, I., Barber, S.A., Phosphorus influx and growth characteristics of corn roots as influenced by phosphorus supply. *Agron. J.* 22, 685-688, 1980.
- Aray, Y., Sparks, D.L., 2007. Phosphate reaction dynamics in soils and soil minerals: a multiscale approach. *Adv. Agron.* 94, 135-179.
- Barreto, A.C., Fernandes, M. S., 2002. Produtividade e absorção de fósforo por plantas de milho em função de doses e modos de aplicação de adubo fosfatado em solo de tabuleiro costeiro. *Rev Bras Cien Solo*, 26:151-156, 2002.
- Bertrand, I., Holloway, R.E., Armstrong, R.D., McLaughlin, M.J., 2003. Chemical characteristics of phosphorus in alkaline soils from southern Australia. *Aust. J. Soil Res.* 41, 61–76.
- Chang, S.C., Jackson, M.L., 1957. Fractionation of soil phosphorus. *Soil Sci.* 84, 133-144.

- Childers, D.L., Corman, J., Edwards, M., Elser, J.J., 2011. Sustainability challenges of phosphorus and food: solutions from closing the human phosphorus cycle. *Bioscience*. 61, 117–124.
- Coelho, M.J.A., Aguiar, A.C.F., Sena, V.G.L., Moura, E.G., 2017. Utilization and fate of phosphorus of different sources applied to cohesive soil of Amazonian periphery. *Sci. Agric*. 74, 242-249.
- Condon, L.M., Turner, B.L., Cade-Menun, B., 2005. Chemistry and Dynamics of Soil Organic Phosphorus. *Phosphorus: agriculture and the environment*. Agron. Monogr. 46, 87-121.
- Condon, L.M., Goh, K.M., Newman, R.H., 1985. Nature and distribution of soil phosphorus as revealed by a sequential extraction method followed by ³¹P nuclear magnetic resonance analysis. *J. Soil Sci*. 36, 199-207.
- Conte, E., Anghinoni, I., Rheinheimer, D.S. 2003. Frações de fósforo acumuladas em Latossolo argiloso pela aplicação de fosfato no sistema plantio direto. *Rev. Bras. Cien. Solo*. 27, 893-900.
- Conte, E., Anghinoni, I., Rheinheimer D.S., 2002. Fósforo da biomassa microbiana e atividade de fosfatase ácida pela aplicação de fosfato em solo no sistema plantio direto. *Rev. Bras. Cien. Solo*. 26, 925-930.
- Cooper, J., Lombardi, R., Boardman, D., Carliell-Marquet, C., 2011. The future distribution and production of global phosphate rock reserves. *Resour. Conserv. Recycl*. 57, 78–86.
- Cordell, D. 2010. The Story of Phosphorus Sustainability implications of global phosphorus scarcity for food security. Ph.D thesis.
- Cornish, P., 2009. Research directions: improving plant uptake of soil phosphorus, and reducing dependency on input of phosphorus fertiliser. *Crop Pasture Sci*. 60, 190-196.
- Cross A.F., Schlesinger, W.H., 1995. A literature review and evaluation of the Hedley fractionation: applications to the biogeochemical cycle of soil phosphorus in natural ecosystems. *Geoderma*. 64, 197-214.
- Dalal, R.C., 1977. Soil organic phosphorus. *Adv Agron*. 29, 83-117.
- Devau, N., Cadre, E.L., Hinsinger, P., Jaillard, B., Gérard, F., 2010. Soil pH controls the environmental availability of phosphorus: Experimental and mechanistic modeling approaches. *Appl. Geochem*. 24, 2163-2174.
- Farmaha, B.S., Fernandez, F.G., Nafziger, E.D., 2012. Distribution of soybean roots, soil water, phosphorus and potassium concentrations with broadcast and subsurface-band fertilization. *Soil Sci. Soc. Am. J*. 76, 1079-1089.

- Hansel, F.D., Ruiz-Diaz, D.A., Amado, T.J.C., Rosso, L.H.M., 2017a. Deep banding increases phosphorus removal by soybean grown under no-tillage production systems. *Agron. J.* 109:1091-1098.
- Hansel, F.D., Amado, T.J.C., Ruiz-Diaz, D.A., Rosso, L.H.M., Nicoloso, F.T., Schorr, M., 2017b. Phosphorus fertilizer placement and tillage affect soybean root growth and drought tolerance. *Agron. J.* 109, 1-9.
- Ferreira, A., Vidal-Torrado, P., Otero, X.L., Macías, F. 2007. Are mangrove forest substrates sediments or soils? A case study in southeastern Brazil. *Catena.* 70, 79-91.
- Filippelli, G.M. 2011. Phosphate rock formation and marine phosphorus geochemistry: the deep time perspective. *Chemosphere.* 84, 759–66.
- Freeman J.S., Rowell, D.L., 1981. The adsorption and precipitation of phosphate onto calcite. *J. Soil Sci.* 32, 75–86
- Gatiboni, L.C., Kaminski, J., Heinheimer, D.S., Flores, J.P.C., 2007. Biodisponibilidade de formas de fósforo acumuladas em solo sob sistema plantio direto. *Rev. Bras. Cien. Solo.* 31, 691-699.
- Gatiboni, L. C. 2003. Disponibilidade de formas de fósforo do solo às plantas. Universidade Federal de Santa Maria, tese de doutorado, Santa Maria, 231p.
- Grant, C.A., Flaten, D.N., Tomasiewicz, D.J., Sheppard, S.C., 2001. A importância do fósforo no desenvolvimento inicial da planta. Piracicaba: Potafos, 95.
- Hedley, M., McLaughlin, M., 2005. Reactions of phosphate fertilizers and by-products in soils. In: Sims JT, Sharpley AN (Eds.), *Phosphorus: agriculture and the environment*, agronomy monograph., Madison, Wisconsin, pp. 181- 252
- Hedley, M.J., Stewart, J.W.B., Chauhan, B.S., 1982. Changes in inorganic and organic soil phosphorus fractions induced by cultivation practices and by laboratory incubations. *Soil Sci. Soc. Am. J.* 46, 970-976.
- Jasinski, S.M., 2018. Phosphate rock: mineral commodity summaries. Reston, VA: United States Geological Survey.
- Khaliwada, R., Hettiarachchi, G.M., Mengel, D.B. Fei, M., 2012. Speciation of phosphorus in a fertilized, reduced-till soil system: in-field treatment incubation study. *Soil Sci. Soc. Am. J.* 76, 2006-2018.
- Kruse, J., M. Abraham, W. Amelung, C. Baum, R. Bol, O. Kühn, H. Lewandowski, J. Niederberger, Y. Oelmann, C. Rüger, J. Santner, M. Siebers, N. Siebers, M. Spohn, J. Vestergren, A. Vogts, and P. Leinweber. 2015. Innovative methods in soil phosphorus research: A review. *J. Plant Nutr. Soil Sci.* 178, 43-88.

- Li, S.X., 1999. The current state and prospect of plant nutrition and fertilizer science. *Plant Nutr. Fert. Sci.* 5, 193-205
- Liu, J.Z., Li, Z.S., Li, J.Y., 1994. Utilization of plant potentialities to enhance the bio-efficiency of phosphorus in soil. *Eco-agric. Res.* 2, 16-23.
- Malavolta, E., 1981. *Manual de química agrícola: adubos e adubação*, third ed. São Paulo, São Paulo.
- Mallarino, A.P., Bordoli, J.M., Borges, R., 1999. Effects of phosphorus and potassium placement on early growth and nutrient uptake of no-till corn and relationships with grain yield. *Agron. J.* 91, 37-45.
- Martinazzo, R., Rheinheimer, D.S., Gatiboni, L.C., Brunetto, G., Kaminski, J., 2007. Fósforo microbiano do solo sob sistema plantio direto afetado pela adição de fosfato solúvel. *Rev. Bras. Cien. Solo.* 31, 563-568.
- McLaughlin, M.J., McBeath, T.M., Smernik, R., Stacey, S.P., Ajiboye, B., Guppy, C., 2011. The chemical nature of P accumulation in agricultural soils-implications for fertiliser management and design: an Australian perspective. *Plant Soil.* 349, 69-87.
- McLaren, T.I., Smernik, R.J., Simpson, R.J., McLaughlin, M.J., McBeath, T.M., Guppy, C.N., Richardson, A.E., 2017. The chemical nature of organic phosphorus that accumulates in fertilized soils of a temperate pasture as determined by solution ^{31}P NMR spectroscopy. *J. Plant Nutr. Soil Sci.* 180, 27-38.
- Menezes-Blackburn, D., Giles, C. Darch, T., George, T.S., Blackwell, M., Stutter, M., Shand, C., Lumsdon, D., Cooper, P., Wendler, R., Brown, L., Almeida, D.S., Wearing, C., Zhang, H., Haygarth, P.M., 2017. Opportunities for mobilizing recalcitrant phosphorus from agricultural soils: a review. *Plant Soil.* 427, 5-16.
- Nash, D.M., Haygarth, P.M., Turner, B.L., Condrón, L.M., McDowell, R.W., Richardson, A.E., Watkins, M., Heaven, M.W., 2014. Using organic phosphorus to sustain pasture productivity: a perspective. *Geoderma.* 221-222, 11-19.
- Neset, T.S.S., Bader, H.P., Scheidegger, R., Lohm, U., 2018. The flow of phosphorus in food production and consumption - Linköping, Sweden, 1870-2000. *Sci. Total Environ.* 396, 111-20.
- Novais, R.F., Smyth, T.J. 1999. *Fósforo em solo e planta em condições tropicais*. UFV, DPS, Viçosa, Minas Gerais.
- Novais, R.F., T.J. Smyth, and F.N. Nunes. 2007. Fosforo. In: R.F. Novais, V.V.H. Alvarez, N.F. Barros, R.L.F. Fontes, R.B. Cantarutti, and J.C.L. Neves, (Eds.) *Fertilidade do solo*. Sociedade Brasileira de Ciencia do Solo, Vicososa, Minas Gerais, pp. 471-550.

- Oelkers, E.H., Valsami-Jones, E., 2008. Phosphates mineral reactivity and global sustainability. *Elements*. 4, 83-87.
- Parfitt, R.L., 1978. Anion adsorption by soils and soil materials. *Adv. Agron.* 30, 1-46.
- Randall, G.W., Vetsch, J., 2008. Optimum placement of phosphorus for corn/soybean rotations in a strip-tillage system. *J. Soil Water Conserv.* 63, 152A–153A.
- Rashid, A., Awan, Z.I., Ryan, J., 2005. Diagnosing phosphorus deficiency in spring wheat by plant analysis: proposed critical concentration ranges, *Commun. Soil Sci. Plant Anal.* 36, 609-622.
- Rheinheimer, D.S., Gatiboni, L.C., Kaminski, J., 2008. Fatores que afetam a disponibilidade do fósforo e o manejo da adubação fosfatada em solos sob sistema plantio direto. *Cien. Rural.* 38, 576-586.
- Rheinheimer, D.S., Anghinoni, I., 2001. Distribuição do fósforo inorgânico em sistemas de manejo de solo. *Pesq. Agropec. Bras.* 36, 151-160.
- Rheinheimer, D.S., Anghinoni, I., Conte, E., 2000. Fósforo da biomassa microbiana em solos sob diferentes sistemas de manejo. *Rev. Bras. Cien. Solo*, 24, 589-597.
- Roberts, T.L., Johnston, A.E., 2015. Phosphorus use efficiency and management in agriculture. *Resour. Conserv. Recycl.* 105, 275-281.
- Rocha, A.T., Duda, G.P., Nascimento, C.W.A., Ribeiro, M.R., 2005. Fracionamento de fósforo e avaliação de extratores de P-disponível em solos da ilha de Fernando de Noronha. *Rev. bras. eng. agríc. ambient.* 2, 178-184.
- Rodrigues, M., Pavinato, P.S., Withers, P.J.A., Teles, A.P.B., Herrera, W.F.B., 2016. Legacy phosphorus and no tillage agriculture in tropical oxisols of the Brazilian savanna. *Sci. Total Environ.* 542, 1050-1061.
- Rowe, H., Withers, P.J.A. Baas, P., Chan, N.I., Doody, D., Holiman, J., Jacobs, B., Li, H., MacDonald, G.K., McDowell, R., Sharpley, A.N., Shen, J., Taheri, W., Wallenstein, M., Weintraub, M.N., 2016. Integrating legacy soil phosphorus into sustainable nutrient management strategies for future food, bioenergy and water security. *Nutr. Cycl. Agroecosyst.* 104, 393-412.
- Sato S., Solomon, D., Hyland, C., Ketterings, Q.M., Lehmann, J., 2005. Phosphorus speciation in manure and manure-amended soils using XANES spectroscopy. *Environ. Sci. Technol.* 39, 7485-7491.
- Sattari, S.Z., Bouwman, A.F., Giller, K.E., van Ittersum, M.K., 2012. Residual soil phosphorus as the missing piece in the global phosphorus crisis puzzle. *Proc. Natl. Acad. Sci.* 109, 6348-6353.

- Sharpley, A.N., 1985. Phosphorus cycling in unfertilized and fertilized agricultural soils. *Soil Sci. Soc. Am. J.* 49, 905-911.
- Shen, J., Yuan, L., Zhang, J., Li, H., Bai, Z., Chen, X., Zhang, W., Zhang, F., 2011. Phosphorus dynamics: from soil to plant. *Plant Physiol.* 156, 997-1005.
- Sleight, D.M., Sander, D.H., Peterson, G.A., 1984. Effect of phosphorus placement on the availability of phosphorus. *Soil Sci. Soc. Am. J.* 48, 336-340.
- Smil, V. 2000. Phosphorus in the environment: Natural flows and human interferences. *Annu. Rev. Energy Environ.* 25, 53–88.
- Song, C., Wang, E., Han, X., Stirzaker, R., 2011. Crop production, soil carbon and nutrient balances as affected by fertilisation in a Mollisol agroecosystem. *Nutr. Cycl. Agroecosyst.* 89, 363-374.
- Souza, R.F., Faquin, V., Andrade, A.T., TORRES, P.R.F., 2007. Formas de fósforo em solos sob influência da calagem e adubação orgânica. *Ver. Bras. Cien. Solo* 31, 1535-1544.
- Stutter, M.I., Shand, C.A., George, T.S., Blackwell, M.S.A., Dixon, L., Bol, R., MacKay, R.L., Richardson, A.E., Condon, L.M., Haygarth, P.M., 2015. Land use and soil factors affecting accumulation of phosphorus species in temperate soils. *Geoderma.* 257, 29–39.
- Theodorou, M.E., Plaxton, W.C., 1993. Metabolic Adaptations. *Plant Physiol.* 101, 339-344.
- Turner, B.L., Richardson, A.E., Mullaney, E.J., 2007. Inositol phosphates: linking agriculture and the environment. *CAB International.*
- Van Kauwenbergh, S.J., Stewart, M., Mikkelsen, R., 2013. World reserves of phosphorus rock... a dynamic and unfolding story. *Better crops.* 97, 18-20.
- Yang, X., Post, W.M., Thornton, P.E., Jain, A., 2013. The distribution of soil phosphorus for global biogeochemical modeling. *Biogeosciences.* 10, 2525-2537.
- Wang, E., Bell, M., Luo, Z., Moody, P., Probert, M.E. 2014. Modelling crop response to phosphorus inputs and phosphorus use efficiency in a crop rotation. *F. Crop. Res.* 155, 120-132.
- Wang, Y., Wang, E., Wang, D., Huang, S., Ma, Y., Smith, C.J., Wang, L., 2009. Crop productivity and nutrient use efficiency as affected by long-term fertilisation in North China Plain. *Nutr. Cycl. Agroecosyst.* 86, 105-119.
- Weihrauch, C., Opp, C., 2018. Ecologically relevant phosphorus pools in soils and their dynamics: The story so far. *Geoderma.* 325, 183-194.
- Withers, P.J.A., Rodrigues, M., Soltangheisi, A., Carvalho, T.S., Guilherme, L.R.G., Benitis, V.M., Gatiboni, L.C., Sousa, D.M.G., Nunes, R.S., Rosolem, C.A., Andreote,

F.D., Oliveira Jr, A., Coutinho, E.L.M., Pavinato., P.S., Transition to sustainable management of phosphorus in Brazilian agriculture. *Nature*. 8, 2537.

Zhu, J., Li, M., Whelan, M., 2018. Phosphorus activators contribute to legacy phosphorus availability in agricultural soils: A review. *Sci, Total Environ*. 612, 522-537.

2. PHOSPHORUS PLACEMENT AND RATES INFLUENCING SOIL LEGACY PHOSPHORUS IN A LONG-TERM COTTON/SOYBEAN ROTATION IN BRAZILIAN CERRADO

ABSTRACT

In weathered soils such as the Brazilian cerrado, phosphorus (P) tends to be strongly adsorbed by Fe and Al oxides and hydroxides, resulting in its low availability to plants. Such behavior demands an adequate P fertilizer management to change the P dynamics and positively affect the accumulation of inorganic (Pi) and organic (Po) labile fractions in the soil (legacy) for improving crops productivity. We aimed to assess changes in soil P pools (labile, moderately labile and non-labile) and legacy soil P accumulation after long-term P fertilization with an initial soil P buildup and annual P applications. A field study was conducted for twelve consecutive years (crop seasons from 2003/04 to 2014/15) under a conventional management with cotton and soybean rotation on an Oxisol. Treatments included an initial P fertilizer addition via broadcast application (phosphating) with five rates (0, 50, 100, 150 and 200 kg P₂O₅ ha⁻¹), and annual P fertilization with five rates (0, 30, 60, 90 and 120 kg P₂O₅ ha⁻¹) applied via band placement. After the last cotton harvest (2015), soil samples were collected from the 0-20 and 20-40 cm and analyzed for soil P pools via a sequential P fractionation scheme. Results of P pools showed a significant effect of P application on labile P fractions with annual rates of 60, 90 and 120 kg P₂O₅ ha⁻¹, while effects of the initial P application were less significant for this tropical high weathered soil. Thus, P accumulation from annual rates in the labile fractions will help to improve the soil legacy P availability and represent a profitable strategy to reduce large inputs of initial (phosphating) inorganic P fertilizer in tropical crop production systems.

Keywords: Initial rates; Annual rates; Conventional tillage; Phosphorus fractionation

2.1. INTRODUCTION

In weathered tropical acid soils, such as those in the Cerrado region, phosphorus (P) is often a primary factor limiting crop growth due to a low content of available P and high phosphate sorption capacity (Vance et al., 2003). The Oxisols cover more than 50% of the Cerrado region of Brazil (Sanchez and Cochrane, 1980), with general fine mineralogy constituted of kaolinite, gibbsite, goethite, and hematite (Fontes and Weed, 1991). In these conditions, P is predominantly high-energy adsorbed and/or precipitated with soil clay minerals. Thus, sorption reactions are therefore the key processes governing the availability of added P fertilizers, although precipitation reactions may also be important (McLaughlin et al., 2011). Therefore, high-input production systems are strongly dependent on inorganic P fertilizers (Dordas, 2009) with high addition of fertilizers every year to maintain crop yields. However, these non-renewable fertilizer sources are finite (Sattari et al., 2012) and there is a monopoly of phosphate reserves in the world in some specific countries (Cooper et al., 2011).

In a conventional soil management system, P deficiency on high P-sorbing soils might be corrected with high initial fertilizer rate application (phosphating) or many annually band applications (Younge and Plucknett, 1966); or by a combination of both (Yost et al., 1979). The initial phosphating may require large amounts to improve the soil P levels to a minimum value, while annual band applications replace gradually phosphate removed by crops and part fixed by the soil. Even though the results so far indicated that with initial and repeated band applications, yields and soil test levels increased (Raij and Diest, 1980; Smyth and Sanchez, 1980; Buresh et al., 1997). Yet, more information about the most efficient long-term rates, placement methods, and their influence under inorganic (Pi) and organic (Po) pool fractions are demanded, especially considering the interaction between labile and non-labile soil P pools and the residual effect (legacy P) of the phosphate fertilizer addition.

Soil P dynamics is complex, and both Pi and Po fractions are important to plant P supply over the crop cycle. Depending on the environmental conditions, soil mineralogy, fertilizer type and soil management, the soil can work as a source or sink of soluble P to the solution from the phosphate fertilizer applied (Novais and Smyth, 1999). Continuum application of soluble phosphates will change soil P dynamics and accumulate it in various forms (Shein et al., 2010), more predominantly in recalcitrant moderately and non-labile P pools in the Cerrado soils (Rodrigues et al., 2016). In addition, this knowledge will be useful to predict potential/better fertilizer sources for increasing P use efficiency and improving the management of cropping systems.

Procedure proposed by Hedley et al. (1982) fractionates the soil P into distinct inorganic and organic pools according to their soil/plant lability. This procedure is one of the most common tools to identify the redistribution of P applied into the soil. Through sequential extraction with a combination of alkali and acid reagents is possible to identify different forms, increasingly harsher treatments extract P fractions that are believed to be less available to plants, and it predict the distribution of labile and non-labile forms of Pi and Po between phosphate and minerals and organic soil components (Condon et al., 1985; Condon and Newman, 2011).

Based on the assumption that soluble P fertilizers should be applied always annually and locally at the establishment of each grain crop for obtaining a better fertilizer efficiency and keeps adequate soil P levels, we aimed in this study to evaluate the effect of long-term P fertilizer placement (initial rates and annual rates) in a conventional rotation of cotton and soybean production on changes in soil chemical properties, in soil P pools (labile, moderately labile and non-labile) and in soil legacy P accumulation.

2.2. MATERIALS AND METHODS

2.2.1. Site description and soil characterization

A long-term field experiment was conducted over twelve successive years (crop seasons from 2003/04 to 2014/15) on a soil classified as Rhodic Eutruxox (Soil Survey Staff, 2014), located in Pedra Preta, Mato Grosso, BR (16°37'23" S, 54°28'26" O, altitude 262 m above sea level) in the Cerrado biome. The regional climate is classified as Aw – Tropical, with mean annual precipitation of 1533 mm, with a well-defined rainy season from October to April and a dry season from May to September and mean annual temperature of 24.7 °C. This site was a native Cerrado (Haridasan, 2008) before the establishment of the experiment. The experiment comprised a conventional tillage system (plow and harrow annually) under commercial cotton and soybean production during the raining season with individual crops grown (nine years for cotton and three years for soybean) during the twelve-years period.

Initial soil was sampled in November of 2003, by collecting a representative sample from the 0–20 and 20–40 cm layers for the characterization of general soil properties (Table 1). The available P content (Mehlich-1), soil organic matter (SOM), pH (CaCl₂), exchangeable acidity (H+Al), aluminum (Al), potassium (K), calcium (Ca), magnesium (Mg) and aluminum saturation index (m) were determined according to the methodologies proposed by EMBRAPA (1997) and bases sum (BS) was obtained by K +, Ca + Mg (in mmol_c kg⁻¹), cation exchange capacity (CEC) was obtained by summing the BS + (H+Al) (in mmol_c kg⁻¹), base saturation index (V) was obtained by dividing the BS by CEC and multiplied by 100. Soil texture was determined by the pipette method of EMBRAPA (1997) after dispersion in 1 mol L⁻¹ NaOH. Soil bulk density was also measured according to EMBRAPA (1997).

The Al and Fe content by ditionite/citrate/bicarbonate (DCB) and by ammonium oxalate (Oxalate) were also determined in initial samples. The amorphous/crystalline Fe ratio (Fe Oxalate/Fe DCB) lower than 0.05 (0.038) indicates that the crystalline oxides are predominant in our soil (Table 1), characterizing it as advanced weathering stage (Kämpf et al., 2000).

Moreover, soil maximum P adsorption capacity (MPAC) and soil mineralogical composition by X-ray diffraction (XRD) were obtained from a sample collected in the control treatment (0-40 cm) after the last crop harvest (2015). MPAC was determined in soil subsamples of 2.5g shaken with eleven levels of added P (0, 4, 8, 12, 20, 28, 36, 44, 56, 68, and 80 mg L⁻¹) as KH₂PO₄ (p.a.), in a 25 mL equilibrium solution of calcium chloride (0.01 M CaCl₂.2H₂O). They were shaken for 24 h at 147 rpm rotation, and centrifuged for 15 min at

3278 g. The P concentration was measured in the supernatant by blue-molybdate colorimetric method (Murphy and Riley, 1962). MPAC was obtained after fitted by Langmuir equation, described by: $P_{so} = [k \times (P_{max} - P_{ce}) / (1 + K \times P_{ce})]$, where P_{so} is the amount of P sorbed in each soil sample, k is a constant related to the sorption energy, P_{max} is the maximum P sorption capacity, and P_{ce} is the equilibrium P concentration in solution. The estimated result of MPAC was 337 mg P kg⁻¹ of soil (Table 1), supported by the presence of Fe and Al sesquioxides in the mineralogical composition, as confirmed by X-ray diffraction (XRD).

Table 1 Initial soil chemical and physical properties for the surface layers (0-20 and 20-40 cm depth) before the experiment establishment.

Soil layer	P ⁽¹⁾	SOM ⁽²⁾	pH	H+Al ⁽³⁾	Al	K	Ca	Mg	BS ⁽⁴⁾	CEC ⁽⁵⁾	V ⁽⁶⁾	m ⁽⁷⁾		
cm	mg kg ⁻¹	g kg ⁻¹	CaCl ₂	----- mmolc kg ⁻¹ -----							%			
0-20	29	25	5.8	14	3	1.7	30	15	46.7	60.7	77	6		
20-40	10	20	5.1	20	1	2.3	17	8	27.3	47.3	58	4		
			Soil texture				<u>DCB⁽⁸⁾</u>		<u>Oxalate⁽⁹⁾</u>			<u>SBD⁽¹⁰⁾</u>	<u>MPAC⁽¹¹⁾</u>	
		<u>Sand</u>		<u>Silt</u>		<u>Clay</u>	Fe	Al	Fe	Al				
		----- g kg ⁻¹ -----										g cm ⁻³	mg kg ⁻¹	
0-20		671		26		303			25.7	6.06	0.97	*	1.27	377
20-40		653		22		324							1.25	

⁽¹⁾Phosphorus extracted by Mehlich-1; ⁽²⁾Soil organic matter; ⁽³⁾Exchangeable acidity; ⁽⁴⁾Sum of bases; ⁽⁵⁾Cation exchange capacity; ⁽⁶⁾Bases saturation index; ⁽⁷⁾Aluminum saturation index; ⁽⁸⁾Ditionite, citrate and bicarbonate; ⁽⁹⁾Acidic ammonium oxalate; ⁽¹⁰⁾Soil bulk density; ⁽¹¹⁾Phosphorus maximum adsorption capacity; *not determined.

To identify the main minerals present in the soil, XRD analysis of the clay fraction was run following the method described by Jackson (1979). A set of different post-treatments were prepared for each soil clay sample after removing Fe oxides. The post-treatments consisted of saturate the clay fraction with 1 M KCl or 1 M MgCl₂ in order to saturate inter-layers and then allowing to identify the phyllosilicates. The following post-treatments were applied: K saturation at room temperature (25°C); K saturation and heating up to 300°C; K saturation and heating up to 500°C; Mg saturation at room temperature (25°C); Mg saturation with solvation using ethylene glycol. Another clay sample with Fe was prepared to identify the Fe oxides, followed by the oriented glass slide preparation prior to analysis. The diffractograms were collected in a Miniflex II Desktop X-Ray Diffractometer (RIGAKU), CuK α radiation, analysis interval from 10 to 50°2 θ to the clay fraction without Fe samples and 5 to 30°2 θ to the clay samples with Fe. The results (Fig. 1) show the dominance of Kaolinite as the only phyllosilicate and the presence of the Hematite and Goethite as Fe oxides and Gibbsite as Al hydroxide in the soil.

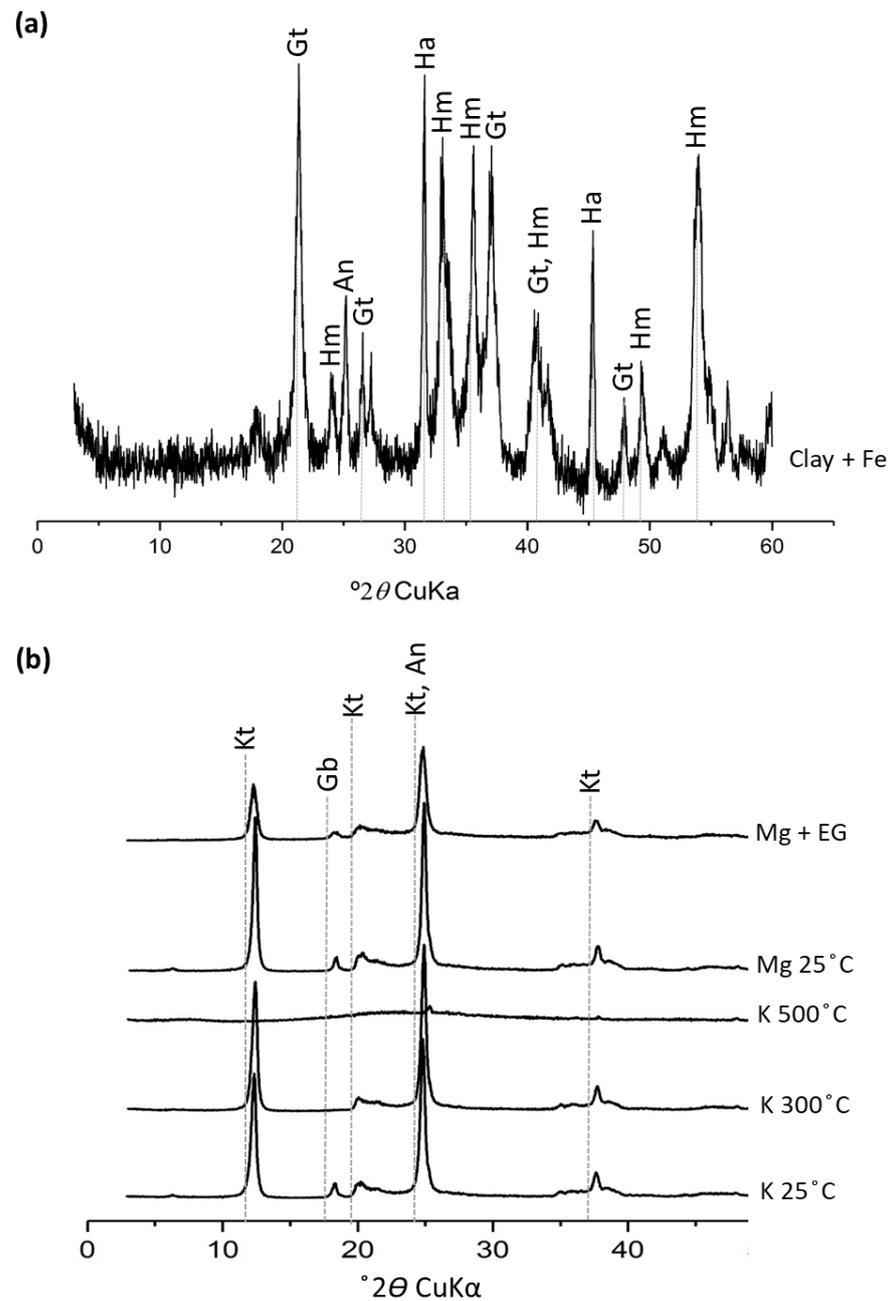


Fig. 1. X-ray diffraction of total clay: (a) Clay + Fe and (b) clay free of Fe [K^+ at room temperature (K 25 $^{\circ}\text{C}$), heated 300 $^{\circ}\text{C}$ (K 300 $^{\circ}\text{C}$) and 500 $^{\circ}\text{C}$ (K 500 $^{\circ}\text{C}$); Mg^{2+} at room temperature (Mg 25 $^{\circ}\text{C}$) and solvated with ethylene glycol (Mg + EG)], oriented blades. Identified minerals: Anatase (An), Gibbsite (Gb), Goethite (Gt), Hematite (Hm) and Kaolinite (Kt).

2.2.2. Experimental design and management practices

The experimental design was a randomized block with a factorial combination of five rates of initial phosphate application (initial P - phosphating), and five rates of annual added P (annual P rates) for a total of 25 treatments and four replications. Individual plots size was 9 x 40 m with a 0.8 m alley between plots. The initial P rates were 0, 50, 100, 150 and 200 kg P₂O₅ ha⁻¹ and the annual P rates were 0, 30, 60, 90 and 120 kg P₂O₅ ha⁻¹. The initial P treatments were broadcast incorporated in the soil by disk plowing (plowing at 15-20 cm depth) at the establishment of the experiment in 2003 and the annual rates of P were band-applied at 5 cm deep and 5 cm to the side (5 by 5) of the seed with the planter for both crops. Conventional tillage with disc harrowing was performed before the planting cotton/soybean and application of phosphate fertilizer for all treatments at each plot every year. Cotton and soybean were planted in the in the same row for each year.

Th The field area was limed before initiating the study in 2001 with 1.0 Mg ha⁻¹. Another 1.2 and 2.0 Mg ha⁻¹ of lime was applied in 2003 and 2006. Moreover, in 2007 and 2011 it was applied 2 and 1.5 Mg ha⁻¹ of lime. Every lime application was immediately incorporated in the soil by disk plowing (plowing at 15-20 cm depth).

The P fertilizer source used for every treatment was triple superphosphate (TSP: 46% soluble/total P₂O₅ and 10% Ca). Complementary fertilization in all cotton-growing years were 135 kg N ha⁻¹ from urea and 120 kg K₂O ha⁻¹ from potassium chloride. Also for the years in which soybean was cultivated, 90 kg K₂O ha⁻¹ from potassium chloride was applied. Planting was made from November to December and harvest from June to July for cotton, and planting in October and harvest in January for soybean.

2.2.3. Soil sampling analysis and phosphorus fractionation

After After the last cotton harvest in August of 2015 (twelve years of conventional cotton and soybean cultivation), a composite soil sample of eight soil cores was collected from the 0-20 and 20-40 cm depth from each plot. Sequential P fractionation was run following the methodology proposed by Hedley et al. (1982) with modifications by Condon et al. (1985). This technique uses chemical extractants sequentially in the same sample to remove progressively from the most available to the most stable inorganic (Pi) and organic (Po) P pools. At each step, 10 mL of extractant was added to 0.5 g soil in 15 mL centrifuge tubes (1:20 soil:solution ratio) and the tubes were shaken end-over-end (vertical shaker, 60

rpm) for 16 h at 25 °C. Soil samples were extracted sequentially using: (i) Anion exchange resin (AER, 2.0 cm² in area) for inorganic P (P_{AER}) readily diffusing into the soil solution; (ii) 0.5 M NaHCO₃ to extract inorganic P (P_{Bic}) weakly adsorbed on the surface of crystalline compounds and organic P (P_{OBic}) compounds with low recalcitrance like ribonucleic acid and glycerophosphate (Tiessen and Moir, 1993); (iii) 0.1 M NaOH to extract inorganic P ($P_{\text{I0.1Hid}}$) strongly adsorbed onto Fe and Al and clay minerals (Hedley et al., 1982), and organic P ($P_{\text{O0.1Hid}}$) mainly associated with fulvic and humic acids adsorbed onto mineral and SOM surfaces (Linguist et al., 1997); (iv) 1 M HCl to extract inorganic P (P_{HCl}) associated with apatite, other sparingly-soluble Ca-P compounds or negatively charged oxide surfaces (Gatiboni et al., 2007); (v) 0.5 M NaOH to extract inorganic P ($P_{\text{I0.5Hid}}$) associated with Fe and Al and clay minerals, and organic P ($P_{\text{O0.5Hid}}$) associated with fulvic and humic acids inside aggregates (Condon et al., 1985). After each extraction, the soil suspensions were centrifuged at 3278 g for 30 min. After centrifugation and removal of the supernatant at each step, the soil was washed with 10 mL 0.5 M NaCl solution to prevent the residual effect of the previous solution (pH change). The (vi) residual P (P_{Residual}) after drying the remaining soil at 40°C for 3 days, was determined by H₂SO₄ + H₂O₂ + MgCl₂ digestion (Brookes and Powlson, 1981), this fraction consists of occluded P forms (Cross and Schelinger, 1995).

The alkaline extracts were added to a 1:1 sulfuric acid solution and 7% ammonium persulfate in an autoclave at 121 °C and 103 kPa for 2 h to determine the P total of each fraction, Pt (Pi + Po) (USEPA, 1971). The amount of Pi in the alkaline extracts (P_{Bic} , $P_{\text{I0.1Hid}}$ and $P_{\text{I0.5Hid}}$) was determined using the colorimetric method of Dick and Tabatabai (1977) at 660 nm wavelength. Inorganic P in each of the acid extract (P_{AER} , P_{HCl} , P_{Residual} and digestion of P_{Bic} , $P_{\text{I0.1Hid}}$ and $P_{\text{I0.5Hid}}$) were determined following the colorimetric method of Murphy and Riley (1962) at 882 nm wavelength. Organic P in each alkaline extract (P_{OBic} , $P_{\text{O0.1Hid}}$ and $P_{\text{O0.5Hid}}$) was determined by the difference between the total P (Pt) and Pi of each respective fraction.

Soil P pools were quantified based on the lability predicted by the extractors (Cross and Schlesinger, 1995). Labile P (LP) consisted of the P_{AER} plus the P_{Bic} (Pi and Po), the moderately labile P (MLP) consisted of the $P_{\text{I0.1Hid}}$ (Pi and Po) plus the P_{HCl} , and the non-labile P (NLP) consisted of the $P_{\text{I0.5Hid}}$ (Pi and Po) plus the P_{Residual} .

2.2.4. Crop productivity, phosphorus efficiency and soil legacy P

Cotton and soybean were harvested using a plot combine, and grain samples collected and analyzed for total P content each year. The values of yield and P content were used to estimate total P removal during the twelve-year period. Phosphorus agronomic efficiency (PAE) was estimated according to Fageria (2001), by relating the cumulative yield, respectively, and the total applied P via fertilizer as follows: $PAE = (Y_f - Y_o) / Q_f$, where PAE is the P agronomic efficiency (kg grain kg⁻¹ of P applied), Y_f is the cumulative yield with fertilized P rate (kg grain ha⁻¹), Y_o is the cumulative yield for the unfertilized control treatment and Q_f is the accumulated P fertilizer rate (kg P ha⁻¹).

The difference between the total P inputs from fertilizers and outputs from crop removal (assuming a 4.0 kg P Mg⁻¹ for cotton (core+fiber) and 4.6 kg P Mg⁻¹ for soybean, data from the Fundação-MT, responsible for the field trial) was used to estimate the P balance. Legacy P was estimated through the amount of total labile P, moderately labile P and non-labile P (kg P ha⁻¹) of each treatment minus the control treatment. The values of P (mg kg⁻¹) at each depth interval (0-20 and 20-40 cm) were added after conversion to kg ha⁻¹ as described in Coelho et al. (2017).

2.2.5. Data analysis

All All statistical analyses were completed in SAS Studio (version 9.4; SAS, Cary, NC). Graphs and regressions were plotted by Sigma Plot (Systat Software, San Jose, CA). Variance homogeneity and normality of data distribution were tested for each parameter before the analysis of variance (ANOVA). The data were submitted to ANOVA using the PROC GLM procedure to test the effect of the phosphate fertilizer rates (initial rates and annual rates of P) on soil P fractions, crop productivity and P agronomic efficiency. When significant, means were compared by the Tukey test, $P < 0.05$, and the annual rates effect was adjusted to linear or quadratic regression model for crop productivity and P agronomic efficiency.

2.3. RESULTS

2.3.1. Soil phosphorus fractions

In general, after twelve-years period, the proportion of all P fractions from the Hedley's fractionation were not significantly affected by the interaction of initial rates \times annual rates of P. However, greater P rates had direct relationship with quantity and distribution of P fractions, mainly for inorganic fractions, and there were significant effects from both initial and annual rates of P, but no interaction between them (Supplementar Table 1).

2.3.1.1. Labile P fractions (P_{AER} , P_{iBic} and P_{oBic})

Overall, Overall, the labile P pool (LP) represents only 6-13 % of the total P (P_{Total}) in the soil profile (0-40 cm) under initial and annual P rates applied (Table 2, Fig. 3). However, the amount of LP was higher in the soil surface (0-20 cm, ~12%) than in the 20-40 cm soil layer (~ 6%). Concentrations of P_{AER} (~36% of LP) and P_{iBic} (~30% LP) in the soil surface (0-20 cm) were significantly affected by initial and annual P rates with the same tendency over all P rates, also P_{oBic} (~ 33% of LP) was affected by annual P rates (Supplementar Table 1 and Table 2). The greatest mean concentration for P_{AER} and P_{iBic} was observed for the 200 kg P_2O_5 ha^{-1} initial rate. Moreover, P_{AER} and P_{iBic} were accumulated in the soil based on the following order for annual rates: 120 > 90 > 60 > 30 > 0 kg P_2O_5 ha^{-1} (Table 2). The smallest concentration of P_{oBic} was observed for 120 kg P_2O_5 ha^{-1} as annual rate, but it differ only from 30 and 60 kg P_2O_5 ha^{-1} (Table 2).

In the 20-40 cm soil layer the LP was less sensitive than in the surface soil. However, the P_{AER} (~25% of LP) was still affected by initial and annual rates of P with the highest concentration observed for 200 kg P_2O_5 ha^{-1} compared to the control. Also, the ranking for mean accumulation based on annual rates was 120 > 90 = 60 > 30 = 0 kg P_2O_5 ha^{-1} (Supplementar Table 1 and Table 2). In addition, P_{iBic} (~38% of LP) was evenly affected by annual P rates and its ranking for mean soil concentration was 120 > 90 = 60 > 30 = 0 kg P_2O_5 ha^{-1} (Supplementar Table 1 and Table 2). Nonetheless, the P_{oBic} (~31% of LP) was not changed by both initial and annual rates of P (Supplementar Table 1).

Table 2. Inorganic P extracted by anion exchange resin and by 0.5 M NaHCO₃, and organic P extracted by 0.5 M NaHCO₃ in two depths twelve-years cultivation period as affected by initial and annual rates of P soil layer after

Initial rates (kg P ₂ O ₅ ha ⁻¹)	Annual P fertilizer rates (kg P ₂ O ₅ ha ⁻¹)											
	0 - 20 cm depth						20 - 40 cm depth					
	0	30	60	90	120	Mean	0	30	60	90	120	Mean
--- Inorganic P extracted by anion exchange resin (P _{AER} , mg kg ⁻¹) ---												
0	5.6	9.6	17	22	24	16b	1.6	1.8	3.0	5.0	8.5	3.9b
50	5.9	13	19	26	26	18ab	1.4	2.8	4.7	6.6	7.8	5.0ab
100	5.3	12	18	22	26	17ab	1.7	3.2	5.0	4.2	8.8	4.5ab
150	6.2	11	17	22	27	17ab	1.8	3.3	5.8	6.6	5.7	4.6ab
200	6.8	14	21	23	27	18a	2.5	3.2	7.7	6.4	8.6	5.7a
Mean	6.0E	12D	18C	23B	26A		1.8C	2.8C	5.2B	5.7B	8.1A	
--- Inorganic P extracted by 0.5 M NaHCO ₃ (P _{Bic} , mg kg ⁻¹) ---												
0	5.9	5.9	13	17	23	13b	3.2	4.2	5.2	7.6	12	6.5ns
50	4.6	9.5	15	18	20	13ab	4.5	4.7	5.7	8.8	18	8.3
100	6.4	8.3	15	18	24	14ab	3.6	4.5	7.9	6.5	13	7.2
150	4.7	8.4	12	17	23	13ab	3.0	5.2	5.8	8.9	7.0	5.9
200	6.3	13	18	21	22	16a	3.5	4.8	12.0	7.9	12	8.1
Mean	5.5E	8.9D	15C	18B	22A		3.2C	4.6C	7.3B	7.9B	12.6A	
--- Organic P extracted by 0.5 M NaHCO ₃ (P _{O_{Bic}} , mg kg ⁻¹) ---												
0	10	13	18	20	11	15ns	5.9	3.3	4.4	3.8	7.7	5.0ns
50	20	18	14	14	12	15	3.9	4.3	5.6	6.9	6.6	5.4
100	16	21	17	15	14	17	4.0	5.3	5.0	4.8	5.7	4.9
150	15	16	16	16	10	15	4.4	6.8	6.2	4.9	5.6	5.6
200	15	17	15	14	14	15	5.8	5.1	6.6	5.3	7.3	6.0
Mean	15AB	17A	16A	16AB	12B		4.8ns	4.9	5.5	5.1	6.6	

Within each depth and P fraction, means values followed by the same capital letter in line and tiny in column were not significantly different by Tukey test ($P < 0.05$). ns, not significant

2.3.1.2. Moderately labile P fractions ($P_{i0.1Hid}$, $P_{o0.1Hid}$, and P_{HCl})

The moderately labile P pool (MLP) represents 17-20% of the P_{Total} in the soil profile (0-40 cm) under initial and annual rates of P applied (Table 3), with the amount of Pi in top surface higher than in the subsoil, although highest contribution by $P_{o0.1Hid}$ and less contribution by P_{HCl} . The $P_{o0.1Hid}$ (~61% of MLP, Fig. 3) was not changed by both initial and annual rates of P in the soil surface, but was more concentrated in the 20-40 soil layer (~87% of MLP) and was affected significantly by initial and annual rates of P at this depth (Supplementar Table 1). The greatest concentration of $P_{o0.1Hid}$ was observed for the 200 kg P_2O_5 ha⁻¹ for initial rates, and the ranking of mean accumulation was $120 = 60 = 90 \geq 30 = 0$ kg P_2O_5 ha⁻¹ for the annual rates of P (Table 3). The $P_{i0.1Hid}$ (~31% of MLP) was also affected by initial and annual rates of P in the 0-20 cm layer, with the greatest concentration for initial rates under 200 kg P_2O_5 ha⁻¹ and the ranking for the annual rates was $120 \geq 90 = 60 \geq 30 = 0$ kg P_2O_5 ha⁻¹ (Supplementar Table 1 and Table 3). Moreover, P_{HCl} (~8% of MLP) in the same layer (0-20 cm) was similarly affected by initial and annual rates of P with greater mean concentration obtained with the 200 kg P_2O_5 ha⁻¹ for initial rates and the ranking of mean accumulation in the annual rates was a little different: $120 = 90 = 60 > 30 = 0$ kg P_2O_5 ha⁻¹ (Table 3). In addition, the $P_{i0.1Hid}$ (~11% of MLP) and P_{HCl} (~2% of MLP) in the 20-40 soil layer were affected just by annual rates of P with the ranking of mean accumulation: $120 \geq 60 = 30 = 0$ kg P_2O_5 ha⁻¹ and $120 > 90 = 60 = 30 = 0$ kg P_2O_5 ha⁻¹, respectively (Supplementar Table 1 and Table 3).

2.3.1.3. Non-labile P fractions ($P_{i0.5Hid}$, $P_{o0.5Hid}$ and $P_{Residual}$)

Cumulative rates of P contributed for the greatest concentration of non-labile P pools (NLP, 67-75% of the P_{Total}) in the soil profile (0-40 cm) under initial and annual rates applied (Table 4, Fig. 3). However, just the $P_{o0.5Hid}$ (~8% of NLP) was affected significantly by initial and annual rates of P and only in the 0-20 cm soil layer (Supplementar Table 1). The greater mean concentration of $P_{o0.5Hid}$ was observed for 50 kg P_2O_5 ha⁻¹ initial rate and its ordering of mean accumulation for annual rates was $120 > 90 = 60 = 30 = 0$ kg P_2O_5 ha⁻¹ (Table 4).

Table 3. Inorganic P extracted by 0.1 M NaOH, organic P extracted by 0.1 M NaOH and inorganic P extracted by 1 M HCl in two depths soil layer after twelve-years cultivation period as affected by initial and annual rates of P.

Initial rates (kg P ₂ O ₅ ha ⁻¹)	Annual P fertilizer rates (kg P ₂ O ₅ ha ⁻¹)											
	0 - 20 cm depth						20 - 40 cm depth					
	0	30	60	90	120	Mean	0	30	60	90	120	Mean
- - - Inorganic P extracted by 0.1 M NaOH (P _{i0.1Hid} , mg kg ⁻¹) - - -												
0	12	21	23	29	23	22b	4.3	5.0	5.7	5.6	6.6	5.4ns
50	15	20	21	27	24	21b	5.3	4.3	4.6	5.6	7.8	5.5
100	15	20	20	22	35	22b	4.6	5.0	5.3	5.6	6.4	5.3
150	15	16	23	21	32	21b	4.5	4.3	5.4	4.9	4.5	4.7
200	23	27	30	28	27	27a	3.8	4.5	5.4	5.5	6.2	5.1
Mean	16C	21BC	23B	25AB	28A		4.5B	4.6B	5.2B	5.4AB	6.3A	
- - - Organic P extracted by 0.1 M NaOH (P _{o0.1Hid} , mg kg ⁻¹) - - -												
0	48	43	58	36	48	47ns	35	32	36	39	46	38b
50	37	43	48	44	43	43	32	36	42	47	58	43ab
100	41	34	39	48	41	41	34	38	46	39	51	41ab
150	37	41	51	54	50	47	36	42	49	50	40	43ab
200	35	35	42	42	55	42	41	42	50	42	53	46a
Mean	40ns	39	48	45	48		35C	38BC	44A	43AB	49A	
- - - Inorganic P extracted by 1 M HCl (P _{HCl} , mg kg ⁻¹) - - -												
0	2.9	3.8	6.1	6.1	5.5	4.9b	0.7	0.6	0.9	0.6	0.8	0.7ns
50	3.6	5.1	6.9	6.7	6.2	5.7ab	0.4	0.6	0.7	0.9	1.5	0.8
100	3.6	4.8	6.4	7.1	7.2	5.8ab	0.5	0.7	0.8	0.7	0.9	0.7
150	3.3	4.4	6.7	6.5	8.1	5.8ab	0.6	0.8	0.8	0.9	0.7	0.7
200	4.6	5.5	7.1	7.4	6.6	6.2a	0.8	0.7	1.1	0.7	1.0	0.8
Mean	3.6B	4.7B	6.6A	6.7A	6.7A		0.6B	0.7AB	0.8AB	0.7AB	0.9A	

Within each depth and P fraction, means values followed by the same capital letter in line and tiny in column were not significantly different by Tukey test ($P < 0.05$). ns, not significant.

Table 4. Inorganic P extracted by 0.5 M NaOH, organic P extracted by 0.5 M NaOH and Residual P in two depths soil layer after twelve-years cultivation period as affected by initial and annual rates of P.

Initial rates (kg P ₂ O ₅ ha ⁻¹)	Annual P fertilizer rates (kg P ₂ O ₅ ha ⁻¹)											
	0 - 20 cm depth						20 - 40 cm depth					
	0	30	60	90	120	Mean	0	30	60	90	120	Mean
- - - Inorganic P extracted by 0.5 M NaOH (P _{i0.5Hid} , mg kg ⁻¹) - - -												
0	28	20	29	32	27	27ns	18	20	25	20	18	20ns
50	33	25	28	28	27	28	20	20	22	16	18	19
100	30	32	28	27	27	29	16	20	17	21	18	18
150	31	27	30	27	30	29	19	20	19	26	20	21
200	32	28	28	32	29	30	20	21	21	20	23	21
Mean	31ns	26	28	29	28		18ns	20	21	21	19	
- - - Organic P extracted by 0.5 M NaOH (P _{o0.5Hid} , mg kg ⁻¹) - - -												
0	15	23	22	25	26	22ab	7.9	13	4.5	6.3	10	8.3ns
50	12	30	26	24	26	24a	3.8	6.7	6.4	12	17	9.2
100	14	19	17	26	24	20ab	7.8	12	13	5.4	12	10
150	10	17	27	23	32	22ab	8.1	10.0	10	8.4	7.1	8.8
200	10	15	20	20	22	17b	7.8	8.1	12	8.2	7.0	8.7
Mean	12B	21A	22A	23A	26A		7.1ns	9.9	9.4	8.1	10	
- - - Residual P (P _{Residual} , mg kg ⁻¹) - - -												
0	204	205	217	208	219	211ns	178	166	171	175	169	172ns
50	211	211	216	225	215	215	174	180	166	168	164	170
100	210	234	217	216	208	217	167	176	181	168	176	173
150	194	206	224	235	222	216	172	165	168	169	163	167
200	215	216	218	220	204	215	174	199	187	189	173	184
Mean	207ns	214	218	221	214		173ns	177	174	174	169	

Within each depth and P fraction, means values followed by the same capital letter in line and tiny in column were not significantly different by Tukey test ($P < 0.05$). ns, not significant.

2.3.2. Crop productivity, agronomic efficiency and soil legacy P

Average Average cumulative yield across the twelve-years period of cotton and soybean cultivation was not influenced by initial rates of P (Fig. 2A) but was affected by annual rates (Fig. 2B). Moreover, it was increased with the fertilizer rates in annually banded application with a peak of maximum yield at 97 kg P₂O₅ ha⁻¹ with a mean yield value of 4140 kg ha⁻¹. The same behavior was observed for P agronomic efficiency (PAE) that was not influenced by initial rates of P (Fig. 2C) but was negatively affected by annual rates of P (Fig. 2D) where lower rates showed proportionally higher PAE, with the greatest mean value of 1.05 kg P kg⁻¹ for the lowest rate (30 kg P₂O₅ ha⁻¹).

Overall, our results have shown no interaction between initial rates and annual rates of P for all parameters analyzed in this study. So that, the balance (legacy P) and changes on the soil P pools (LP, MLP, and NLP) after twelve-years period was isolated by initial and annual rates of P (Fig. 3). Furthermore, the legacy P had accumulated at both initial and annual rates with the greatest amount showing proportionally under higher P inputs, but outputs were not affected (Fig. 3A and 3B). However, there was a soil P mining under the smallest rates annually applied (0 and 30 kg P₂O₅ ha⁻¹), with a depletion of -218 and -74 kg P ha⁻¹, respectively (Fig. 3B). The greatest positive balance were founded for the 60, 90 and 120 kg P₂O₅ ha⁻¹ annual rates (Fig. 3D). Initial P rates had no effect on legacy P overall (Fig. 3C). Also, LP and MLP increases according to the increase of annual rates but for NLP was the opposite, decreasing as annual P rates increased (Fig. 3D).

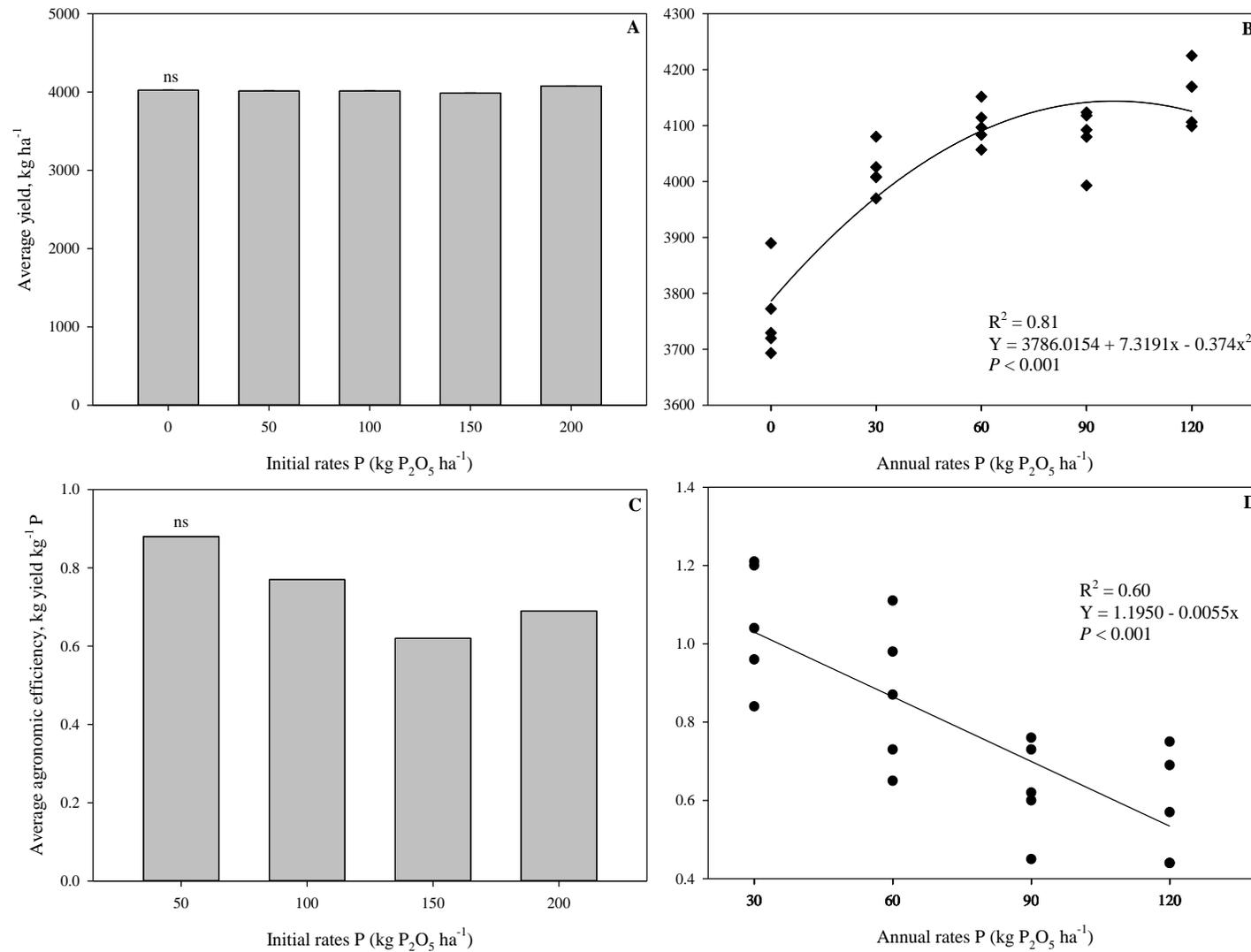


Fig. 2. Average cumulative yield and agronomic efficiency of cotton and soybean after twelve years as affected by initial rates (A and C) and annual rates of P (B and D). Mean values followed by the same letter do not differ by Tukey test ($P < 0.05$). ns, not significant.

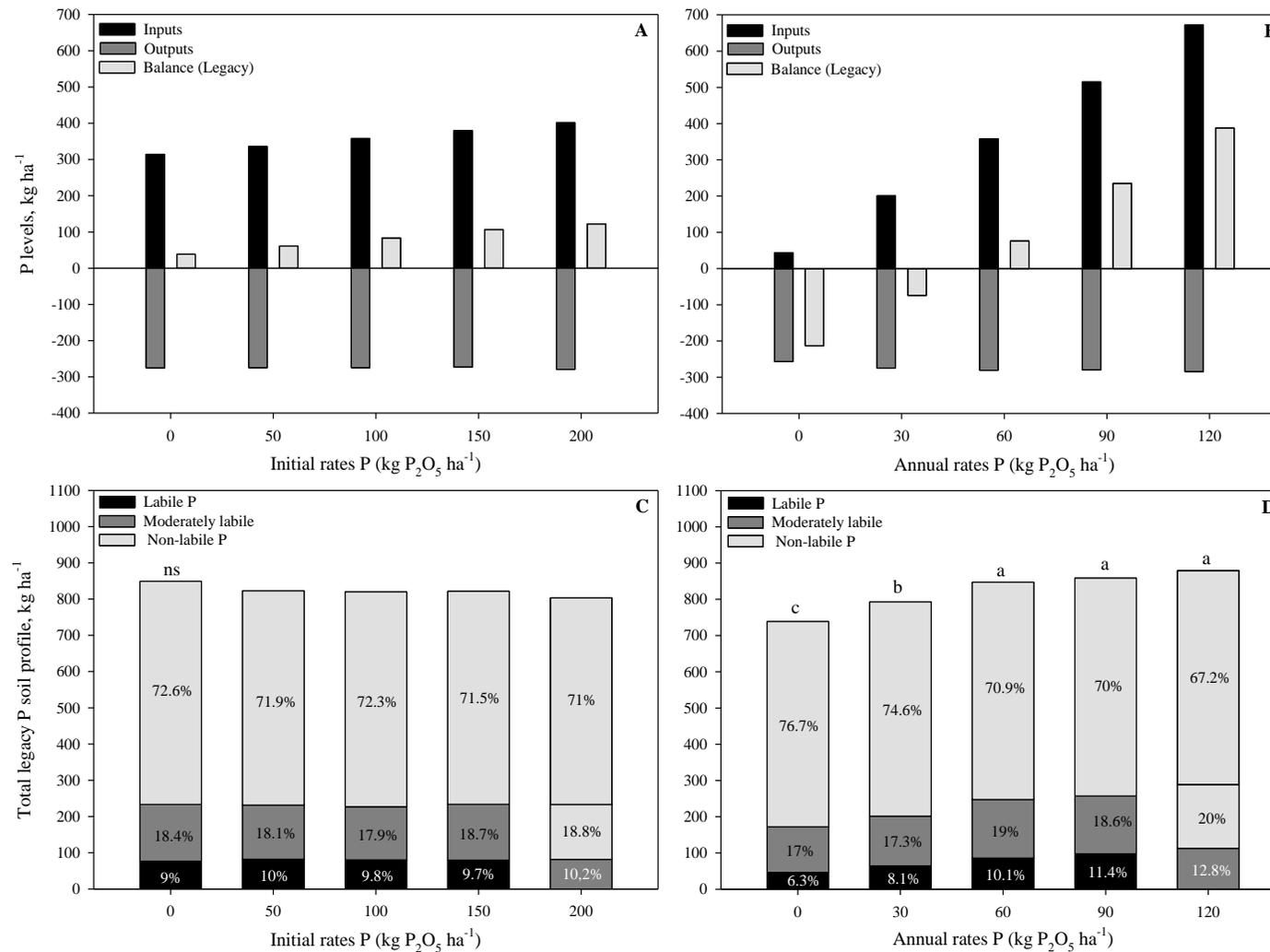


Fig. 3. Legacy soil P after twelve years of cotton and soybean: P inputs, outputs and balance during twelve year period of study as affected by initial rates of P (A) and by annual rates of P (B); Changes in total legacy soil P in soil profile (0-40 cm) as a function of labile P, moderately labile P and non-labile P as affected by initial rates of P (C) and by annual rates of P (D); Mean values followed by the same letter do not differ by Tukey test ($P < 0.05$). ns, not significant.

2.4. DISCUSSION

2.4.1. Labile P fractions – LP (P_{AER} , P_{iBic} and P_{oBic})

The P fertilizer rates (initial and annual) under conventional tillage had an expressive effect on P dynamics in the soil surface after twelve years, more pronounced in MLP and NLP pools than in LP pool. These results agree in some way to previous studies in a clayey Rhodic Hapludox, in which it was reported an increase of only 5-7% in LP pool after 3 years of P fertilizer application (Teles et al., 2017), but 5-10% increase in total P after 6 years of P fertilization (Soltangheisi et al., 2018). Otherwise, Herrera et al. (2016) reported nearly 14% increase in LP pool when studying cumulative rates of P fertilizer over four years cultivation in a sandy clay loam Hapludox. The dominance of less labile P accumulation in most Brazilian soils is due to the high capacity of these weathered soils (Oxisols), dominated by kaolinite and Al-/Fe- oxides (Fontes and Weed, 1991; Schaefer et al., 2008), to retain P by high specificity and strong adsorption reactions (Cross and Schlesinger, 1995; Fink et al., 2016). This sense can explain the low LP pool and the higher accumulation of total P in the soil surface without mobility in soil profile, since our soil is predominantly constituted by kaolinite, gibbsite, goethite and hematite (Fig. 1). Yet, usually there is high amount of LP (P_{AER} , P_{iBic} and P_{oBic}) in the surface soil (0-20 cm) and lower levels of LP in the subsoil (20-40cm) for Cerrado soils (Sousa and Lobato, 2004), as observed here.

The greatest LP concentrations observed in 0-20 cm under higher initial P rate for both P_{AER} and P_{iBic} may be attributed to P saturation by fertilizer band placement, influencing consequently its distribution in the soil profile. Since the adsorption sites are gradually saturated, binding energy of P solubilized later is weakly adsorbed and consequently increase P availability (Rheinheimer et al., 2003). The same behavior can be attributed for levels of P for P_{AER} and P_{iBic} under initial and annual rates of P in the subsurface (20-40 cm). However, the continuous application of soluble P fertilizer via broadcast in a conventional system provides more contact between soil-fertilizer, which enhances P sorption reactions, what explains the small influence of the initial rates in readily available and inorganic labile P levels in soil surface (0-20 cm). Also, soils under intensive physical disturbance by plowing and disc harrowing, such our case, are more susceptible to SOM decomposition, consequently exposing potential P adsorption sites present Fe oxide surfaces (Tiecher et al., 2018).

Greater levels of organic P in the soil surface layers are usually positive for plant absorption (Prasad et al., 2017). Our results of $P_{O_{Bic}}$ were from 12 to 17 mg kg⁻¹ (Table 3), representing ~30% of LP, what is a reasonable amount for supplying plant demand, although the greatest annual P rate has depleted a little this fraction over the time of cultivation. Thus, in a continuum addition of phosphate fertilizers both organic and inorganic forms of P have similar ability to provide P for the plants (Gatiboni et al., 2007; Bunemann et al., 2006; Daroub et al., 2000). Although Daroub et al. (2000) reported that inorganic LP pool participation is more expressive for plant absorption than organic LP.

2.4.2. Moderately labile P fractions – MLP ($P_{i0.1Hid}$, $P_{o0.1Hid}$ and P_{HCl})

The The higher rates of initial and annual P fertilizer application increased the concentration of inorganic MLP in both layers of our soil profile (Table 4). Still, the greater concentrations found for P_{HCl} analogue to the higher phosphate rates may come from calcium phosphates present in fertilizer (10% of Ca). Also, P_{HCl} is usually a very small fraction of P in strongly weathered soils from tropical regions (Pavinato et al., 2009; Tiecher et al., 2018), since this P fractions is associated to apatite, other moderately soluble Ca-P compounds or negatively charged oxide surfaces (Hedley et al., 1982).

It's known that weathered tropical Oxisols strongly sorb P onto Al- and Fe- oxides and hydroxides (Margenot et al., 2017; Kruse et al., 2015), what may explain the small value and unexpressive effect of fertilizer application on inorganic MLP concentration in the subsurface layer (20-40 cm) when compared to the soil surface (0-20 cm). Otherwise, the crescent rates of both initial and annual P fertilizer addition promoted an increase in $P_{O_{0.1Hid}}$ concentrations in subsurface (20-40 cm, Table 4), without any effect on surface layer (0-20 cm). Our results are in good agreement with Conte et al. (2003) who also observed a high concentration of organic MLP pool with P fertilizer application. Moreover, organic MLP pool can be a source (without or with small P fertilizer application) or drain (with high P fertilizer application) of P in soil solution, according to soil P dynamics and soil composition (Teles et al., 2017).

2.4.3. Non-labile P fractions ($P_{i0.5Hid}$, $P_{o0.5Hid}$ and $P_{Residual}$)

Non-labile P usually represents the largest pool found in tropical cultivated soils due to the long-term cumulative phosphate dosages applied. Both annual and initial rates of P

fertilizer have increased the inorganic NLP due the strong soil P-buffering capacity of our soil, more significantly decreasing available P contents over the time of cultivation under soluble P fertilizer addition (Table 4). Thus, that soils can fix P added from soluble phosphate source before it can be uptake by crops (Syers et al., 2008). This behavior is strongly related to amorphous/crystalline Fe ratio (Fe Oxalate/Fe DCB = 0.038) indicating that crystalline oxides are predominant (~31% of clay fraction) and confirmed by X-ray diffraction (Table 1 and Fig. 1), with higher P adsorption capacity (MPAC = 377 mg kg⁻¹) and consequently reduced LP and MLP pools. Also, small content of organic NLP fraction (Po_{0.5Hid}) found in our soil is explained by the stabilization of organic compounds by mineral weathering products such as Fe- and Al-oxides (Turner and Blackwell, 2013), which can be increased with the continuous P fertilizing in conventional tillage system, as observed in our results of surface layer (0-20 cm) under annual P rates (Table 4).

2.4.4. Crop productivity, agronomic efficiency and soil legacy P

Our Average yield of cotton and soybean increased proportionally with the increase in annual rate of P fertilizer, with the best yield under 97 kg P₂O₅ ha⁻¹. When excessively P supplied over the crop demand for optimal growth, excess grain P is unlikely to increase grain yield (Wang et al., 2017). On the other hand, the PUE decreased when increasing phosphate rates. High levels of initial available P (29 mg dm⁻³, Table 1) for Cerrado soils (Sousa and Lobato, 2004), which promoted a great yield in the control treatment and may explain our very small value of PUE, even under the smallest rate annually applied (PUE = 1.05 kg P kg⁻¹ under 30 kg ha⁻¹), it also support the small difference in P outputs between control and the highest P rate used (256 *versus* 284 kg P ha⁻¹).

Twelve years of annual phosphate fertilizer application contributed for a positive P balance, remaining in the soil -218, -74, 76, 235 and 387 kg P ha⁻¹ under 0, 30, 60, 90 and 120 kg P₂O₅ ha⁻¹ annually applied respectively. In fact, the higher adsorption capacity of Oxisols is able to maintain the majority of P from fertilizer in non-available fractions, but the positive balance in higher rates is more a consequence of the excessive P added via fertilizer. Similar studies using Hedley's fractionation reported that the legacy P has accumulated predominantly as MLP and NLP (Conte et al., 2003; Rodrigues et al., 2016; Teles et al., 2017). However, total legacy P in our soil profile (0-40 cm) was also increased in LP (10.1, 11.4 and 12.8%) and MLP (19, 18.6 and 20%) pools with annual phosphate rates higher than plant export (60, 90 and 120 kg P₂O₅ ha⁻¹ respectively). In addition, these changes in LP and

MLP over the time were more sensitive than NLP for a continuous annual P fertilization compared to the initial P fertilization. Thus, a great proportion of this legacy P may potentially be accessible for crops (Rowe et al., 2016).

2.5. CONCLUSIONS

Phosphate fertilizer applied initially (phosphating) or at annual base have affected the crop production and P efficiency, P dynamics and the amount of legacy P accumulated over a long-term evaluation. However, the annual P application had more effect than initial rates in both soil layers evaluated (0-20 and 20-40 cm). The rate of 97 P₂O₅ ha⁻¹ as annual base was the one responsible for optimum yield of both crops soybean and cotton.

Soil P pools suggested a positive effect of P application on labile P with addition of 60, 90 and 120 kg P₂O₅ ha⁻¹ annual rates without any effect of initial P application. Thus, annual application according to plant demand is more effective to improve soil legacy P availability and represent a profitable strategy for inputs of inorganic P fertilizer in tropical crop production systems.

REFERENCES

- Brookes, P.C., and D.S. Powlson. 1981. Preventing phosphorus losses during perchloric acid digestion of sodium bicarbonate soil extracts. *J. Sci. Food Agric.* 32:671–674.
- Bunemann, E.K., Heenan, D.P., Marschner, P., McNeill, A.M., 2006. Long-term effects of crop rotation, stubble management and tillage on soil phosphorus dynamics. *Aust. J. Soil Res.* 44, 611–618.
- Buresh, R. J., Smithson, P.C., Hellums, D.T., 1997. Building soil phosphorus capital in Africa. In: R. J. Buresh, P. A. Sanchez, F. Calhoun, editors, *Replenishing Soil Fertility in Africa*, SSSA Spec. Publ. 51. SSSA and ASA, Madison, WI. p. 111-149.
- Chien, S.H., Prochnow, L.I., Mikkelsen, R., 2010. Agronomic use of phosphate rock for direct application. *Better Crops*, 94, 21–23.
- Coelho, M.J.A., Aguiar, A.C.F., Sena, V.G.L., Moura, E.G., 2017. Utilization and fate of phosphorus of different sources applied to cohesive soil of Amazonian periphery. *Sci. Agric.* 74, 242-249.

- Condrón, L.M., Goh, K.M., Newman, R.H., 1985. Nature and distribution of soil phosphorus as revealed by a sequential extraction method followed by ^{31}P nuclear magnetic resonance analysis. *J. Soil Sci.* 36, 199-207.
- Condrón, L.M., Newman, S., 2011. Revisiting the fundamentals of phosphorus fractionation of sediments and soils. *J. Soils Sediments.* 11, 830-840.
- Conte, E., Anghinoni, I., Rheinheimer, D.D.S., 2003. Phosphorus accumulation fractions in a clayey oxisol in relation to phosphate doses under no-tillage. *Rev. Bras. Cienc. Solo* 27, 893–900.
- Cooper, J., Lombardi, R., Boardman, D., Carliell-Marquet, C., 2011. The future distribution and production of global phosphate rock reserves. *Resour. Conserv. Recycl.* 57, 78-86.
- Cross, A.F., Schlesinger, W.H. 1995. A literature review and evaluation of the Hedley fractionation: applications to the biogeochemical cycle of soil phosphorus in natural ecosystems. *Geoderma*, 64, 197–214.
- Darou, S.H., Ellis, B.G., Robertson, G.P., 2000. Phosphorus fractions and fate of phosphorus-33 in soils managed under plowing and no-tillage. *Soil Sci. Soc. Am. J.* 51, 1547-1553.
- Dick, W.A., Tabatabai, M.A., 1977. Determination of orthophosphate in aqueous solutions containing labile organic and inorganic phosphorus. *J. Environ. Qual.* 6, 82-85.
- Dorbas, C. 2009. Dry matter, nitrogen and phosphorus accumulation, partitioning and remobilization as effected by N and P fertilization and source-sink relations. *Eur. J. Agron.* 30, 129-139.
- EMBRAPA, 2013. Sistema brasileiro de classificação de solos, Terceira ed. Brasília, Distrito Federal.
- EMBRAPA, 1997. Manual de métodos de análise de solo, segunda ed. Rio de Janeiro, Rio de Janeiro.
- Fageria, N.K., 2001. Nutrient management for improving upland rice productivity and sustainability. *Commun. Soil Sci. Plant Anal.* 32, 2603–2629.
- Fink, J.R., Inda, A.V., Bavaresco, J., Barrón, V., Torrent, J., Bayer, C., 2016. Adsorption and desorption of phosphorus in subtropical soils as affected by management system and mineralogy. *Soil Tillage Res.* 155, 62–68.
- Fontes, M.P.F., Weed, S.B., 1991. Iron oxides in selected Brazilian Oxisols. I. Mineralogy. *Soil Sci. Soc. Am. J.* 55, 1143-1149.

- Gatiboni, L.C., Kaminski, J., Rheinheimer, D.S., Flores, J.P.C., 2007. Biodisponibilidade de formas de fósforo acumuladas em solo sob Sistema de plantio direto (Bioavailability of soil phosphorus forms in no-tillage system). *Rev. Bras. Ciênc. Solo (Braz. J. Soil Sci.)* 31, 691-699.
- Haridasan, M., 2008. Nutritional adaptations of native plants of the cerrado biome in acid soils. *Braz. J. Plant Physiol.* 20, 183-195.
- Hedley, M.J., Steward, W.B., Chuauhan, B.S., 1982. Changes in inorganic and organic soil phosphorus fraction induced by cultivation practices and laboratory incubation. *Soil Sci. Soc. Am. J.* 46, 970-976.
- Herrera, W.F.B., M. Rodrigues, A.P.B. Teles, G. Barth, and P.S. Pavinato. 2016. Crop yield and soil phosphorus lability under soluble and humic-complexed phosphate fertilizers. *Agron. J.* 108, 1692-1702.
- Jackson, M.L., 1979. *Soil Chemical Analysis Advanced Course*, second ed. Madison, Wisconsin.
- Kämpf, N., Scheinost, A.C., Schulze, D.G., 2000. Oxide minerals, in: Sumner, M. E. (Eds.), *Handbook of soil science*. University of Georgia, Georgia, pp. 125-168.
- Kruse, J., Abraham, M., Amelung, W., Baum, C., Bol, R., Kühn, O., Lewandowski, H., Niederberger, J., Oelmann, Y., Rüger, C., Santner, J., Siebers, M., Siebers, N., Spohn, M., Vestergren, J., Vogts, A., Leinweber, P., 2015. Innovative methods in soil phosphorus research: a review. *J. Plant Nutr. Soil Sci.* 178, 43–88.
- Linquist, B.A., Singleton, P.W., Cassaman, K.G., 1997. Inorganic and organic phosphorus dynamics during a build-up decline of available phosphorus in an ultisol. *Soil Sci.* 162, 254-264.
- Margenot, A.J., Singh, B.R., Rao, I.M., Sommer, R., 2017. Phosphorus fertilization and management in soils of Sub-Saharan Africa. In: Lal, R., Stewart, B.A. (Eds.), *Soil Phosphorus*. CRC Press, Boca Raton/Florida, pp. 151–208.
- McLaughlin, M.J., McBeath, T.M., Smernik, R., Stacey, S.P., Ajiboye, B., Guppy, C., 2011. The chemical nature of P accumulation in agricultural soils – implications for fertilizer management and design: an Australian perspective. *Plant Soil.* 349, 69-87.
- Mehra, O.P., Jackson, M.L., 1960. Iron oxide removal from soils and clays by a dithionite-citrate system buffered with sodium bicarbonate. *Clays Clay Miner.* 7, 317–327.
- Murphy, J., Riley, J.P., 1962. A modified single solution method for the determination of phosphate in natural waters. *Anal. Chim. Acta.* 27, 31 –36.

- Novais, R.F., Smyth, T.J., 1999. Fósforo em solo e planta em condições tropicais. Universidade Federal de Viçosa, Viçosa, Minas Gerais. Brazil.
- Pavinto, P.S., Merlin, A., Rosolem, C.A., 2009. Disponibilidade de cátions no solo alterada pelo sistema de manejo (Soil cation availability as affected by the tillage system). R. Bras. Ci. Solo. 33, 1031-1040.
- Raij, B., Quaggio, J.A., Silva, N.M., 2008. Extraction of phosphorus, potassium, calcium, and magnesium from soils by an ionexchange resin procedure. Commun. Soil Sci. Plant Anal. 17, 547-566.
- Raij, B.van, Diest, A.van, 1980. Phosphate supplying power of rock phosphates in an oxisol. Plant Soil. 55, 97-107.
- Rheinheimer, D.S., Anghinoni, I., Conte, E., Kaminski, J., Gatiboni, L.C., 2003. Dessorção de fósforo avaliada por extrações sucessivas em amostras de solo provenientes dos sistemas plantio direto e convencional. Ci. Rural. 33, 1053-1059.
- Rodrigues, M., Pavinato, P.S., Withers, P.J.A., Teles, A.P.B., Herrera W.F.B., 2016. Legacy phosphorus and no tillage agriculture in tropical oxisols of the Brazilian savanna. Sci. Total Environ. 542, 1050–1061.
- Rowe, H., Withers, P.J.A., Baas, P., Chan, N.I., Doody, D., Holiman, J., Jacobs, B., Li, H., MacDonald, G.K., McDowell, R., Sharpley, A.N., Shen, J., Taheri, W., Wallenstein, M., Weintraub, M.N., 2016. Integrating legacy soil phosphorus into sustainable nutrient management strategies for future food, bioenergy and water security. Nutr. Cycl. Agroecosyst. 104, 393-412.
- Sanchez, P.A., Cochrane, T.T., 1980. Soil constraints in relation to major farming systems in tropical America. In: priorities for Alleviating Soil-Related Constraints to Food Production in the Tropics, IRRI: Philippines. pp. 107-140.
- Sattari, S.Z., Bouwman, A.F., Giller, K.E., van Ittersum, M.K., 2012. Residual soil phosphorus as the missing piece in the global phosphorus crisis puzzle. Proc. Natl. Acad. Sci. 109, 6348–6353.
- Schaefer, C.E.G.R., Fabris, J.D., Ker, J.C., 2008. Minerals in the clay fraction of Brazilian latosols (Oxisols): a review. Clay Miner. 43, 137-154.
- Schwertmann, U., 1964. Differenzierung der eisenoxide des bodens durch extraction mit ammoniumoxalat-losung. Z. Pflanzenernahr. Bodenkd. 105, 194–202.
- Smyth, T.J., Sanchez, P.A., 1980. Effects of lime, silicate and phosphorus applications an Oxisol on phosphorus sorption and ion retention. Soil Sci. Soc. Am. J. 44, 500-505.

- Soil Survey Staff., 2014. Keys to Soil Taxonomy, twelfth ed. Washington, District of Columbia.
- Soltangheisi, A., Rodrigues, M., Coelho, M.J.A., Gasperini, M., Sartor, L.R., Pavinato, P.S., 2018. Changes in soil phosphorus lability promoted by phosphate sources and cover crops. *Soil tillage res.* 179, 20-28.
- Sousa, D.M.G., Lobato, L., 2004. Cerrado: correção do solo e adubação. 2ª ed. EMBRAPA Informação Tecnológica, Brasília-DF, Brasil. 416p.
- Stewart, W.M., Reiter, J.S., Kreig, D.R., 2005. Cotton response to multiple applications of phosphorus fertilizer. *Better crops.* 89, 18-20.
- Syers, J.K., Johnston, A.E., Curtin, D., 2008. Efficiency of soil and fertilizer phosphorus use: Reconciling changing concepts of soil phosphorus behavior with agronomic information. *FAO Fertilizer and Plant Nutrition Bulletin.* Food and Agriculture Organization of the United State Nations, pp. 18.
- Teles, A.P.B., Rodrigues, M., Bejarano Herrera, W.F., Soltangheisi, A., Sartor, L.R., Withers, P.J.A., Pavinato, P.S., 2017. Do cover crops change the lability of phosphorus in a clayey subtropical soil under different phosphate fertilizers?. *Soil Use Manage.* 33, 34-44.
- Tiecher, T., Gomes, M.V., Ambrosiani, V.G., Amorim, M.B., 2018. Assessing linkage between soil phosphorus forms in contrasting tillage systems by path analysis. *Soil tillage res.* 175, 276-280.
- Tiessen, H., Moir, J.O., 1993. Characterization of available P by sequential extraction, in: Carter, M.R., (Ed.), *Soil sampling and methods of analysis.* Canadian Soc. of Soil Sci. Lewis Publ., Boca Raton, Florida. pp. 75–86.
- Turner, B.L., Blackwell, M.S.A., 2013. Isolating the influence of pH on the amounts and forms of soil organic phosphorus. *Eur. J. Soil Sci.* 64, 249–259.
- USEPA - United States Environmental Protection Agency, 1971. *Methods of chemical analysis for water and wastes.* Environmental Protection Agency, Cincinnati, Ohio.
- Vance, C.P., Unde-Stone, C., Allan, D.L. 2003. Phosphorus acquisition and use: critical adaptation by plants for securing a nonrenewable resource. *New Phytol.* 157, 423-447.
- Yost, R.S., Kamprath, E.J., Lobato, E., Naderman, G.C., 1979. Phosphorus response of corn on an Oxisol as influenced by rates and placement. *Soil Sci. Soc. Am. J.* 43, 338-343.
- Younge, O.R., Plucknet, D. L., 1966. Quenching the high phosphorus fixation on Hawaiian Latosols. *Soil Sci. Soc. Am. Proc.* 30, 653-655.

3. LONG-TERM FERTILIZER PLACEMENT AFFECTS SOIL PHOSPHORUS FRACTIONS AND LEGACY IN A CORN-SOYBEAN ROTATION IN NORTH-CENTRAL KANSAS, USA

ABSTRACT

Phosphorus (P) fertilizer placement can affect plant P uptake during the growing season. However, the long-term interaction of placement and plant root P uptake and cycling can also affect soil P pools. The objective of this study was to evaluate the influence of long-term P fertilizer placement on soil P pools (labile, moderately labile, and non-labile) and legacy soil P accumulation under a long-term corn-soybean rotation. A field study was conducted for 10 years from 2006 to 2015. Three treatments were evaluated, including a control with no P fertilizer application and two fertilizer treatments (39 kg P ha^{-1}): (1) surface broadcast and (2) deep band at approximately 15 cm depth. All treatments received strip-tillage. After 10 years, soil samples were collected from the row and between the row at two sampling depths (0-7.5 and 7.5-15 cm) and soil P pools via sequential P fractionation were measured. Significant changes in soil P pools for treatments compared to initial soil and control were observed due to the long-term effect of P fertilizer placement. Broadcast P fertilizer placement increased the amount of labile P and moderately labile P in the topsoil (0-7.5 cm) and had a greater level of P fertilizer use efficiency compared to deep-band placement. With reduced tillage, organic carbon amount increased in the soil surface (0-7.5 cm) that may have affected differences between P placements and P dynamics in the soil. Placing P fertilizer exclusively using one strategy over many years can promote depletion of P pools from some soil layers and row locations.

Keywords: Phosphorus placement; Phosphorus pools; Strip-till tillage; Legacy phosphorus

3.1. INTRODUCTION

Phosphorus (P) is considered the second macronutrient that most often limits agricultural production, next to nitrogen (N). The demand of P fertilizer to obtain optimal crop yields in order to produce enough food for the global population will continue to increase (Chowdhury et al., 2017), however, the amount of P, with the quantity of phosphate rock remaining being uncertain, is practically finite (Reijnders, 2014). In many soils in the world, P added as fertilizer becomes quickly fixed by sorption to mineral phases and converted to compounds of more limited bioavailability (Stutter et al., 2015). Required in relatively large quantities, P fertilizer is often applied beyond the total demand of plants. Thus, excess applied P can be considered a waste of resources and can increase potential environmental problems (Luo et al., 2017), such as eutrophication of surface waters from the P transported by runoff and eroded particulates (Withers et al., 2014; Cassidy et al., 2017). Efficient P management in crop production is mandatory to minimize depletion of phosphate rock reserves, environmental issues, and production costs. A good understanding of the pools

and processes of P dynamics in the soil can form a solid basis for efficient P management and reduce the dependence on excessive rates of fertilizer (Bünemann, 2015).

In the soil, P exists in inorganic (Pi) as well as organic (Po) forms of comparable solubility (Weihrauch and Opp, 2018), but differ in their amounts and proportions (Song et al., 2017), and shows a strong interaction and sorption reactions with the soil components. Plants take up only 10-20% of the P applied as fertilizer during the year of application (Vu et al., 2008).

Thus, P added in soils as fertilizers in predominately soluble Pi forms can be found adsorbed to a high-energy with Al- and Fe- oxides and hydroxides on the surface of 1:1 clay minerals through ligand exchange and precipitated as Fe and Al phosphate in acidic soils, such as Tropical soils, and reactions with Ca to form less soluble or insoluble compounds in alkaline soils, such as Temperate soils, or be physicochemically stabilized into soil organic matter (SOM) complexes (Shen et al, 2011; Eriksson et al., 2015).

Due to these reactions, P often accumulates in soils following long-term continuous annual application of P fertilizers (Vu et al., 2007), resulting in a buildup of the residual P pool (Pi and Po) in soils (Condrón et al., 2013). Residual P, also called legacy P, is defined as accumulated P in soils, which can be calculated as the difference between inputs and outputs of P over time (Havens and James, 2005; Sattari et al., 2012; Rowe et al., 2016).

Previous studies in temperate regions (Sattari et al., 2012; Rowe et al., 2016) and some specific ally from North America (Kamprath, 1999; Dodd and Mallarino, 2005) suggest that legacy soil P remains accessible to plants and helps maintains productivity in agroecosystems, enabling reductions in P inputs and P transfer from land to water (Condrón et al., 2013). Fertilizer P placement can affect crop P utilization, change soil P dynamics, and the legacy-P concentration at various soil depths and soil-plant interactions. Broadcast application is the simplest and most popular method of fertilizer P. Broadcast fertilization can result in uniform fertilizer distribution with high-speed operations, potentially reducing working time and labor (Hansel et al. 2017a). Increased contact of fertilizer with soil in broadcast-P application over large soil areas can increase the potential for P retention and fixation by some soils. However, the use of deep-band fertilizer placement may minimize potential P fixation in higher P-fixing soils due to the smaller volume of soil in contact with the fertilizer (Anghinoni and Barber, 1980). Previous research extensively evaluated the differences between broadcast and deep-band P fertilizer placement on short-term crop response under different soils, tillage, and cropping systems (Sleight, et al., 1984; Khatiwada et al., 2012; Mallarino et al., 2012; Adeo et al., 2016; Hansel et al., 2017ab). However, few studies have evaluated the long-term effect of

different P fertilizer placement strategies and particularly the preferential forms of P retained in the soil pool and their contribution to increasing P use efficiency by crops over time.

Hedley's P fractionation is the most common approach to identify the redistribution of applied P in the soil through sequential extraction with a combination of alkali and acid reagents to identify different forms of soil P (Hedley et al. 1982). Furthermore, the method has been used extensively to investigate the distribution of labile and non-labile forms of P_i and P_o among phosphate and minerals and organic soil components (Condrón et al., 1985; Condrón and Newman, 2011). The hypothesis of this study was that in long-term P fertilization in reduced tillage the broadcast placement can be as effective as deep band placement in keeping available P levels in the soil. Thus, this study aimed to evaluate the effects of long-term P fertilizer placement (broadcast versus deep band) in a corn-soybean crop rotation on changes in soil P pools (labile, moderately labile and non-labile) and legacy soil P accumulation in a Mollisol in north-central in Kansas.

3.2. MATERIALS AND METHODS

3.2.1. Site description and soil characterization

A field study was conducted for 10 consecutive years (2006-2015) on a Crete silt-loam soil (fine, smectitic, mesic Pachic Udertic Argiustolls) (Soil Survey Staff, 2014) in Scandia, Kansas, USA (39°46'23" N; 97°47'19" W). The regional climate is classified as a Humid Continental (warm summer) climate, with mean annual precipitation of 760 mm and mean annual temperature of 12 °C. The site had a history of no-tillage production practices for approximately five years before the implementation of this study and received supplemental irrigation. A corn-soybean rotation was established, with a total of five years of corn and five years of soybean during the 10 years.

Initial Initial soil samples were collected in April 2006 before initiating the study by collecting a representative sample from the 0-7.5 and 7.5-15 cm layers for the characterization of general soil properties of the experimental area (Table 1). Soil samples were air-dried and passed through a 2-mm sieve prior to chemical analyses. Soil pH was determined in a 1:1 (m/v) soil:water ratio and soil exchangeable acidity (H+Al) were determinate according to the methodologies proposed by Watson and Brown (1998). Soil test P (STP) was determined colorimetrically using the Mehlich-3 extraction (Frank et al., 1998). Total organic carbon

(TOC) and total organic N (TON) were determined by dry combustion (LECO Corporation, 2005). Exchangeable potassium (K), calcium (Ca), magnesium (Mg), and sodium (Na) were determined by inductively coupled plasma optical emission spectrometry (ICP-OES) after extraction with 1 M ammonium acetate (Warncke and Brown, 1998). Cation exchange capacity (CEC) was determined by summing the exchangeable cations (in $\text{cmol}_c \text{ kg}^{-1}$). Extractable iron (Fe), zinc (Zn), copper (Cu), and manganese (Mn) were determined using diethylenetriaminepentaacetic acid (DTPA) extraction technique described by Whitney (1998) and analyzed with by ICP-OES.

Particle-size distribution was determined using a modification of the pipette method of Kilmer and Alexander (1949), and the 3A1 method from the Soil Survey Laboratory Methods Manual (Soil Survey Laboratory Staff, 2004). To evaluate maximum P adsorption capacity (MPAC), 2.5g subsamples of soil were mixed with 11 rates of added P (0, 4, 8, 12, 20, 28, 36, 44, 56, 68, and 80 mg L^{-1}) as potassium dihydrogen phosphate (KH_2PO_4 (p.a.)), in a 25 mL equilibrium solution of calcium chloride (0.01 M $\text{CaCl}_2 \cdot 2\text{H}_2\text{O}$). Suspensions were shaken for 24 h at 147 g and centrifuged for 15 min at 3278 g, the P concentration was measured in the supernatant by the blue-molybdate colorimetric method (Murphy and Riley, 1962). The MPAC was obtained after fitting the Langmuir equation, calculated as: $P_{\text{so}} = [k \times (P_{\text{max}} - P_{\text{ce}}) / (1 + K \times P_{\text{ce}})]$, where P_{so} is the amount of P sorbed in each soil sample, k is a constant related to the sorption energy, P_{max} is the maximum P sorption capacity, and P_{ce} is the equilibrium P concentration in solution. The estimated result of MPAC was 288 mg P kg^{-1} of soil (Table 1).

Table 1. Initial soil chemical and textural parameters for a Pachic Udertic Argiustolls from the experimental site at Scandia, Kansas, United States.

Depth	pH	TON†	TOC‡	P§	K	Ca	Mg	Na	H+Al¶	CEC#
	H ₂ O	--- g kg ⁻¹ ---		----- mg kg ⁻¹ -----			cmol _c kg ⁻¹			
0 - 7.5	6.5	1.8	20	21	586	2159	371	31	2.5	17
7.5 - 15	6.5	1.6	14	12	452	2443	426	45	2.5	21
	Fe	Zn	Cu	Mn	MPAC††			Clay	Silt	Sand
	----- mg kg ⁻¹ -----					----- g kg ⁻¹ -----				
0 - 7.5	32	1.7	1.2	88	288			210	590	200
7.5 - 15	45	1.0	1.4	77				290	550	160

† TON, total organic nitrogen

‡ TOC, total organic carbon

§ Mehlich III-extractable phosphorus

¶ H+Al, exchangeable acidity

CEC, cation exchange capacity

†† MPAC, maximum phosphorus adsorption capacity

3.2.2. Experimental set-up and management

A detailed description of the experimental design and crop management was provided in Preston et al. (2018). Briefly, the experiment was set up as a randomized complete block design with four replications. For this particular study, three treatments were evaluated, including a control with no P fertilizer application during the 10 years of the study, and two fertilizer treatments (39 kg P ha⁻¹) applied with two placements: (1) surface broadcast and (2) deep band at approximately 15 cm depth applied with the strip-tillage operation. The P fertilizer treatments were applied before corn only. A strip-tillage operation was performed before planting corn and soybean was planted into the corn residue with no prior tillage. Strip-tillage was used for all plots including the control with no P fertilizer application. Deep-band P fertilizer application was completed with the strip-tillage operation applied in a concentrated zone spaced at 76 cm and made in the same row location in all 10 years. Corn and soybean were planted in the center of the strip in the same row each year. The plot size was 16 m long by 3 m wide with four rows per plot at 76 cm row spacing.

Phosphorus fertilizer source for the broadcast treatment was triple superphosphate (0-20-0) applied by hand to the soil surface before corn planting. The P fertilizer source for deep

banding was ammonium polyphosphate (10-15-0). Nitrogen fertilizer was applied to all plots before corn using anhydrous ammonia at 224 kg N ha⁻¹, such that the N applied with the P fertilizer treatments were balanced to provide the same amount of N for all plots. Corn and soybean were harvested using a plot combine and grain samples were collected and analyzed for total P content each year. Yield and grain P content were used to estimate total P removal during the 10-year period.

3.2.3. Soil sampling, analysis, and phosphorus fractionation

After the last soybean harvest in 2015 following 10 years of corn-soybean rotation, a composite soil sample of eight to 10 soil cores was collected from the 0-7.5 and 7.5-15 cm depths from each plot. Samples were collected at two positions: in row (IR) and between rows (BR) at 38 cm from the rows. Samples were collected from the central two rows in each plot and randomly distributed along the length of the plot. Soil samples were air-dried and passed through a 2-mm sieve prior to chemical analyses. Chemical analysis included measurement of pH, TON, TOC and the sequential P fractionation. Soil pH was determined in a 1:1 (m/v) soil:water ratio (Watson and Brown, 1998). TON and TOC were determined by dry combustion (LECO Corporation, 2005).

Sequential P fractionation was completed following the methodology proposed by Hedley et al. (1982) with modifications by Condon et al. (1985). The technique uses sequential chemical extractants on the same sample to progressively remove the most available to the most stable inorganic (Pi) and organic (Po) P pools. At each step, 10 mL of extractant were added to 0.5 g soil in 15 mL centrifuge tubes [1:20 (m/v) soil:solution ratio] and the tubes were shaken horizontally in a reciprocal shaker at 60 revolutions per minute (rpm) for 16 h at 25°C. Soil samples were extracted sequentially using: (i) anion exchange resin (AER) for inorganic P (P_{AER}) readily diffusing into solution using a resin membrane (2.0 cm² in area); (ii) 0.5 M NaHCO₃ to extract inorganic P (P_{Bic}) weakly adsorbed on the surface of crystalline compounds and organic P (P_{OBic}) compounds with low recalcitrance like ribonucleic acid and glycerophosphate (Tiessen and Moir, 1993); (iii) 0.1 M NaOH to extract inorganic P (P_{i0.1Hid}) strongly adsorbed onto Fe and Al and clay minerals (Hedley et al., 1982) and organic P (P_{o0.1Hid}) mainly associated with fulvic and humic acids adsorbed onto mineral and SOM surfaces (Linquist et al., 1997); and (iv) 1 M HCl to extract inorganic P (P_{HCl}) associated with apatite, other sparingly-soluble Ca-P compounds or negatively charged oxide surfaces (Gatiboni et al., 2007); (v) 0.5 M NaOH to extract inorganic P (P_{i0.5Hid}) associated

with Fe and Al, clay minerals, and organic P ($P_{O_{0.5Hid}}$) associated with fulvic and humic acids inside aggregates (Condon et al., 1985). After each extraction, the soil suspensions were centrifuged at 3278 g for 30 min to collect clear supernatants. After centrifugation and removal of the supernatant at each step of the fractionation, the soil was washed with 10 mL of 0.5 M NaCl solution to prevent the residual effect of the previous solution (pH change). After drying the remaining soil at 40°C for 3 days, the residual P ($P_{Residual}$) was determined by $H_2SO_4 + H_2O_2 + MgCl_2$ digestion (Brookes and Powlson, 1981). This fraction consists of occluded P forms (Cross and Schelinger, 1995).

The alkaline extracts were added to a 1:1 sulfuric acid solution and 7% ammonium persulfate in an autoclave at 121 °C and 103 kPa for 2 h to determine the Pt ($P_i + P_o$) (USEPA, 1971). The amount of P_i in the alkaline extracts (P_{iBic} , $P_{i0.1Hid}$ and $P_{i0.5Hid}$) was determined using the colorimetric method of Dick and Tabatabai (1977). Inorganic P in each of the acid extracts (P_{AER} , P_{HCl} , $P_{Residual}$ and digestion of P_{tBic} , $P_{t0.1Hid}$ and $P_{t0.5Hid}$) were determined following the colorimetric method of Murphy and Riley (1962). Organic P in each alkaline extract was determined by the difference between the total P (Pt) and P_i (P_{oBic} , $P_{o0.1Hid}$ and $P_{o0.5Hid}$).

The pools of P were quantified based on the lability predicted by the extractors (Cross and Schlesinger, 1995). Labile P (LP) consisted of the P_{AER} plus the P_{Bic} (P_i and P_o). The moderately labile P (MLP) consisted of the $P_{0.1Hid}$ (P_i and P_o) plus the P_{HCl} . The non-labile P (NLP) consisted of the $P_{0.5Hid}$ (P_i and P_o) plus the $P_{Residual}$.

3.2.4. Phosphorus efficiencies and soil legacy P

The difference between the total P inputs from the fertilizers and outputs from crop removal was used to estimate the P balance. Phosphorus agronomic efficiency (PAE) and P recovery efficiency (PRE) were estimated according to Fageria (2001) by relating the cumulative P uptake and grain yield, respectively, and the total applied P fertilizer as follows: $PAE = (Y_f - Y_o) / Q_f$, where PAE is the P agronomic efficiency ($kg \text{ grain } kg^{-1}$ of P applied), Y_f is the cumulative yield with fertilized P rate ($kg \text{ grain } ha^{-1}$), Y_o is the cumulative yield for the unfertilized control treatment and Q_f is the P fertilizer application rate ($kg \text{ P } ha^{-1}$). For PRE, grain yield (Y_f and Y_o) were replaced by P uptake ($kg \text{ P } kg^{-1}$ of P applied).

Soil legacy P was estimated by summing the amount of total labile P, moderately labile P, and non-labile P ($mg \text{ P } kg^{-1}$) of each treatment (control, broadcast, deep band) and

subtracting the amount of total labile P, moderately labile P and non-labile P (mg P kg^{-1}) from the soil at the beginning of the study.

3.2.5. Statistical analysis

All statistical analyses were completed in SAS Studio (version 9.3; SAS, Institute, Inc, Cary, NC). The GLIMMIX procedure was used for analysis of variance (ANOVA). Phosphorus fertilizer placement and soil sampling position were considered as fixed factors in the model and blocks were considered as random factors (Littell et al., 2006). Corrected-denominator degrees of freedom were obtained using the Kenward-Roger adjustment. Mean comparisons were conducted using the LSMEANS and SLICE option in PROC GLIMMIX. Statistical differences were established at the 0.05 probability level.

3.3. RESULTS

3.3.1. Soil Chemical properties

The The main P fertilization treatments imposed for 10 years did not affect soil TON and pH; however, these parameters were differed significantly by depth, with the greatest values in the 0-7.5 cm layer (Table 2 and Fig. 1A and 1B). TOC differed between sampling position by depth, where values were statistically greater between-row compared to the in-row position for the 0-7.5 cm depth (Table 2 and Fig. 1C). These results suggest that changes in soil chemical properties were likely generated by the combination of cropping system management and fertilizer placement.

Table 2. Statistical significance of P fertilizer placement treatment, soil sampling position and depth for soil pH, total organic nitrogen, and total organic carbon. Soil samples collected at the 0-7.5 and 7.5-15 cm depths, two positions (between-row and in-row), for three P fertilizer treatments (deep-band, broadcast, and control). Samples collected after ten years of corn and soybean rotation.

Fixed effect	Soil chemical properties		
	pH	Total organic nitrogen (TON)	Total organic carbon (TOC)
	----- p > F -----		
	-		
Treatment (T)	0.799	0.090	0.656
Position (P)	0.114	0.427	<0.001
Depth (D)	<0.001	<0.001	<0.001
T × P	0.883	0.530	0.541
T × D	0.409	0.530	0.163
P × D	0.908	1.000	0.001
T × P × D	0.519	0.160	0.169

Significant different detected at $p < 0.05$.

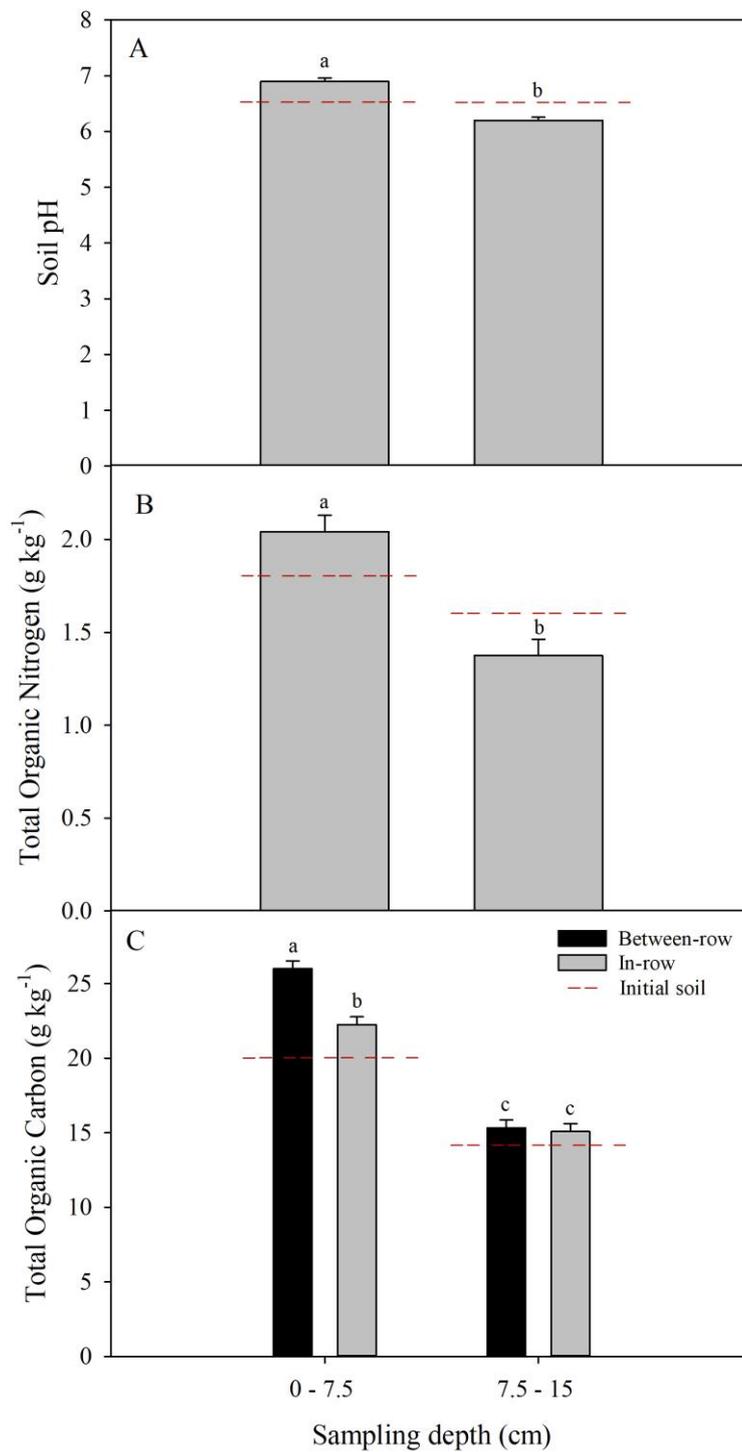


Figure 1. Soil pH (A), total organic nitrogen - TON (B) and total organic carbon - TOC (C) at two sampling depths under two sampling positions (between-row and in-row) after ten years of a corn-soybean rotation and P fertilizer treatment application. Error bars indicate the standard error of the mean. Significance of the main effects are presented in table 2

3.3.2. Labile P pool ($LP = P_{AER}$, P_{iBic} and P_{oBic})

In After 10 years, the labile P pool (i.e., P_{OLP} , P_{iLP} and P_{tLP}) differed significantly among treatment - sampling position - depth combinations (Table 3). The labile pool represented approximately 8% of total P (P_{Total}), with a decrease in average concentration in the following order: P_{iBic} (3.5%) > P_{AER} (2.5%) > P_{oBic} (2%) (Table 4). The continuous P fertilizer placement affected inorganic P (P_{iLP}) by increasing cycling more than the organic P (P_{OLP}) fraction and enhancing the proportion of total labile P (P_{tLP}) (Fig. 2). In the 0-7.5 cm depth, the greatest proportions of P_i and P_{tLP} were observed for the broadcast treatment for both sampling positions (between-row and in-row) compared to the deep-band and control treatments (Fig. 2B and 2C). However, in the 7.5-15 cm depth the largest P_{iLP} and P_{tLP} concentrations were observed for the deep-band treatment compared with the broadcast and control treatments at the in-row sampling position. At the between-row sampling position, P_{tLP} concentrations did not differ among the broadcast, deep-band, and control treatments (Fig. 2C).

The greatest concentrations of organic P (P_{OLP}) occurred in the 7.5-15 cm depth for the control and broadcast treatments at the in-row position and for the deep-band treatment at the between-row position. However, the smallest concentrations occurred for the deep-band treatment at the in-row position in the 7.5-15 cm depth and for the broadcast treatment at the between-row position in the 0-7.5 cm depth (Fig. 2A).

The proportion of the inorganic P pools ($P_{AER} + P_{iBic}$) for the broadcast treatment in the 0-7.5 cm depth at the between-row and in-row positions represented approximately 92 and 87% of P_{tLP} , respectively (Table 4). Conversely, the organic P fraction (P_{OLP}) for the broadcast represented 8 and 13% of the P_{tLP} for the between-row and in-row positions, respectively. Moreover, in the 7.5-15 cm depth, the proportion of the P_{OLP} for the control and broadcast treatments at the in-row position and the deep-band treatment at the between-row position represented approximately 53, 33 and 44% of the P_{tLP} , respectively (Table 4). In addition, the P_{iLP} ($P_{AER} + P_{iBic}$) for the broadcast treatments in the 7.5-15 cm depth represented 67% and 56% of the P_{tLP} for in-row and between-row positions, respectively. For the deep-band treatment at the in-row position, P_{OLP} (P_{oBic}) represented 10% and P_{iLP} represented 90% of the P_{tLP} . Results from this study showed that treatments with the largest amount of P_i showed the smallest amount of P_o , indicating the reverse dynamics for these two pools for each sampling position and depth (Fig. 2A and 2B).

Table 3. Statistical significance of P fertilizer placement, soil sampling position and depth on P lability fractions. Soil samples collected at two sampling depths (0-7.5 and 7.5-15 cm) and two positions (between-row and in-row) for three P fertilizer treatments (deep-band, broadcast, and control). Samples collected after ten years of corn and soybean rotation.

Fixed effect	Labile P (LP)			Moderately labile P (MLP)			Non-labile P (NLP)		
	Inorganic (Pi)	Organic (Po)	Total (Pt)	Inorganic (Pi)	Organic (Po)	Total (Pt)	Inorganic (Pi)	Organic (Po)	Total (Pt)
	----- p > F -----								
Treatment (T)	<0.001	0.005	<0.001	<0.001	0.045	0.009	0.004	0.844	0.004
Position (P)	<0.001	0.164	<0.001	0.011	0.350	0.900	0.107	0.701	0.227
Depth (D)	<0.001	<0.001	<0.001	<0.001	0.111	0.002	0.166	0.697	0.123
T × P	<0.001	0.001	<0.001	0.002	0.786	0.403	0.119	0.837	0.325
T × D	<0.001	0.005	<0.001	0.003	0.030	0.055	0.307	0.102	0.039
P × D	<0.001	0.032	0.001	0.831	0.335	0.385	0.563	0.013	0.019
T × P × D	<0.001	<0.001	0.003	0.274	0.851	0.621	0.674	0.488	0.897

Significant different detected at $p < 0.05$.

Table 4. Phosphorus content in inorganic (Pi) and organic (Po) fractions at two soil sampling depths (0-7.5 and 7.5-15 cm) and two sampling positions (between-row and in-row) for three P fertilizer treatments (deep-band, broadcast, and control). Samples collected after ten years of corn and soybean rotation.

Position	Treatment	Inorganic P fractions					Organic P fractions				P _{Total} †
		P _{AER}	P _{iBic}	P _{i0.1Hid}	P _{HCl}	P _{i0.5Hid}	P _{Residual}	P _{OBic}	P _{O0.1Hid}	P _{O0.5Hid}	
----- mg P kg ⁻¹ -----											
<u>0 - 7.5 cm</u>											
Between-row	Control	9	14	27	106	77	150	11	63	80	528
	Deep band	17	16	27	102	96	154	7	55	88	563
	Broadcast	33	32	33	123	89	171	6	63	82	633
	Mean	20	21	48	110	87	158	8	60	83	575
In-row	Control	9	19	22	109	93	146	10	64	60	527
	Deep band	20	23	28	101	82	161	10	60	78	558
	Broadcast	26	35	33	116	91	162	9	65	80	610
	Mean	18	26	27	109	89	156	10	63	72	565
<u>7.5 - 15 cm</u>											
Between-row	Control	4	15	26	86	82	147	12	78	69	530
	Deep band	6	13	28	77	97	152	15	46	75	510
	Broadcast	6	14	34	90	91	162	13	58	69	539
	Mean	6	14	29	84	90	154	13	61	71	526
In-row	Control	5	10	26	78	86	167	17	90	89	553
	Deep band	15	39	37	79	90	183	6	61	79	596
	Broadcast	17	15	30	83	84	177	16	70	73	560
	Mean	13	21	31	80	87	176	13	73	80	570
----- p > F, for fixed effects -----											
Treatment (T)		<0.001	<0.001	<0.001	<0.001	0.167	0.023	0.005	0.045	0.844	0.002
Position (P)		0.000	<0.001	0.412	0.010	0.776	0.062	0.164	0.350	0.701	0.102
Depth (D)		<0.001	<0.001	0.088	<0.001	0.972	0.139	<0.001	0.111	0.697	0.020
T × P		0.000	<0.001	0.017	0.026	0.011	0.253	0.001	0.786	0.837	0.140
T × D		<0.001	<0.001	0.092	0.013	0.548	0.394	0.005	0.030	0.102	0.001
P × D		<0.001	0.031	0.229	0.564	0.047	0.064	0.032	0.335	0.013	0.018
T × P × D		<0.001	<0.001	0.153	0.116	0.707	0.913	<0.001	0.851	0.488	0.586

† Sum of all inorganic and organic P fractions

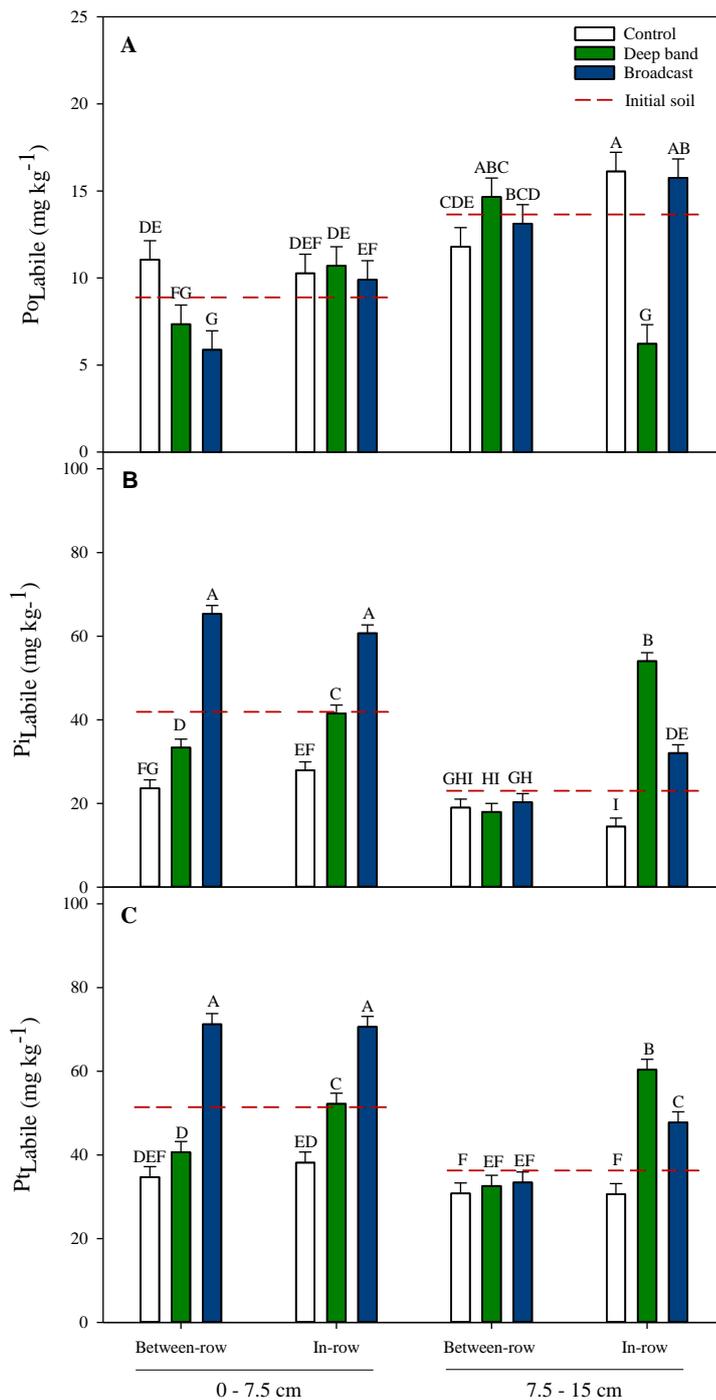


Figure 2. Labile P (LP) pool: organic - Po (A), inorganic - Pi (B) and total - Pt (C) for two soil sampling depths and two sampling positions (between-row and in-row) as affected by P fertilizer treatments (deep-band, broadcast, and control) after ten years of a corn-soybean rotation. Error bars indicate the standard error of the mean and mean values followed by the same letter are not statistically different ($p > 0.05$).

3.3.3. Moderately labile P pool (MPL = $P_{i0.1Hid}$, $P_{o0.1Hid}$ and P_{HCl})

The average concentration of the moderately labile P pool was 34% of the P_{Total} and the relative size of the MLP increased in the following order: $P_{i0.1Hid}$ (5.5%) < $P_{o0.1Hid}$ (11.5%) < P_{HCl} (17%) (Table 4). This is likely due to the predominant concentration of Ca-precipitated P (P_{HCl}) under Pi and Po compared to the Fe/Al oxides and hydroxides.

The moderately labile organic P (P_{OMLP}) differed among treatment – depth combinations (Table 3). The greatest P_{OMLP} concentration occurred in the control treatment in the 7.5-15 cm depth (Fig. 3A). For all other treatments and sampling depths the P_{OMLP} concentrations were similar and smaller than the control at the 7.5-15 cm depth (Fig. 3A). In contrast, a greater proportion of concentration of the moderately labile inorganic P (P_{iMLP}) occurred in the 0-7.5 cm depth and differed among treatment - position and treatment – depth combinations (Table 3). For both interactions, the greatest P_{iMLP} was observed in the broadcast treatment in the 0-7.5 cm depth and in the between-row position (Fig. 3B and 3C).

The total moderately labile P (P_{tMLP}) was influenced by the Po and Pi pools in the control and broadcast treatments and differed among treatments – depths combinations (Table 3). Consequently, the greater P_{tMLP} occurred in the 0-7.5 cm depth and the broadcast treatment was significantly greater than the deep-band treatment (Fig. 3D and 3E).

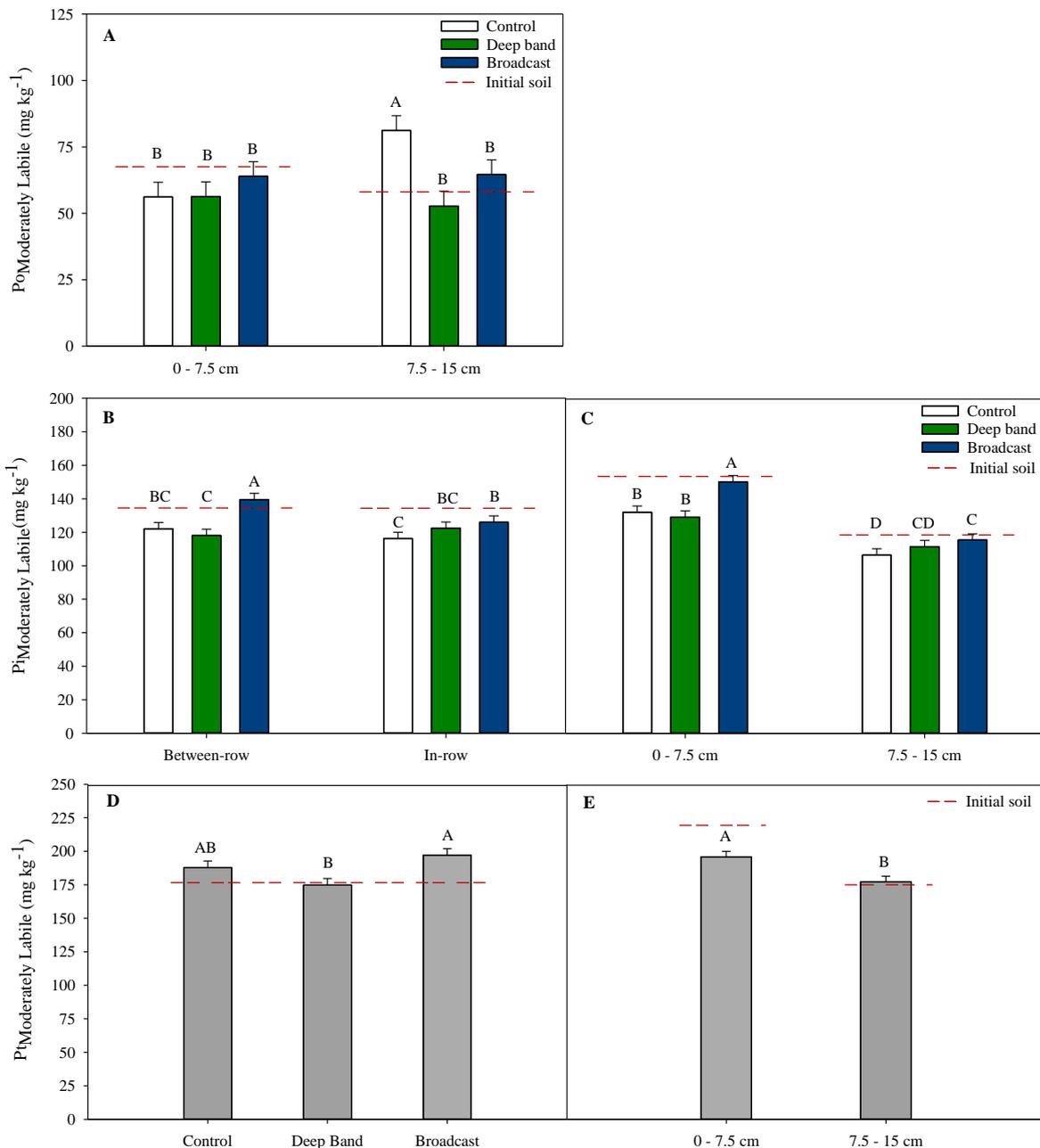


Figure 3. Moderately labile P (MLP) pool: organic - Po (A), inorganic - Pi (B and C) and total - Pt (D and E) for two soil sampling depths and two sampling positions (between-row and in-row) as affected by P fertilizer treatments (deep-band, broadcast, and control) after ten years of a corn-soybean rotation. Error bars indicate the standard error of the mean and mean values followed by the same letter are not statistically different ($p > 0.05$).

3.3.4. Non-labile P pool (NLP = $P_{i0.5Hid}$, $P_{o0.5Hid}$, and $P_{Residual}$)

The non-labile P pools represented the greatest proportion (58%) of P_{Total} in the soil, with overall average concentrations increasing in the following order: $P_{o0.5Hid}$ (13.5%) < $P_{i0.5Hid}$ (15.8%) < $P_{Residual}$ (28.7%) (Table 4). The non-labile organic P (P_{ONLP}) differed among position – depth combinations (Table 3). In the 0-7.5 cm depth the greatest P_{ONLP} concentration occurred at the between-row position, but did differ between positions in the 7.5-15 cm depth (Fig. 4A). Both P application methods, broadcast and deep-band, increased the non-labile inorganic P (P_{INLP}) compared to the control (Table 3 and Fig. 4B). However, a greater proportion of the P_{INLP} occurred in the $P_{Residual}$ fractions and did not differ among treatments, positions, or sampling depths after 10 years of cropping (Table 3). Thus, the amount of total non-labile P (P_{tNLP}) differed among treatment - depth and position – depth combinations (Table 3) but was primarily influenced by the proportions of P in the $P_{o0.5Hid}$ and $P_{i0.5Hid}$ fractions (Table 4). The control treatment in the 0-7.5 cm depth had a lower concentration compared to the other treatment-sampling depth combinations (Fig. 4C). Furthermore, in the 7.5-15 cm depth, the largest proportion of P_{tNLP} occurred at the in-row position (Fig. 4D).

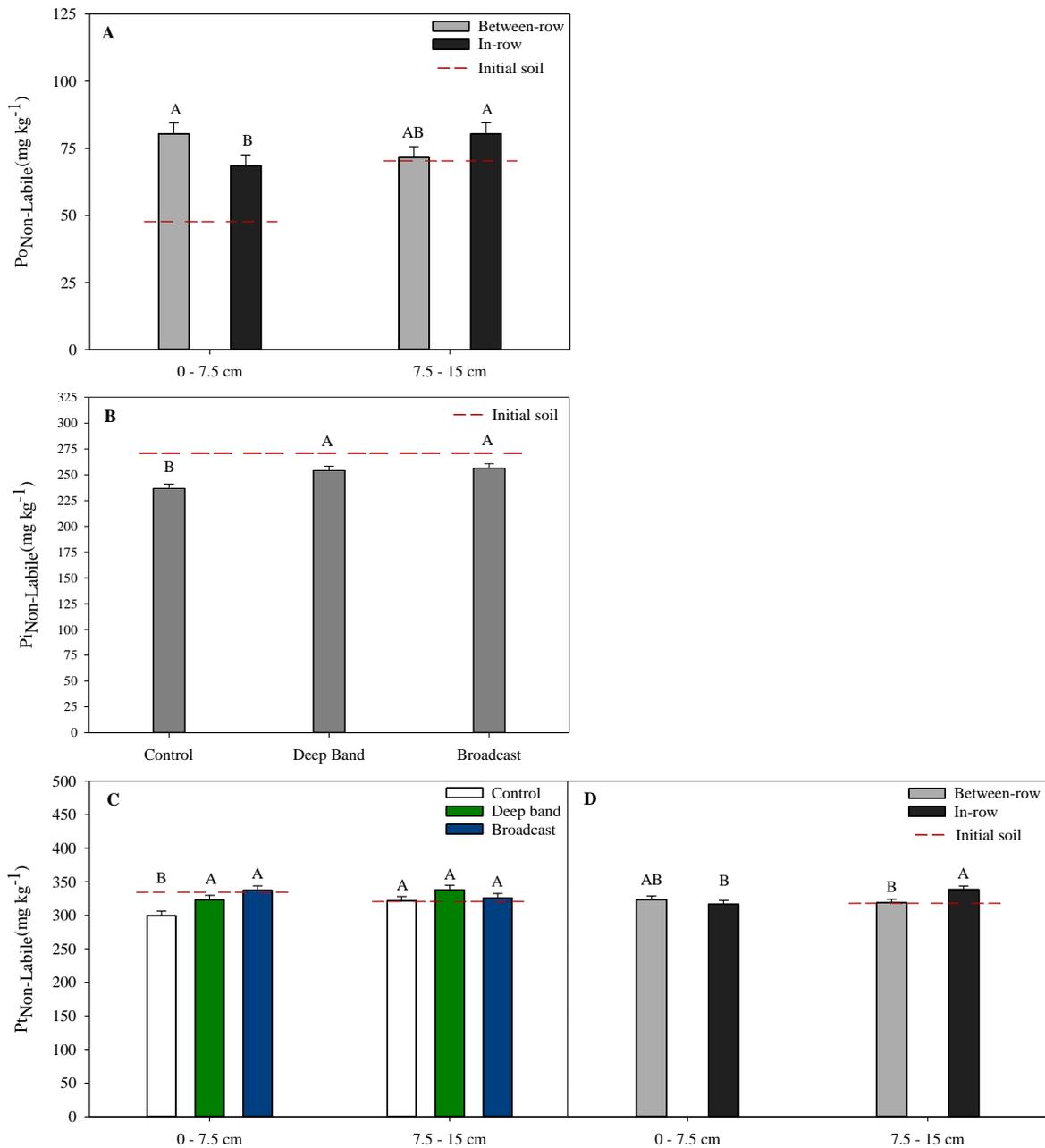


Figure 4. Non-labile P (NLP) pool: organic - Po (A), inorganic - Pi (B) and total - Pt (C and D) for two soil sampling depths and two positions (between-row and in-row) as affected by P fertilizer treatments (deep-band, broadcast, and control) after ten years of a corn-soybean rotation. Error bars indicate the standard error of the mean and mean values followed by the same letter are not statistically different ($p > 0.05$).

3.3.5. Legacy soil P

Soil P placement affected the amount and fractional distribution of P after 10 years. Figure 5 shows the overall relationship between the P balance and changes in the soil P pools (labile, moderately labile and non-labile) for all treatments. Considering the P fertilizer inputs and outputs, P mining occurred for all treatments (Fig. 5). The P balance for the two fertilized treatments showed that P mining and efficiency were greater with the broadcast compared to the deep-band treatment, with -73 and -46 kg P ka⁻¹, respectively (Fig. 5A). Therefore, in the broadcast treatment, the P recovery efficiency percentage (PRE) was 39% and the P agronomic efficiency (PAE) was 55 kg P kg⁻¹ applied versus 28% and 44 kg P kg⁻¹ applied for the deep-band treatment.

Changes in P pools were evaluated by the difference between initial soil P and after 10 years [total labile (TLP), total moderately labile (TMLP) and total non-labile (TNLP)]. The relative contribution of all fractions after 10 years are presented in the Supplemental Figure 1. The largest positive change in TLP fraction occurred for the broadcast treatment (Fig. 5B). The broadcast treatment promoted increased TLP for both positions in the 0-7.5 cm depth and in the in-row for the 7.5-15 cm depth (Fig. 5B). Deep-band promoted decreased TLP and TMLP for the 0-7.5 cm depth in the between-row and TMLP in the in-row position. In contrast, the deep-band treatment promoted increased TLP and TNLP in the in-row position in the 7.5-15 cm depth (Fig. 5B). In the control treatment, a decrease occurred for all P pools (TLP, TMLP, and TNLP) for both positions in the 0-7.5 cm depth after 10 years, but was unaffected in the 7.5-15 cm depth (Fig. 5B). These results suggest that the broadcast placement resulted in the largest P-mineralization and promoted an increase in TLP pools in the soil after 10 years in a corn-soybean rotation.

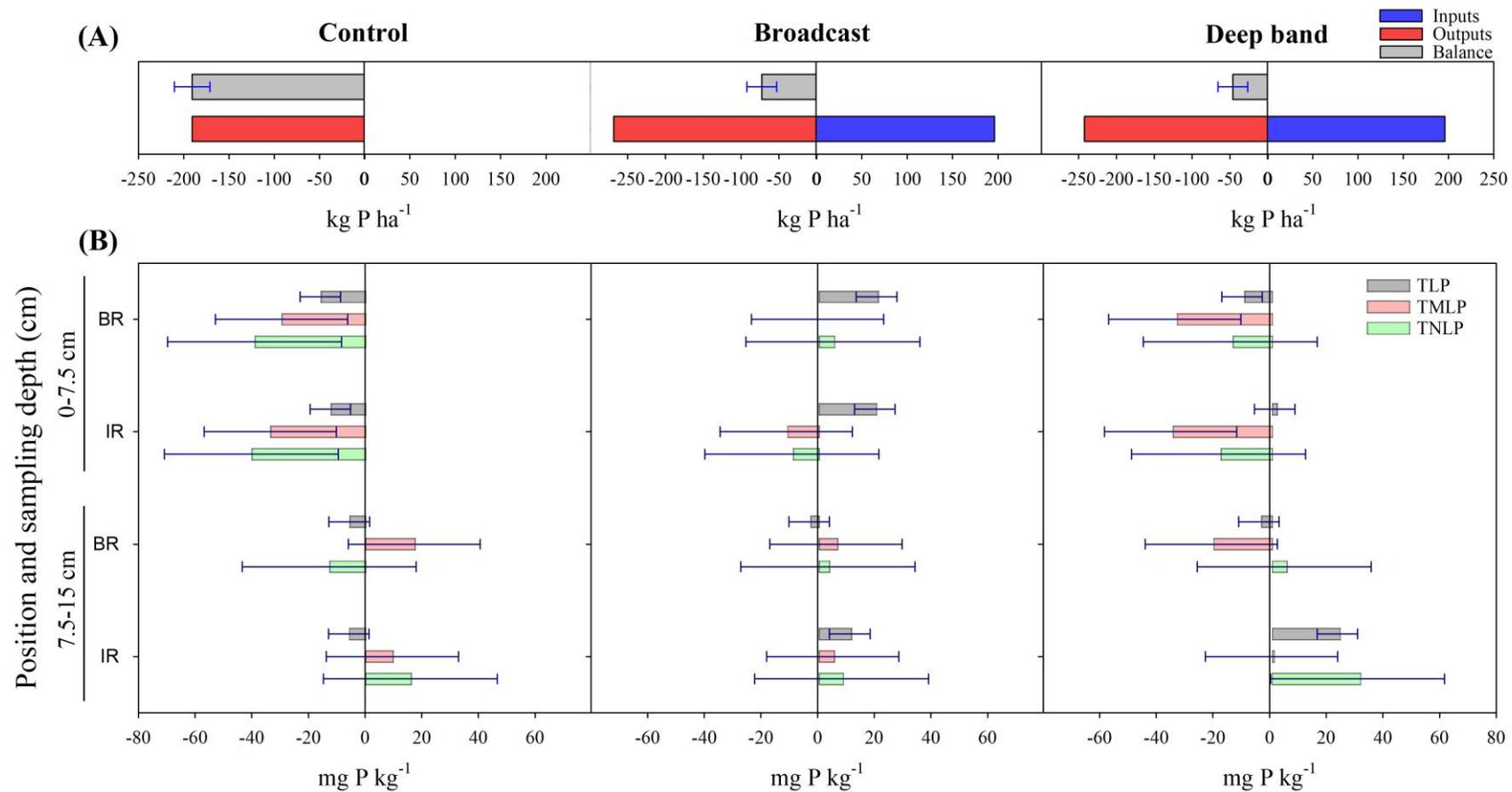


Figure 5. Legacy soil P after ten years of a corn-soybean rotation and P fertilizer treatment application; A) P inputs, outputs and balance during the ten year period of the study; B) change in soil total P for each treatment, sampling position and depth as a function of total labile P (TLP), Total moderately labile P (TMLP) and Total non-labile P (TNLP). Error bars indicate the 95% confidence limit.

3.4. DISCUSSION

3.4.1. Soil chemical properties

Ten years of reduced tillage (strip-till) in a corn-soybean rotation promoted changes in the concentration of TOC and TON in the topsoil likely due to an increase in the amount of crop residues returned to the soil surface and subsequent increased soil organic matter (SOM). The largest TOC and TON occurred in the 0-7.5 cm depth, corroborating results reported by Ortega et al. (2002). According to Stewart et al. (2008), the increase in organic C in agricultural soils occurs by increasing C inputs with minimal soil disturbance. Thus, the largest amount of the TOC in the topsoil promoted by SOM accumulation could have also promoted changes, although small, in soil pH and TON due to its influence on N mineralization. Similar results were described by Busare and Salako (2013), who showed that reduced tillage resulted in significantly greater pH and TOC in the surface soil, which was also observed by Rahman et al. (2008), who reported greater SOM and TON near the soil surface in reduced tillage practices..

3.4.2. Labile P pool (P_{AER} , P_{BiC} , and P_{OBiC})

The cumulative inorganic P fertilizer applied over 10 years promoted changes in the concentrations of the labile P pool. The greatest concentrations occurred from the broadcast treatment for both sampling positions (between-row and in-row), which was attributed to the stratification of P from the broadcast placement and the P-fertilizer source applied (granular TSP) that likely influenced P distribution in the soil. Thus, the continued addition of soluble P fertilizer via broadcast application in a reduced tillage crop system provided less contact between the soil and the fertilizer, which likely contributed to reducing large P sorption reactions and that may have contributed to increasing labile P concentrations near the soil surface. Previous research has shown that long-term P fertilization under no-tillage agriculture led to the increase the labile P pool, especially in the top soil, 0-5 cm depth (Rodrigues et al., 2016). Moreover, the presence of low-molecular-weight compounds present in SOM near the surface from crop residues might block P adsorption sites, increasing P availability (Hue, 1991; Rheinheimer and Anghinoni, 2001; Guppy et al., 2005). Soils under reduced tillage management also contribute to increased in labile P by deposition of organic/humic

substances during residue decomposition, which are also involved in the P solubilization process (Singh and Amberger, 1990; Coelho et al., 2017).

The P-fertilizer placement with deep-band (~11-15 cm depth) promoted increased inorganic P accumulation in the 7.5-15 cm soil layer in the in-row position (Fig. 2B), which was also supported by Hansel et al. (2017b). The smallest proportion of P near the soil surface and the increase in P concentration in the subsoil could also be exacerbated by root-P uptake that still occurs primarily from the soil surface (Randall and Vetsch, 2008). The deep-band may have contributed to increasing P fixed by soil components (1:1 clay and Fe oxides and hydroxides) and to decreasing labile P concentration compared to the broadcast application. However, Khatiwada et al. (2012) suggested that when P was deep-banded in a reduced-tillage system, more P remained in relatively soluble forms in soil in the absence of plants.

Previous studies have shown an increase in organic P close to the surface (Prasad et al., 2017) and analogously to increase in SOM, which also decreased with depth (Achat et al., 2009) when a no-tillage system was used for crop production. Results of the present study showed the opposite, with the greatest quantity of organic P present in deeper soil depth for the control treatment, while the other treatments had less inorganic P in deeper soil depth. For example, Spodosols presenting similar soil characteristics include horizons that became enriched with Po by vertical translocation from the topsoil (Achat et al., 2009). The Pi and Po pools act in a similar way in buffering the absorbed P by plants in soils with low or no addition of P fertilizers. The Po pool is considered as the main supply of P for plant uptake when no fertilizer is added to the soil (Gatiboni et al., 2007). The labile Po fraction (Po_{Bic}) has been considered an active fraction of soil Po because the labile Po is readily exchangeable in the soil solution as glycerophosphate and ribonucleic acid (Bowman and Cole, 1978) and as by-products of phospholipids (Jørgensen et al., 2011). The large proportion of labile Pi and the decrease in the proportion of labile Po, and vice-versa, suggests that mineralization plays a potential role in P availability and that diffusion of Po also indicates an increase in the relative importance of labile P pool for supplying P in soil solution at deeper soil depths.

3.4.3. Moderately labile P pool (Pi_{0.1Hid}, Po_{0.1Hid} and P_{HCl})

The largest overall proportion of moderately labile P was attributed to the greater concentration of Ca-precipitated P (P_{HCl}). The P_{HCl} consisted in part of P linked to apatite, other moderately soluble Ca-P compounds or negatively charged oxide surfaces (Hedley et al., 1982). These results were expected as the soil studied was characterized by a pH close to

neutral (6.9). The large base saturation, with Ca^{2+} as the main exchangeable cation in this soil, and after 10 years of cultivation might have resulted in more soluble Ca in the soil solution promoting Ca-P formation as opposed to Fe-associated P ($\text{Pi}_{0.1\text{Hid}}$). These results were in agreement with previous studies that reported a large proportion of P_{HCl} in the moderately labile pool in Mollisols soil under different tillage systems (Zamuner et al. 2008; Khatiwada et al. 2012).

The control treatment in the 7.5-15 cm depth had the greatest concentration of moderately labile Po , what may have a similar explanation to that for the labile Po fraction. The broadcast treatment had the greatest concentration of moderately labile Pi in the 0-7.5 cm depth (Fig. 3). As result of this tradeoff in P fractions, the large proportion of moderately Pi and the decrease in the proportion of moderately Po , and vice-versa, the total moderately labile pool showed no difference when compared the control and the broadcast treatments. The moderately labile P pool is important for soil-P buffering and the larger concentration in the broadcast treatment may have contributed to the large concentration of the labile Pi pool. When P depletion occurs in one fraction, another fraction may be desorbed to replace P as well as maintain the plant-P supply (Pavinato et al., 2009). Similarly, Gatiboni et al. (2007) showed that as the labile-P fraction is depleted, the moderately labile P acts as buffer for the soil solution.

3.4.4. Non-labile P pool ($\text{Pi}_{0.5\text{Hid}}$, $\text{Po}_{0.5\text{Hid}}$, and $\text{P}_{\text{Residual}}$)

Long-term cumulative fertilization promoted increased non-labile P, which represented the largest pool of the total soil P. Fertilizer P use, both broadcast or deep band, increased Pi in the non-labile pool, which agreed with the MPAC (288 mg kg^{-1}) indicating a medium P adsorption capacity for the soil. The MPAC may be attributed to a slight to intermediate degree of weathering of this soil studied, with on average 25% clay, and large Ca concentration, moderate to large organic matter and Al- and Fe- oxides and hydroxides and 1:1 clay mineralogy. In addition, the greater total non-labile P was showed at the in-row sampling position in the 7.5-15 cm depth, which could be explained by the deep-band placement that increased the concentration of P from the fertilizer, as well the specific adsorption and subsequent reduction in more labile and moderately labile pools.

Non-labile Po consisted of organic P associated with fulvic and humic acids inside aggregates extracted by 0.5 M NaOH (Condon et al., 1985). Under these circumstances, the

greater quantity of organic matter near the soil surface likely resulted in a greater P_o fraction for the between-row position in the top 0-7.5 cm depth.

3.4.5. Legacy soil P

The largest amount of P for most fractions evaluated (i.e., labile and moderately labile P) occurred in the broadcast treatment, likely contributed to the greater P recovery efficiency (i.e., about 11% more than for the deep-band treatment) and consequently greater P agronomic efficiency. Despite P recovery by crops rarely exceeds 25% and frequently ranging between 10-15% of the P applied (Roberts and Johnston, 2015) or 10-20% overall (Cornish, 2009), results of this study showed that the broadcast treatment was able to increase P recovery efficiency after 10 years of crop production. In addition, the broadcast treatment had the largest soil legacy P accumulated in the labile P fractions as well as moderately labile fractions. Typically, a significant proportion of legacy P occurs in forms that are potentially available for crop uptake, especially in temperate soils (Rowe et al., 2016).

The trend of broadcast-P accumulation suggests the possibility of reducing P fertilization requirements over time by increasing labile P pools. Furthermore, results showed that the changes over time in available P were more sensitive than the changes in total P to continuous P fertilization and depletion by plant uptake. The preferential accumulation of labile P in temperate soils has recently been attributed to the blocking of P immobilization pathways when P sorption sites become more and more negatively charged as they saturate with phosphate (PO_4^{3-}) (Barrow and Debnath, 2014). With reduced P adsorption, more of the P added can remain in a labile form (Rodrigues et al., 2016). This situation enhances the redistribution of P pools over time, resulting in increased P efficiency (Herrera et al., 2016). For the deep-band treatment, the largest legacy P accumulation was at the in-row position in the 7.5-15 cm depth due to P-fertilizer placement over time. Results suggested that even with large labile legacy P accumulation with the deep-band treatment, P recovery efficiency and P agronomic efficiency were still greater for broadcast P application in this temperate soil.

3.5. CONCLUSIONS

Long-term Long-term P fertilizer placement resulted in a large amount of legacy P in the soil, with larger differences between the treatments in labile and moderately labile pools.

The broadcast P fertilizer placement increased labile and moderately labile P pools in the topsoil (0-7.5 cm) and likely resulted in more P recycling, promoting greater P use efficiency as compared to the deep-band placement. Furthermore, crop P uptake was more active for the 0-7.5 cm soil layer, regardless of P-fertilizer placement, which resulted in significant depletion of all P pools in the upper layer when fertilizer was applied as deep band in the 7.5-15 cm layer or the control with no P fertilizer application. The supplemental irrigation with no water restriction may have contributed to this results.

Phosphorus fertilizer placement via broadcast application over multiple years resulted in legacy soil P in available fractions, which can potentially contribute as a secondary source of P for plant uptake and possibly substituting, at least in part, for future fertilizer applications programs to help to mitigate environmental risk. Moreover, increased of organic carbon in the soil surface that might have affected the performance of P fertilizer placement and soil P dynamics, as well as final P uptake by the crop.

Results of this study suggest that placing P fertilizer exclusively using one strategy over many years can promote depletion of P pools from some soil layers and row locations. Therefore, crop production in the field might benefit from a combined P fertilizer placement strategy, with changes in the position of crop rows and the location of band-applied P fertilizers. Allocating the total recommended P fertilizer rate in various placement methods, such as a combination of deep-band, starter fertilizer with the planter, and broadcast, might minimize depletion of available P pools from certain soil layers or row positions, while maintaining or increasing plant P uptake.

REFERENCES

- Adee, E., Hansel, F.D., Ruiz Diaz, D.A., Janssen, K., 2016. Corn response as affected by planting distance from the center of strip-till fertilized rows. *Front Plant Sci.* 7, 1232-1241.
- Achat, D.L., Bakker, M.R., Augusto, L., Saur, E., Dousseron, L., Morel, C., 2009. Evaluation of the phosphorus status of P-deficient podzols in temperate pine stands: combining isotopic dilution and extraction methods. *Biogeochemistry*, 92, 183–200.
- Anghinoni, I., Barber, S.A., 1980. Phosphorus influx and growth characteristics of corn roots as influenced by phosphorus supply. *Agron. J.* 72, 685–688.
- Barrow, N.J., Debnath, A., 2014. Effect of phosphate status on the sorption and desorption properties of some soils of northern India. *Plant Soil*, 378, 383-395.

- Bowman, R.A., Cole, C.V., 1978. An exploratory method for fractionation of organic phosphorus from grassland soils. *Soil Sci.* 125, 95–101.
- Brookes, P.C., Powlson, D.S., 1981. Preventing phosphorus losses during perchloric acid digestion of sodium bicarbonate soil extracts. *J. Sci. Food Agric.* 32, 671–674.
- Bünemann, E.K., 2015. Assessment of gross and net mineralization rates of soil organic phosphorus - A review. *Soil Biol. Biochem.* 89, 82-98.
- Busari, M.A., Salako, F.K., 2013. Effect of tillage, poultry manure and NPK fertilizer on soil chemical properties and maize yield on an Alfisol at Abeokuta, south-western Nigeria. *Nigerian J. Soil Sci.* 23, 206-218
- Cassidy, R., Doody, D.G., Watson, C.J., 2017. Impact of legacy soil phosphorus on losses in drainage and overland flow from grazed grassland soils. *Sci. Total Environ.* 575, 474–484.
- Chowdhury, R.B., Moore, G.A., Weatherley, A.J., Arora, M., 2017. Key sustainability challenges for the global phosphorus resource, their implications for global food security, and options for mitigation. *J. Cleaner Prod.* 140, 945-963.
- Coelho, M.J.A., Aguiar, A.C.F., Sena, V.G.L., Moura, E.G., 2017. Utilization and fate of phosphorus of different sources applied to cohesive soil of Amazonian periphery. *Sci. Agric.* 74, 242-249.
- Condon, L.M., Spears, B.M., Haygarth, P.M., Turner, B.L., Richarddson, A.E., 2013. Role of legacy phosphorus in improving global phosphorus-use efficiency. *Environ. Dev.* 8,147-148.
- Condon, L.M., Newman, S., 2011. Revisiting the fundamentals of phosphorus fractionation of sediments and soils. *J. Soils Sediments*, 11, 830-840.
- Condon, L.M., Goh, K.M., Newman, R.H., 1985. Nature and distribution of soil phosphorus as revealed by a sequential extraction method followed by ³¹P nuclear magnetic resonance analysis. *Eur. J. Soil Sci.* 36, 199–207.
- Cornish, P., 2009. Research directions: improving plant uptake of soil phosphorus, and reducing dependency on input of phosphorus fertiliser. *Crop Pasture Sci.* 60, 190–196.
- Cross, A.F., Schlesinger, W.H., 1995. A literature review and evaluation of the Hedley fractionation: applications to the biogeochemical cycle of soil phosphorus in natural ecosystems. *Geoderma*, 64,197–214.
- Dick, W.A., Tabatabai, M.A., 1977. Determination of orthophosphate in aqueous solutions containing labile organic and inorganic phosphorus compounds. *J. Environ. Qual.* 6, 82–85.

- Dodd, J.R., Mallarino, A.P., 2005. Soil-test phosphorus and crop grain yield responses to long-term phosphorus fertilization for corn-soybean rotations. *Soil Sci. Soc. Am. J.* 69, 1118-1128.
- Eriksson, A.K., Gustafsson, J., Hesterberg, D., 2015. Phosphorus speciation of clay fractions from long-term fertility experiments in Sweden. *Geoderma*, 241-242, 68-74.
- Fageria, N.K., 2001. Nutrient management for improving upland rice productivity and sustainability. *Commun. Soil Sci. Plant Anal.* 32, 2603-2629.
- Frank, K., Beegle, D., Denning, J., 1998. Phosphorus. In: J.R. Brown (Eds.), *Recommended chemical soil test procedures for the North Central Region*. North Central Region. Missouri Agric. Exp. Stn., Columbia.
- Gatiboni, L.C., Kaminski, J., Rheinheimer, D. S., Flores, J.P.C., 2007. Biodisponibilidade de formas de fósforo acumuladas em solo sob Sistema plantio direto (Bioavailability of soil phosphorus forms in no-tillage system). *Rev. Bras. Ciênc. Solo (Braz. J. Soil Sci.)*, 31, 691–699.
- Guppy, C.N., Menzies, N.W., Moody, P.W., Blamey, F.P.C., 2005. Competitive sorption reactions between phosphorus and organic matter in soil: a review. *Soil Research*, 43, 189-202.
- Hansel, F.D., Amado, T.J.C., Ruiz-Diaz, D.A., Rosso, L.H.M., Nicoloso, F.T., Schorr, M., 2017a. Phosphorus fertilizer placement and tillage affect soybean root growth and drought tolerance. *Agron. J.* 109, 1-9.
- Hansel, F.D., Ruiz-Diaz, D.A., Amado, T.J.C., Rosso, L.H.M., 2017b. Deep banding increases phosphorus removal by soybean grown under no-tillage production systems. *Agron. J.* 109, 1091-1098.
- Havens, K.E., James, R.T., 2005. The phosphorus mass balance of Lake Okeechobee, Florida: Implications for eutrophication management. *Lake Reservoir Manage.* 21, 139-148.
- Hedley, M.J., Stewart, J.W.B., Chauhan, B.S., 1982. Changes in inorganic and organic soil phosphorus fractions induced by cultivation practices and by laboratory incubations. *Soil Sci. Soc. Am. J.* 46, 970–976.
- Herrera, W.F.B., Rodrigues, M., Teles, A.P.B., Barth, G., Pavinato, P.S., 2016. Crop yield and soil phosphorus lability under soluble and humic-complexed phosphate fertilizers. *Agron. J.* 108, 1692-1702.
- Hue, N.V., 1991. Effects of organic acids/anions on P sorption and phytoavailability in soils with different mineralogies. *Soil Sci.* 152, 463–471.

- Jørgensen, C., Jensen, H.S., Andersen, F.Ø., Egemose, S., Reitzel, K., 2011. Occurrence of orthophosphate monoesters in lake sediments: significance of myo- and scyllo-inositol hexakisphosphate. *J. Environ. Monit.* 13, 2328-2334.
- Kamprath, E.J., 1999. Changes in phosphate availability of Ultisols with long-term cropping. *Commun. Soil Sci Plant Anal.* 30, 909–919.
- Khaliwada, R., Hettiarachchi, G.M., Mengel, D.B., Fei, M., 2012. Speciation of phosphorus in a fertilized, reduced-till soil system: in-field treatment incubation study. *Soil Sci. Soc. Am. J.* 76, 2006-2018.
- Kilmer, V.J., Alexander, L.T., 1949. Methods of making mechanical analysis of soils. *Soil Sci.* 68, 15-24.
- LECO TruSpec CN Carbon/Nitrogen combustion analyzer reports total levels (inorganic and organic) of C and N on a weight percent basis, according to the TruSpec CN instrument method "Carbon, Hydrogen, and Nitrogen in Flour and Plant Tissue", LECO Corporation, St. Joseph, MI, 2005.
- Linquist, B.A., Singleton, P.W., Cassman, K.G., 1997. Inorganic and organic phosphorus dynamics during a build-up decline of available phosphorus in an ultisol. *Soil Sci.* 162, 254–264.
- Littell, R.C., Milliken, G.A., Stroup, W.W., Wolfinger, R.D., Schabenberber, O., 2006. SAS for mixed models, second ed. SAS Inst., Cary, North Carolina.
- Luo, L., Ma, Y., Sanders, R.L., Xu, C., Li, J., Myneni, S.C.B., 2017. Phosphorus speciation and transformation in long-term fertilized soil: evidence from chemical fractionation and P K-edge XANES spectroscopy. *Nutr. Cycl. Agroecosyst.* 107, 215-226.
- Mallarino, A.P., Schwarte, K., Havlovic, B.J., 2012. Broadcast and band phosphorus and potassium placement for corn and soybean managed with till or no till. Iowa State Research Farm Progress Reports. 3. http://lib.dr.iastate.edu/farms_reports/3/ (accessed 27 March 2019)
- Murphy, J., Riley, J.P., 1962. A modified single solution method for the determination of phosphate in natural waters. *Anal. Chim. Acta*, 27, 31–36.
- Ortega, R.A., Peterson, G.A., Westfall, D.G., 2002. Residue management and changes in soil organic matter as affected by cropping intensity in no-till dryland agroecosystems. *Agron. J.* 94, 944–954.
- Pavinato, P.S., Merlin, A., Rosolem, C.A., 2009. Phosphorus fractions in Brazilian Cerrado soils as affected by tillage. *Soil Tillage Res.* 105, 149-155.

- Prasad, R., Singh Shivay, Y., Majumdar, K., Prasad, S., 2017. Phosphorus management. In: Lal, R., Stewart, B.A., (Eds.), Soil Phosphorus. CRC Press, Boca Raton, Florida, pp. 81–113.
- Preston, C.L., Ruiz Diaz, D.A, Mengel, D., 2018. Corn response to long-term phosphorus fertilizer application rate and placement with strip-tillage. *Agron J. in press*
- Rahman, M.H., Okubo, A., Sugiyama, S., Mayland, H.F., 2008. Physical, chemical and microbiological properties of an Andisol as related to land use and tillage practice. *Soil Tillage Res.* 101, 10-19.
- Randall, G.W., Vetsch, J., 2008. Optimum placement of phosphorus for corn/soybean rotations in a strip-tillage system. *J. Soil Water Conserv.* 63, 152A–153A.
- Reijnders, L., 2014. Phosphorus resources, their depletion and conservation, a review. *Resour. Conserv. Recycl.* 93, 178-187.
- Rheinheimer, D.S., Anghinoni, I., 2001. Distribuição do fósforo inorgânico em sistemas de manejo de solo (Distribution of inorganic phosphorus fraction in soil management systems). *Braz. J. Agric. Res.* 36, 151–160.
- Roberts, T.L., Johnston, A.E., 2015. Phosphorus use efficiency and management in agriculture. *Resour. Conserv. Recycl.* 105, 275–281.
- Rodrigues, M., Pavinato, P.S., Withers, P.J.A., Teles, A.P.B., Herrera, W.F.B., 2016. Legacy phosphorus and no tillage agriculture in tropical oxisols of the Brazilian savanna. *Sci. Total Environ.* 542, 1050–1061.
- Rowe, H., Withers, P.J.A., Baas, P., Chan, N.I., Doody, D., Holiman, J., Jacobs, B., Li, H., MacDonald, G.K., McDowell, R., Sharpley, A.N., Shen, J., Taheri, W., Wallenstein, M., Weintraub, M.N., 2016. Integrating legacy soil phosphorus into sustainable nutrient management strategies for future food, bioenergy and water security. *Nutr. Cycl. Agroecosyst.* 104, 393-412.
- Sattari, S.Z., Bouwman, A.F., Giller, K.E., van Ittersum, M.K., 2012. Residual soil phosphorus as the missing piece in the global phosphorus crisis puzzle. *Proc. Natl. Acad. Sci.* 109, 6348-6353.
- Singh, C.P., Amberger, A., 1990. Humic substances in straw compost with rock phosphate. *Biological Wastes*, 31, 165-174.
- Sleight, D.M., Sander, D.H., Peterson, G.A., 1984. Effect of fertilizer phosphorus placement on the availability of phosphorus. *Soil Sci. Soc. Am. J.* 48, 336-340.
- Shen, J., Yuan, L., Zhang, J., Li, H., Bai, Z., Chen, X., Zhang, W., Zhang, F., 2011. Phosphorus dynamics: from soil to plant. *Plant Physiol.* 156, 997-1005.

- Soil Survey Staff. 2014. Keys to Soil Taxonomy, twelfth ed. Washington, District of Columbia.
- Soil Survey Laboratory Staff. 2004. Soil survey laboratory methods manual. National Soil Survey Center, Lincoln, Nebraska.
- Song, C., Han, X.Z., Tang, C., 2007. Changes in phosphorus fractions, sorption and release in Udic Mollisols under different ecosystems. *Biol. Fertil. Soils*. 44, 37-47.
- Stewart, C.E., Plante, A.F., Paustian, K., Conant, R.T., Six, J., 2008. Soil carbon saturation: linking concept and measurable carbon pools. *Soil Sci. Soc. Am. J.* 72, 379-392.
- Stutter, M.I., Shand, C.A., George, T.S., Blackwell, M.S.A., Dixon, L., Bol, R., Mackay, R.L., Richardson, A.E., Condon, L.M., Haygarth, P.M., 2015. Land use and soil factors affecting accumulation of phosphorus species in temperate soils. *Geoderma*, 257-258, 29-39.
- Tiessen, H., Moir, J.O., 1993. Characterization of available P by sequential extraction. In: Carter, M.R. (Eds), *Soil Sampling and Methods of Analysis*. Canadian Society of Soil Science. Lewis Publications, Boca Raton, FL, pp. 75–86.
- USEPA - United States Environmental Protection Agency, 1971. *Methods of chemical analysis for water and wastes*. Environmental Protection Agency, Cincinnati, US.
- Vu, D.T., Tang, C., Armstrong, R.D., 2008. Changes and availability of P fractions following 65 years of P application to a calcareous soil in a Mediterranean climate. *Plant Soil*, 304, 21-33.
- Vu, D.T., Tang, C., Armstrong, R.D., 2007. Changes and availability of P fractions following 65 years of P application to a calcareous soil in a Mediterranean climate. *Plant Soil*, 304, 21-33.
- Warncke, D., Brown, J.R., 1998. Potassium and other cations. In: J.R. Brown (Eds.), *Recommended chemical soil test procedures for the North Central Region*. North Central Region. Missouri Agric. Exp. Stn., Columbia.
- Watson, M.E., Brown, J.R., 1998. Boron. p. 45–48. In: J.R. Brown (ed.) *Recommended chemical soil test procedures for the North Central Region*. North Central Reg. Res. Publ. 221 (Rev.) SB 43 1001. Missouri Agric. Exp. Stn., Columbia, pp. 31-34.
- Weihrauch, C., Opp, C., 2018. Ecologically relevant phosphorus pools in soils and their dynamics: The story so far. *Geoderma*, 325, 183-194.
- Withers, P.A.J., Sylvester-Bradley, R., Jones, D.L., Healey, J.R., Talboys, P.J., 2014. Feed the crop not the soil: Rethinking phosphorus management in the food chain. *Environ. Sci. Technol.* 48, 6523–6530.

- Whitney, D.A., 1998. Micronutrients: Zinc, Iron, Manganese and Copper. In Recommended chemical soil test procedures for North Central Region. North Central Regional, Missouri Agric. Exp. Stn. Columbia, pp. 41-44.
- Zamuner, E.C., Picone, L.I., Echeverria, H.E., 2008. Organic and inorganic phosphorus in Mollisol soil under different tillage practices. *Soil Tillage Res.* 99, 131-138

4. MANAGEMENT OF PHOSPHATE FERTILIZER FOR GRAIN PRODUCTION INFLUENCING SOIL LEGACY PHOSPHORUS IN BRAZILIAN CERRADO

ABSTRACT

The Brazilian cerrado is one of the last viable alternatives for expansion of production with high agricultural potential. Therefore, to increase agricultural efficiency and preserve the native savanna by converting degraded pasture to agricultural crop production, phosphorus fertilizer placements can affect the bioavailability forms and pools of P in the soil. Our objective was to evaluate the effects of long-term P fertilizer placement (initial placement and annual rates and placement of P) in the transition of pasture to double-crop of soybean and corn under no-till crop production on changes in the soil P pools (labile, moderately labile and non-labile) and legacy soil P accumulation. A field study was conducted for five consecutive years (crop seasons from 2010/11 to 2014/15) of a double-crop of soybean and corn under no-till crop production on an Oxisol. The treatments consisted of P fertilizer management and rates combinations: initial phosphate fertilization placement with four forms, including a control (0) with no P fertilizer application, and three fertilizer placements (200 kg P₂O₅ ha⁻¹ / 87 kg P ha⁻¹): (1) surface broadcast (S), (2) incorporated to 20 cm (I20) and (3) incorporated to 40 cm (I40) application at the implantation of the experiment in 2010; and annual phosphate fertilization rates and placements with five combinations, including a control (0A) with no P fertilizer application during the 5 years of the study and two rates (50 kg P₂O₅ ha⁻¹ / 22 kg P ha⁻¹ and 100 kg P₂O₅ ha⁻¹ / 22 kg P ha⁻¹) combined with two placements (band and broadcast): (1) band + 50 (50B), (2) band+100 (100B), (3) broadcast+50 (50SB) and (4) broadcast+100 (100SB) application for each year of cultivation. After the last corn harvest (2015), soil samples were collected from the 0-10, 10-20 and 20-40 cm and analyzed for soil P pools via a sequential P fractionation. Results of P pools in the soil showed a significant effect of P application on labile P fractions in soil surface with the adoption of annual rates and placement of 100B and 100SP kg P₂O₅ ha⁻¹ under all initial P application, and 100B for the all others P fractions and soil depths, while effects of the initial P application were less significant. Thus, P accumulation from the initial (P remains in the soil after five years) and annual P fertilizer application in the labile P pools could help (at least in part) to reduce current high dependency and large inputs of P fertilizer in no-till systems.

Keywords: Initial P placements; Annual P placements; No-till tillage; Phosphorus fractionation

4.1. INTRODUCTION

The Brazilian Cerrado biome consists in 203.4 M ha with 49.4 M ha cultivated with pastures (EMBRAPA 1999). However, this area used extensive beef cattle production on unimproved pastures (Lopes and Guilherme, 2016) correspond about 25 to 32 M ha (EMBRAPA 1999, De Oliveira et al., 2004) which compromising productivity and generating economic and environmental issues. Thus, the Cerrado biome is one of the last viable alternative areas for expansion of production with high agricultural potential. Therefore, to increase agricultural efficiency, mitigate environmental issues, and preserve the native

savanna by converting degraded pasture to agricultural lands. Although, in the higher weathered soil, such as Cerrado soils, one of the most limiting nutrients for agricultural production is the phosphorus (P) availability (Vence et al., 2003; Teles et al., 2017) due to the natural deficiency plus the severe P fixation capacity.

The higher fixation of P in most tropical soils is attributed to the higher amount of Fe and Al oxy-hydroxides (mainly goethite, hematite, gibbsite, and amorphous Fe and Al oxides) and 1:1 clay silicate such as kaolinite (Fontes and Weed, 1996, Rodrigues et al., 2016). In addition, the higher concentration of Fe and Al in the soil solution further enhance the retention capacity by precipitation of Fe-P and Al-P and decrease P availability for plant uptake (Dhillon et al., 2017). To supply this lack of P availability, farmers often apply phosphate fertilizers beyond plant requirements. Usually, just 10-20% of the P applied as fertilizer is absorbed by the plants in short-term growing season (Vu et al., 2008). Thus, the addition of P as inorganic phosphate fertilizer is the principal P source for plant uptake in agricultural systems (Rodrigues et al., 2016; Withers et al., 2017), but P is a non-renewable resource, and it might run out soon (Cordell and White, 2013; Reijnders, 2014). Therefore, strategies of management of cropping systems and phosphate placement can reduce the inputs of P fertilizer, also increasing soil P availability and use that P more efficiently by crops.

The conservation tillage such a no-till (NT) is developed during the last 20 years in Brazilian Cerrado area (Pavinato et al., 2009) but comparison between the conventional tillage and NT associated to phosphate fertilizer placement on soil P fractions and its availability is scarce, mostly evaluating similar dosage and/or in the same P management (Pavinato et al., 2009; Zumuner et al., 2008; Tiecher et al., 2012; Rodrigues et al., 2016). Moreover, is poorly understood the dynamics of P under effect of placement methods and long-term rates under NT in tropical soils.

Commonly, there is an integration of initial high rates of P fertilizer addition, designed to improve the general P level of the soil, with annual crop rates of P fertilizer to supply the plant demand. The initial P application can be placed as superficial broadcast or incorporated in the soil profile, and the most common annual application is broadcast and band placements. However, no studies under NT so far have evaluated the best combination of phosphate placement on P availability and dynamics in soil in long-term. Thus, phosphate fertilizing may change substantially the remaining forms of P in the soil, both inorganic (Pi) and organic (Po), called residual P (Condrón et al., 2013). Yet, at a time scale, the correct management of tillage system and P fertilizer placement may increase the access of the plants to P and reduce fertilizer inputs.

Soil P availability and solubility in tropical soils is usually studied through chemical fractionation into Po and Pi forms that differ in their availability for plant acquisition using different extractors (Pavinato et al., 2009; Ciampitti et al., 2011). The P fractionation developed by Hedley et al. (1982) and adapted by Cross and Schlesinger (1995) has been the most applied approach to identify and interpret this P dynamics in the soil.

We hypothesized here that initial P fertilization as phosphating acts as a buffer of soil labile P levels, reducing its respective adsorption when applied at planting. Also, annual P fertilization via broadcast or band has a similar response in soil labile P levels, in no-tillage system. Thus, the objective of this study was to evaluate the effects of long-term P fertilizer placement (initial and annual rates and placement of P) in transition of pasture to double-crop of soybean and corn under no-till crop production on changes in the soil P pools (labile, moderately labile and non-labile) and legacy soil P accumulation.

4.2. MATERIALS AND METHODS

4.2.1. Site description and soil characterization

A long-term field experiment, established in the Cerrado biome, was conducted over five successive years (crop season from 2010/11 to 2014/15) at an Oxisols classified as Rhodic Haplustox (Soil Survey Staff, 2014), and as Latossolo Vermelho distrófico típico in Brazilian classification (EMBRAPA, 2013), located in Itiquira, Mato Grosso, BR (16°37'23" S, 54°28'26" O, altitude 262 m above sea level). The regional climate is classified as Aw – Tropical, with mean annual precipitation of 1559 mm, with a well-defined rainy season from October to April and a dry season with very little rain from May to September and mean annual temperature of 23.7 °C. This site had a history of extensive pasture (*Brachiaria decumbens*) for more than two decades before the establishment of the study. The experiment comprised a no-till system under double-crop soybean and corn production with five years for soybean (three first years with a single crop for soybean) and two last years for corn (double crop soybean-corn). In addition, to the transition of pasturelands to the no-till system, millet was planted as cover crop in two consecutive years (2008/09 and 2009/10) before the implantation of the study and in the third year of the study after corn (2012/13).

Initial soil samples were collected in November of 2008, before initiating the study, by collecting a representative sample from the 0–20 and 20–40 cm layers for the characterization of general soil properties (Table 1). The P level (Mehlich-1), soil organic matter (SOM), pH

(CaCl₂), exchangeable acidity (H+Al), aluminum (Al), potassium (K), calcium (Ca) and magnesium (Mg) content and aluminum saturation index (m) were determined according to the methodologies proposed by EMBRAPA (1997) and sum of bases (BS) was obtained by summing K, Ca and Mg (mmol_c kg⁻¹), cation exchange capacity (CEC) was obtained by summing the exchangeable cations (BS + H+Al, mmol_c kg⁻¹), and bases saturation index (V) was obtained by (BS/CEC)*100. The granulometric analysis was determined by pipette method after dispersion in 1 mol L⁻¹ NaOH, and dry bulk density was also determined, both following procedures described in EMBRAPA (1997). The levels of Fe and Al were determined by extraction with Ditionite, citrate, and bicarbonate (DCB) and acidic ammonium oxalate (Oxalate) from initial soil. The amorphous/crystalline Fe ratio (FeOxalate/FeDCB) lower than 0.05 (0.041) indicates that in this soil the crystalline oxides are predominant (Table 1), characteristics of soils in advanced weathering stage (Kämpf et al., 2000).

Maximum P adsorption capacity (MPAC) and X-ray diffraction (XRD) were analyzed from a sample collected in the control treatment (0-40 cm) after the last crop harvest (2015), the results are shown in Table 1. To evaluate MPAC, soil subsamples of 2.5g were mixed with eleven rates of added P (0, 5.5, 11, 16.5, 27.5, 38.5, 49.5, 60.5, 77, 93.5, and 110 mg L⁻¹) as KH₂PO₄ (p.a.), in a 25 mL equilibrium solution of calcium chloride (0.01 M CaCl₂.2H₂O). They were shaken for 24 h at 147 rpm and centrifuged for 15 min at 3278 g, and the P concentration was measured in the supernatant by the blue-molybdate colorimetric method (Murphy and Riley, 1962). Final MPAC value was obtained after fitted the Langmuir equation, described by: $P_{so} = [k \times (P_{max} - P_{ce}) / (1 + K \times P_{ce})]$, where P_{so} is the amount of P sorbed in each soil sample, k is a constant related to the sorption energy, P_{max} is the maximum P sorption capacity, and P_{ce} is the equilibrium P concentration in solution. The estimated MPAC was 826 mg P kg⁻¹ of soil (Table 1), supported by the presence of Fe and Al sesquioxides in the mineralogical composition, as confirmed by X-ray diffraction (XRD).

Table 1 Initial soil chemical and physical properties of the soil surface layers (0-20 and 20-40 cm depth) before the experiment establishment

Soil layer	P ⁽¹⁾	SOM ⁽²⁾	pH	H+Al ⁽³⁾	Al	K	Ca	Mg	BS ⁽⁴⁾	CEC ⁽⁵⁾	V ⁽⁶⁾	m ⁽⁷⁾
cm	mg kg ⁻¹	g kg ⁻¹	CaCl ₂	----- mmol _c kg ⁻¹ -----						%		
0-20	3,7	35	4.5	50	4	9.2	17	10	36.2	86.2	42	10
20-40	0.9	36	4.3	56	7	1.3	12	8	21.3	77.3	28	25
			Granulometry			DCB ⁽⁸⁾		Oxalate ⁽⁹⁾		SBD ⁽¹⁰⁾	MPAC ⁽¹¹⁾	
		<u>Sand</u>	<u>Silt</u>	<u>Clay</u>	Fe	Al	Fe	Al				
		----- g kg ⁻¹ -----								g cm ⁻³	mg kg ⁻¹	
0-20		222	91	687	75.3	6.5	3.1	*			1.23	826
20-40		217	55	728							1.25	

⁽¹⁾ Phosphorus extracted by Mehlich-1; ⁽²⁾ Soil organic matter; ⁽³⁾ Exchangeable acidity; ⁽⁴⁾ Sum of bases; ⁽⁵⁾ Cation exchange capacity; ⁽⁶⁾ Bases saturation index; ⁽⁷⁾ Aluminum saturation index; ⁽⁸⁾ Dithionite, citrate and bicarbonate extraction; ⁽⁹⁾ Ammonium oxalate extraction; ⁽¹⁰⁾ Soil bulk density; ⁽¹¹⁾ Phosphorus maximum adsorption capacity; *not determined

To identify the main minerals present in soil, XRD analysis of the clay fraction was run following the method described by Jackson (1979). A set of different post-treatments were prepared for each soil sample, with the clay fraction after removing Fe oxides. The post-treatments consisted of saturating the clay fraction with 1 M KCl or 1 M MgCl₂ in order to saturate inter-layers and then allow the identification of phyllosilicates. The following post-treatments applied were: K saturation at room temperature (25°C); K saturation and heating up to 300°C; K saturation and heating up to 500°C; Mg saturation at room temperature (25°C); Mg saturation with solvation using ethylene glycol. Another sample of clay fraction with Fe was prepared to identify the Fe oxides present in the soils, followed by the oriented glass slide preparation. The diffractograms were collected in a Miniflex II Desktop X-Ray Diffractometer (RIGAKU), CuK α radiation, analysis interval from 10 to 50°2 θ to the clay fraction without Fe samples and 5 to 30°2 θ to the samples of clay + Fe. The results (Fig. 1) show the dominance of Kaolinite as the only phyllosilicate and the presence of the Hematite and Goethite as the Fe oxides and Gibbsite as Al hydroxide. These minerals will be referred conjunctly among this text as Al and Fe oxides.

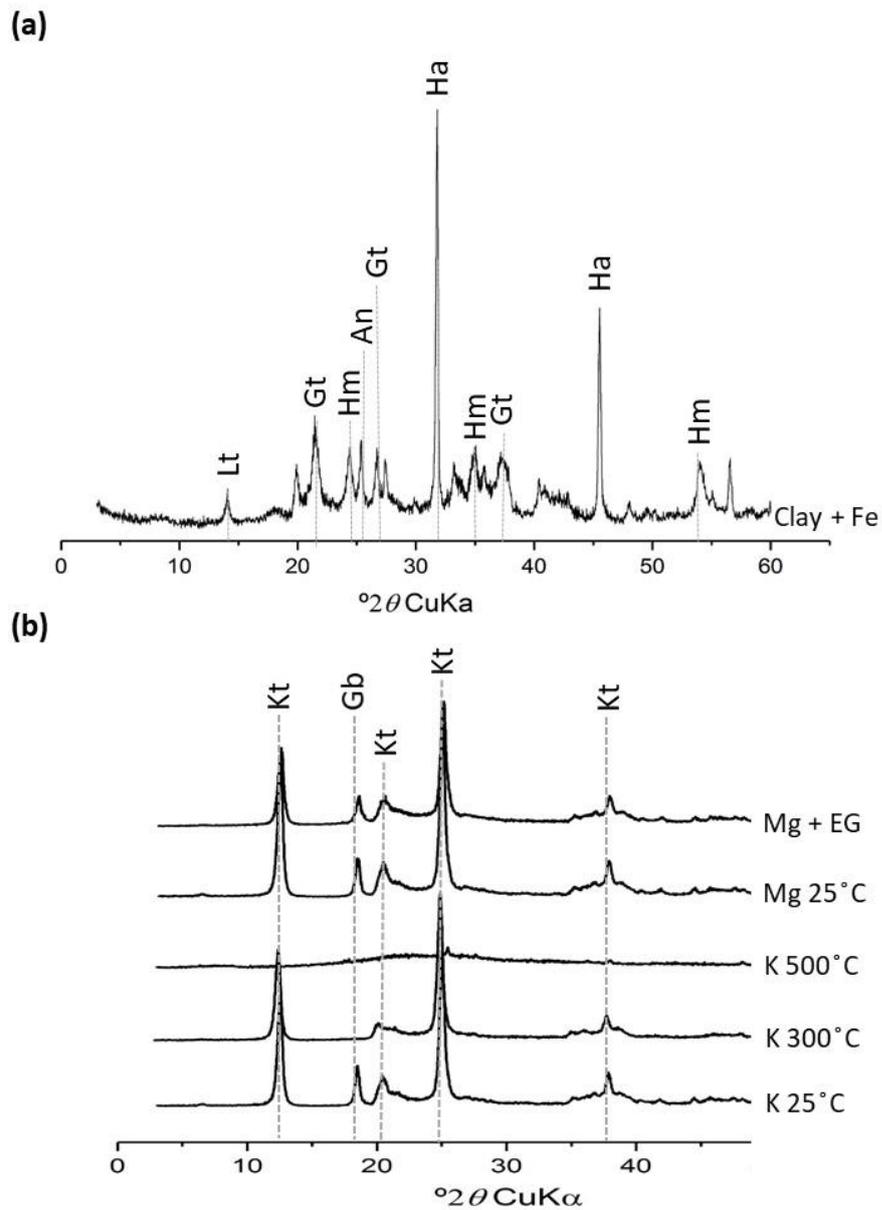


Figure 1. X-ray diffraction of total clay: (a) Clay + Fe and (b) clay free of Fe [K^+ at room temperature (K 25 °C), heated 300 °C (K 300 °C) and 500 °C (K 500 °C); Mg^{2+} at room temperature (Mg 25 °C) and solvated with ethylene glycol (Mg + EG)], oriented blades. Identified minerals: Anatase (An, Gibbsite (Gb), Goethite (Gt), Halite (Ha), Hematite (Hm), Kaolinite (Kt) and Lepidocrocite (Le).

4.2.2. Experimental design and management practices

The experiment was set up as a randomized complete block design with three replications and 20 treatments. Individual plots size was 6.3 by 10 m with a 3.0 m alley between the plots, totaling 63 m². The treatments consisted of P fertilizer management and rates combinations: initial phosphate fertilization placement applied at the implantation of the experiment in 2010 in four distinct forms, including a control (0) with no P fertilizer application, and three fertilizer placements (200 kg P₂O₅ ha⁻¹ / 87 kg P ha⁻¹): (1) surface broadcast (S), (2) incorporated to 20 cm (I20) and (3) incorporated to 40 cm (I40); and annual phosphate fertilization rates and placements with five combinations, including a control (0A) without P fertilizer application and two rates (50 kg P₂O₅ ha⁻¹/22 kg P ha⁻¹ and 100 kg P₂O₅ ha⁻¹/22 kg P ha⁻¹) combined with two placements (band and superficial broadcast): (1) band + 50 (50B), (2) band+100 (100B), (3) superficial broadcast + 50 (50SB) and (4) superficial broadcast + 100 (100SB). The P fertilizer source for all treatments was triple superphosphate (TSP: 46% soluble/total P₂O₅ and 10% Ca). The initial placements of P was carried out as broadcast incorporated to 20 and 40 cm in the soil by subsoiler and disk plowing (plowing at 20 or 40 cm depth) and the annual rates of P in band placement were carried out applied 5 cm deep and 5 cm to the side (5 by 5) of the seed with the planter for soybean and corn sowing in each year of cultivation.

The area was limed before initiating the study in 2008 with 4 Mg ha⁻¹ of hydrated calcium, which corresponds to 1116 and 312 kg ha⁻¹ of Ca and Mg [Ca(OH)₂ + Mg(OH)₂] incorporated in the soil by disk plowing (plowing at 20 cm depth), and 1 and 2 Mg ha⁻¹ in 2013 and 2014, which corresponds to 279 and 558 kg ha⁻¹ of Ca and 78 and 156 kg ha⁻¹ of Mg, respectively, applied superficial in the soil. Complementary fertilization in all soybean-growing years was 102 kg K₂O ha⁻¹ from potassium chloride. Already for the years in which the corn was cultivated, 45 kg N ha⁻¹ from urea and 60 kg K₂O ha⁻¹ from potassium chloride after the planting and 45 kg N ha⁻¹ from urea at the V4/V5 stage of development. The soybean establishment was always in October and the harvest in January and the corn establishment was always in January and the harvest in July.

4.2.3. Soil sampling, analysis, and phosphorus fractionation

After the last harvest in August of 2015 a composite soil sample of eight soil cores was collected from the 0-10, 10-20 and 20-40 cm depth from each plot. Sequential P

fractionation was following the methodology proposed by Hedley et al., (1982) with modifications by Condon et al. (1985). This technique uses chemical extractants sequentially in the same sample to remove progressively from the most available to the most stable inorganic (Pi) and organic (Po) P pools. At each step, 10 mL of extractant was added to 0.5 g soil in 15 mL centrifuge tubes (1:20 soil:solution ratio), and the tubes were shaken end-over-end (vertical shaker, 60 rpm) for 16 h at 25 °C. Soil samples were extracted sequentially using: (i) Anion exchange resin (AER) for inorganic P (P_{AER}) readily diffusing into solution using a resin membrane (2.0 cm² in area); (ii) 0.5 M NaHCO₃ to extract inorganic P (P_{Bic}) weakly adsorbed on the surface on crystalline compounds and organic P (P_{OBic}) compounds with low recalcitrance like ribonucleic acid and glycerophosphate (Tiessen and Moir, 1993); (iii) 0.1 M NaOH to extract inorganic P ($P_{i0.1Hid}$) strongly adsorbed onto Fe and Al and clay minerals (Hedley et al., 1982), and organic P ($P_{o0.1Hid}$) mainly associated with fulvic and humic acids adsorbed onto mineral and SOM surfaces (Linquist et al., 1997); (iv) 1 M HCl to extract inorganic P (P_{HCl}) associated with apatite, other sparingly-soluble Ca-P compounds or negatively charged oxides surfaces (Gatiboni et al., 2007); (v) 0.5 M NaOH to extract inorganic P ($P_{i0.5Hid}$) associated with Fe and Al and clay minerals, and organic P ($P_{o0.5Hid}$) associated with fulvic and humic acids inside aggregates (Condon et al., 1985). After each extraction, the soil suspensions were centrifuged at 3278 g for 30 min to collect clear supernatants. After centrifugation and removal of the supernatant at each step of the fractionation, the soil was washed with 10 mL 0.5 M NaCl solution, to prevent the residual effect of the previous solution (pH change). The (vi) residual P ($P_{Residual}$) after drying the remaining soil at 40°C for 3 days, was determined by H₂SO₄ + H₂O₂ + MgCl₂ digestion (Brookes and Powelson, 1981), this fraction consists of occluded P forms (Cross and Schelinger, 1995).

The alkaline extracts were added to a 1:1 sulfuric acid solution and ammonium persulfate (7%) in an autoclave at 121 °C and 103 kPa for 2 h to determine the Pt (Pi + Po) (USEPA, 1971). The amount of Pi in the alkaline extracts (P_{Bic} , $P_{i0.1Hid}$ and $P_{i0.5Hid}$) was determined using the colorimetric method of Dick and Tabatabai (1977). Inorganic P in each of the acid extract (P_{AER} , P_{HCl} , $P_{Residual}$ and digestion of P_{tBic} , $P_{t0.1Hid}$ and $P_{t0.5Hid}$) were determined following the colorimetric method of Murphy and Riley (1962). Organic P in each alkaline extract was determined by the difference between Pt and Pi (P_{OBic} , $P_{o0.1Hid}$, and $P_{o0.5Hid}$).

In the end, the pools of P were grouped based on the lability predicted by the extractors (Cross and Schlesinger, 1995). Labile P consisted of the P_{AER} plus the P_{Bic} (Pi and

Po), moderately labile P consisted of the $P_{0.1\text{Hid}}$ (P_i and P_o) plus the P_{HCl} , and non-labile P consisted of the $P_{0.5\text{Hid}}$ (P_i and P_o) plus the P_{Residual} .

4.2.4. Statistical analysis

All statistical analyses were completed in SAS Studio (version 9.3; SAS, Cary, NC). The GLIMMIX procedure was used for ANOVA analysis. Initial P fertilizer placement and annual P fertilizer rates were considered as a fixed factor in the model, and blocks were considered as a random factor (Littell et al., 2006). Corrected denominator degrees of freedom were obtained using the Kenward-Roger adjustment. Mean comparisons were done using the LSMEANS and SLICE option in PROC GLIMMIX. Statistical differences were established at the 0.05 probability level.

4.3. RESULTS

4.3.1. Labile P fractions (P_{AER} , P_{Bic} and P_{OBic})

Overall, after five years, the labile P pool (P_{AER} , P_{Bic} and P_{OBic}) was significantly affected by the initial P fertilizer treatments and annual rates and placement in all soil layers (Table 2). However, just in the topsoil (0-10 cm depth) the labile P pool was significantly affected by the interaction of initial P fertilizer treatments \times annual rates and placements (Table 2) and presents them from 5 to 19% of the total P in the soil. In this layer (0-10 cm), the 100B under I20 presented the highest concentration of labile P (Figure 2 - A1). Yet, 100B was not different significantly to 100SB but presented a higher level of P than 0A, 50B, and 50SB under all other initial P fertilizer treatments. Under 50B and 100SB annual rates and placements, I20 and I40, and S and I40 initial P fertilizer treatments, present the higher level of P compared to 0. However, under 100B the S and I20 presented the higher values of P.

Hence, the P_{AER} fraction was the most dominant fraction of labile P at all soil depth layer. The P_{AER} levels in the 0-10 cm were affected by the annual rates and placement treatments (Table 3) with the highest concentrations for 100B and 100SB (Table 4). However, the P_{Bic} and P_{OBic} were affected by the interaction of initial P fertilizer treatments \times annual rates and placements (Table 2). For the P_{Bic} , 100B under S and I20 presented the higher concentration of P. However, under 0 and I40 did not show any significant differences to 100SB (Table 4). Under annual 100B, the S and I20 presented higher concentrations of P, otherwise S was higher than 0 and I40 under 100SB (Table 4). For P_{OBic} , under I40 the annual

100SB maintained the highest level. Under 0, S and I20, the annual 100B was higher than 0A and 50B, 50SB, respectively (Table 4). Under 0A, the initial S was higher than 0 and I20. Under 50B and 100B, the 0 and I40 present the lower concentrations of $P_{O_{Bic}}$ (Table 4).

In the soil layer 10-20 cm the labile P pool was also affected by annual rates and placements (Table 3), representing concentrations from 3 to 12% of the total P (Figure 2 – A2). The highest P concentration was observed at 100B, mostly represented by P_{AER} and $P_{i_{Bic}}$ levels that were affected by annual rates and placements treatments as well (Table 3). Otherwise, the $P_{O_{Bic}}$ level was affected by the interaction of initial P fertilizer treatments \times annual rates and placements (Table 2). Under 0 and I20, the annual 100SB presented the lowest $P_{O_{Bic}}$ concentration. However, under S treatment presented the higher concentration. Also, under I20, the 0A presented the lowest P concentration. Under annual 100B and 100SB, the I40 and S presented the higher concentration of $P_{O_{Bic}}$.

The labile P pool in the bottom soil layer (20-40 cm) was similarly affected by initial P fertilizer treatments and annual rates and placements (Table 3), meaning 2 to 10% of total P in the soil. Lower levels of labile P were observed for 0 and the highest level was observed for 100B (Figure 2 - A3). In the same way to the other layers, P_{AER} was affected by the interaction of initial P fertilizer treatments \times annual rates and placements (Table 2), with the highest concentration observed at 100B under evenly all initial P fertilizer treatments (Table 4). However, under initial 0 the 100B treatment presented the lowest concentration of P and under 40I and 50SB presented the highest concentration of P. In this layer, $P_{i_{Bic}}$ was also affected by initial P fertilizer treatments and annual rates and placements (Table 2), with lower concentration observed at initial 0 and the highest concentration observed at annual 100B (Table 4). However, the $P_{O_{Bic}}$ was not affected by both initial P fertilizer treatments and annual rates and placements (Table 2).

Table 2. Statistical significance of P initial fertilizer treatment and annual rates in three soil sampling depths on P lability fractions. Soil samples collected at the 0-10, 10-20 and 20-40 cm depths, for four initial P fertilizer treatments (surface-S, incorporated to 20 cm-I20, incorporated to 40 cm-I40 and control-0) and five annual rates and placement treatments (50-Band, 100-Band, 50-Broadcast,100-Broadcast, and control). Samples collected after five years of double-crop soybean and corn.

Fixed effect	Hedley P fractions			
	Labile P	Moderately labile P	Non-labile P	Total P
	----- p > F -----			
	<u>0 – 10 cm</u>			
Initial treatments (IT)	0.008	0.146	0.001	0.004
Annual rates and placements (ARP)	<0.001	<0.001	<0.001	<0.001
IT × ARP	0.015	0.266	0.167	0.065
	<u>10 – 20 cm</u>			
Initial treatments (IT)	0.281	0.054	0.685	0.865
Annual rates and placements (ARP)	<0.001	<0.001	0.093	<0.001
IT × ARP	0.713	0.671	0.759	0.879
	<u>20 – 40 cm</u>			
Initial treatments (IT)	0.008	0.716	0.588	0.310
Annual rates and placements (ARP)	<0.001	<0.001	0.272	<0.001
IT × ARP	0.062	0.154	0.274	0.642

Significant different detected at $p < 0.05$.

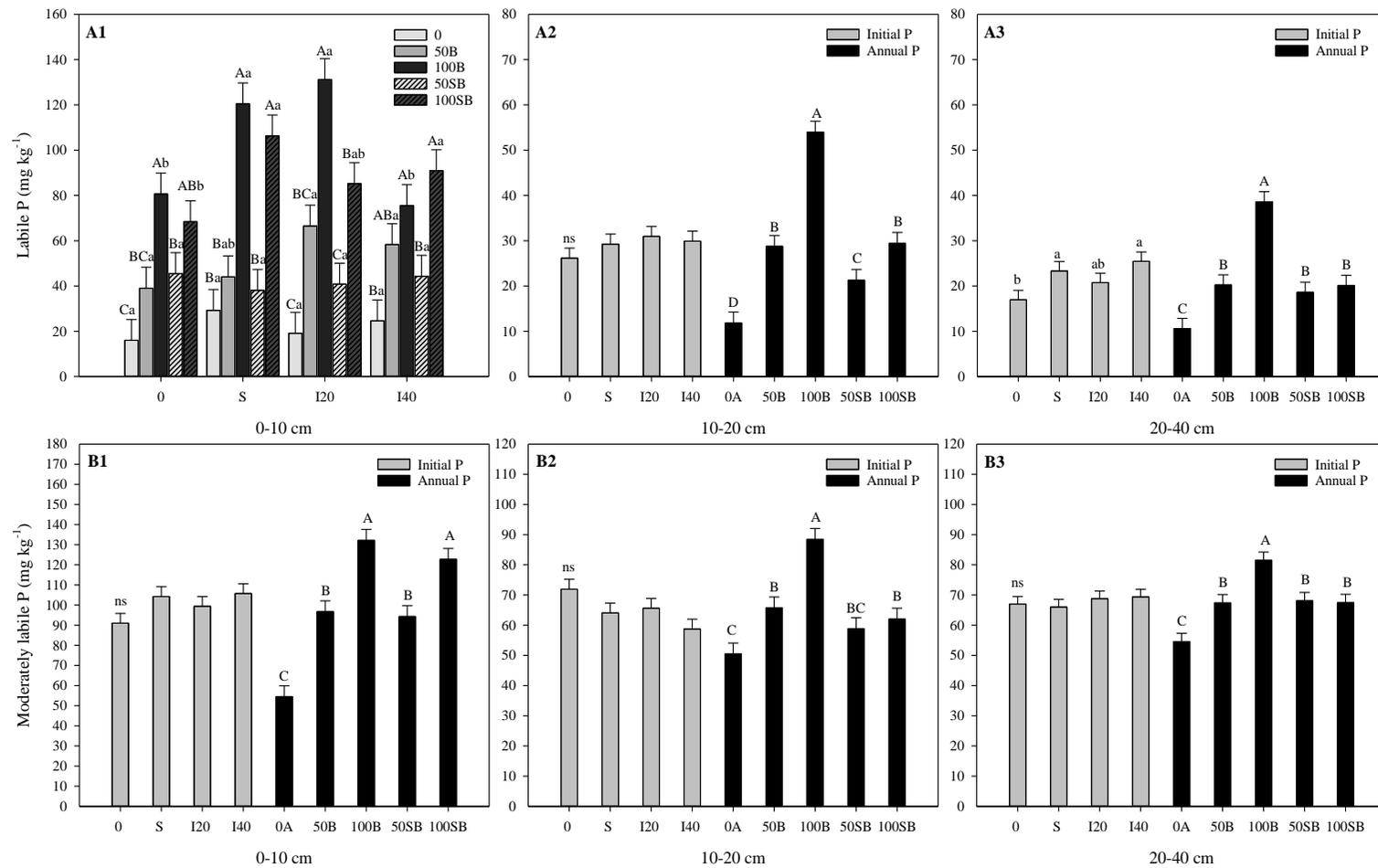


Figure 2. Soil P levels in labile (A) and moderately labile (B) pools for three soil sampling depths as affected by four initial P fertilizer treatments (surface-S, incorporated to 20 cm-I20, incorporated to 40 cm-I40, and control), and five annual rates and placement treatments (50-band, 100-band, 50-superficial broadcast, 100-superficial broadcast and control) after five years of double-crop soybean and corn in a no-till system. Within each depth and P fractions, means followed by the same uppercase letter (initial treatments) and lowercase letter (annual rates and placement) are not statistically different ($p > 0.05$). ns: not significant

Table 3. Statistical significance of P initial fertilizer treatment and annual rates in three soil sampling depth on P fractions. Soil samples collected at 0-10, 10-20 and 20-40 cm depths, for four initial P fertilizer treatments (surface-S, incorporated to 20 cm-I20, incorporated to 40 cm-I40 and control-0) and five annual rates and placement treatments (50-Band, 100-Band, 50-Broadcast,100-Broadcast, and control). Samples collected after five years of double-crop soybean and corn.

Fixed effect	Hedley P fractions								
	P _{AER}	P _{Bic}	P _{O_{Bic}}	P _{i0.1Hid}	P _{O0.1Hid}	P _{HCl}	P _{i0.5Hid}	P _{O0.5Hid}	P _{Residual}
	----- p > F -----								
	<u>0 - 10 cm</u>								
Initial treatments (IT)	0.288	0.005	0.001	0.154	0.419	0.042	0.154	0.419	0.003
Annual rates and placements (ARP)	<0.001	<0.001	0.033	0.151	<0.001	0.003	0.151	<0.001	0.006
IT × ARP	0.069	0.004	0.043	0.993	0.022	0.323	0.993	0.021	0.196
	<u>10 - 20 cm</u>								
Initial treatments (IT)	0.262	0.239	0.281	0.084	0.050	0.383	0.084	0.050	0.301
Annual rates and placements (ARP)	<0.001	<0.001	<0.001	0.747	0.002	<0.001	0.747	0.002	0.116
IT × ARP	0.871	0.192	0.004	0.553	0.452	0.928	0.553	0.452	0.586
	<u>20 - 40 cm</u>								
Initial treatments (IT)	0.001	0.005	0.123	0.542	0.208	0.769	0.542	0.208	0.227
Annual rates and placements (ARP)	<0.001	<0.001	0.702	0.950	0.257	0.001	0.950	0.257	0.094
IT × ARP	0.026	0.229	0.367	0.930	0.196	0.553	0.930	0.195	0.260

P_{AER}, Inorganic P extracted by anion exchange resin; P_{Bic}, Inorganic P extracted by 0.5 M NaHCO₃; P_{O_{Bic}}, Organic P extracted by 0.5 M NaHCO₃; P_{i0.1Hid}, Inorganic P extracted by 0.1 M NaOH; P_{O0.1Hid}, Organic P extracted by 0.1 M NaOH; P_{HCl}, Inorganic P extracted by 1 M HCl, P_{i0.5Hid}, Inorganic P extracted by 0.5 M NaOH; P_{O0.5Hid}, Organic P extracted by 0.5 M NaOH; P_{Residual}, Residual P.

Table 4. Labile P fractions (P_{AER} , P_{iBic} and P_{oBic}) at three soil sampling depths (0-10, 10-20 and 20-40 cm) for four initial P fertilizer treatments (surface-S, incorporated to 20 cm-I20, incorporated to 40 cm-I40 and control-0), and five annual rates and placement treatments (50-band, 100-band, 50- superficial broadcast, 100- superficial broadcast and control). Samples collected after five years of double-crop soybean and corn.

Hadley phosphorus fractions / annual rates and placements / depth																				
		P_{AER} (mg kg⁻¹)					P_{iBic} (mg kg⁻¹)						P_{oBic} (mg kg⁻¹)							
		0-10 cm																		
IT	0A	50B	100B	50SB	100SB	Mean	0A	50B	100B	50SB	100SB	Mean	0A	50B	100B	50SB	100SB	Mean		
0	5.5	19	43	27	41	27ns	4.4Ba	13Aba	23Ab	11Ba	20Ab	14	6.2Bc	6.9Bb	14Ab	7.7ABa	7.8ABb	8.6		
S	7.5	20	58	19	62	33	5.2Ca	12Ca	46Aa	11Ca	30Ba	21	17Aa	12ABab	17Aab	9Ba	14ABab	14		
I20	5.8	30	66	18	47	33	4.2Ca	18Ba	44Aa	8.4Ca	23Bab	20	9.1Bbc	19Aa	22Aa	15ABa	15ABab	16		
I40	5.9	29	38	25	51	30	3.8Ca	16Ba	27Ab	10BCa	21ABb	16	15ABab	13Bab	10Bb	9.4Ba	19Aa	13		
Mean	6.2C	25B	51A	22B	50A		4.4	15	35	10	24		12	13	16	10	14			
		10-20 cm																		
0	3.2	11	25	7.6	13	12ns	1.9	6.2	13	3.8	10	7.1ns	6.7ABa	8.2Aa	8.3Ab	6.4ABa	5.4Bb	7.0		
S	3.7	16	27	8.1	13	14	1.7	10	19	4.1	8.0	8.5	6.2Ba	6.7Ba	7.8ABb	6.1Ba	9.5Aa	7.2		
I20	4.2	13	29	14	14	15	2.7	7.8	18.5	6.1	7.1	8.4	6.4ABa	8.4ABa	9.2Ab	7.9ABa	5.7Bb	7.5		
I40	3.3	14	31	9.1	16	15	1.8	5.6	15	5.5	8.4	7.2	5.6BCa	8.2Ba	14Aa	6.2BCa	6.8BCab	8.2		
Mean	4D	13B	28A	10C	14B		2.0D	7.4B	16A	4.9C	8.5B		6.2	7.9	9.8	6.6	6.9			
		20-40 cm																		
0	2.4Ba	4.5ABa	9.2Ac	4.7ABb	8.0ABa	5.8	4.5	5.8	9.1	5.0	6.1	6.0c	4.8	5.8	4.6	4.3	6.3	5.2ns		
S	2.4Ca	8.2Ba	25Aa	8.9Bab	6.6BCa	10	3.7	6.8	20	7.2	8.3	9.2ab	4.2	4.7	4.2	4.4	2.3	3.9		
20I	1.7Ca	9.3Ba	18Ab	4.3BCb	7.2BCa	8.1	3.3	8.0	15	4.5	6.3	7.3bc	5.9	4.7	5.7	5.9	4.3	5.3		
40I	2.4Ca	9.4Ba	21Aab	10Ba	11Ba	11	4.4	9.0	17	8.9	9.5	9.7a	3.1	4.9	6.4	5.8	4.9	5.0		
Mean	2.2	7.9	18	7.1	8.1		3.9C	7.4B	15A	6.4B	7.5B		4.5ns	5.0	5.2	5.1	4.7			

Within each depth and P fraction, means followed by the same capital letter in rows and tiny letter in columns are not statistically differently ($p > 0.05$). ns: not significant; IT, initial treatments Annual rates; 50B, 50 band; 100B, 100 band; 50SB, 50 superficial broadcast; 100SB, 100 superficial broadcast.

4.3.2. Moderately labile P fractions ($P_{i0.1Hid}$, $P_{o0.1Hid}$ and P_{HCl})

The moderately labile P pool was affected by annual rates and placement treatments at the 0-10 cm soil layer (Table 2), representing from 13 to 23 % of the total P in the soil with the highest level of P observed at 100A and 100SB (Figure 2-B1). Although the $P_{i0.1Hid}$ was the most dominant fraction of moderately labile P at all soil depth layers it was not affected by any treatment (Table 3). Otherwise, $P_{o0.1Hid}$ was affected by the interaction of initial P fertilizer treatments \times annual rates and placements (Table 3). The annual 0A under 0, S and I20 and 0A and 50SB under I40, were observed the lowest concentration of P for this fraction (Table 5). Under 0A, the I40 was higher than S and I20. However, under 50SB, the I40 was lower than 0 and S. Also, under 100B the highest concentration was observed for I20. In addition, P_{HCl} was affected by both initial P fertilizer treatments and annual rates and P placements (Table 2), with a concentration of P for the initial S higher than 0 and I40 and annual 100B higher than 0, 50SB and 100SB (Table 5).

In the deep layers of 10-20 and 20-40 the moderately labile P pool was affected only by annual rates and placements (Table 2), and presented from 12 to 19 and 12 to 18 % of the total P in each layer respectively, with the highest level of P observed at 100B treatment (Figure 2-B2 and B3). Yet, the $P_{i0.1Hid}$ for both soil layers was not affected for both initial P fertilizer treatments and annual rates and placements, being not discussed here. Otherwise, $P_{o0.1Hid}$ was affected by annual rates and placement treatments with the highest concentration observed at 100B (Table 3 and 5). The P_{HCl} for both soil layers was affected by annual rates and placements (Table 2) with the highest concentration of P observed at 100B (Table 5).

Table 5. Moderately labile P fractions ($P_{i0.1Hid}$, $P_{o0.1Hid}$ and P_{HCl}) at three soil sampling depths (0-10, 10-20 and 20-40 cm) for four initial P fertilizer treatments (surface-S, incorporated to 20 cm-I20, incorporated to 40 cm-I40 and control-0), and five annual rates and placement treatments (50-band, 100-band, 50- superficial broadcast, 100- superficial broadcast and control). Samples collected after five years of double-crop soybean and corn.

Hadley phosphorus fractions / annual rates and placements / depth																			
		$P_{i0.1Hid}$ (mg kg⁻¹)					$P_{o0.1Hid}$ (mg kg⁻¹)					P_{HCl} (mg kg⁻¹)							
		0-10 cm																	
IT		0A	50B	100B	50SB	100SB	Mean	0A	50B	100B	50SB	100SB	Mean	0A	50B	100B	50SB	100SB	Mean
0		40	42	45	45	43	43ns	20Cab	35ABa	49Ab	33Ba	33Ba	34	1.1	2.7	5.3	1.8	2.1	2.6b
S		44	51	55	45	52	49	6.2Cb	48Aa	52Ab	27Ba	41Aa	35	1.7	11	10	2.1	4.4	5.9a
I20		42	46	48	39	47	45	10Cb	43Ba	68Aa	20Cab	33BCa	35	1.3	5.1	13	2.0	3.7	5.0ab
I40		42	49	50	46	55	48	24Ba	38Aa	44Ab	16Bb	28ABa	30	1.4	3.5	3.6	1.9	3.5	2.8b
Mean		42ns	47	49	44	49		15	41	53	24	34		1.4C	5.6AB	8.0A	1.9C	3.4BC	
		10-20 cm																	
0		41	39	47	50	49	45ns	10	13	26	8.0	15	15ns	1.1	1.4	2.3	1.0	1.5	1.5ns
S		44	49	46	36	44	44	5.5	7.3	18	8.0	8.7	9.5	1.1	1.3	2.2	0.9	1.1	1.3
I20		37	39	43	33	36	38	11	19	24	21	11	17	0.9	1.7	2.8	1.6	1.2	1.6
I40		42	42	43	44	36	41	3.6	16	21	6.6	20	13	0.8	1.5	2.4	1.1	1.4	1.4
Mean		41ns	42	45	41	41		7.5C	14B	22A	11BC	14B		1.0C	1.5B	2.4A	1.2BC	1.3BC	
		20-40 cm																	
0		35	43	42	42	38	40ns	12	4.1	7.8	6.9	11	8.1ns	0.2	0.2	0.4	0.1	0.2	0.2ns
S		43	38	38	40	40	40	5.0	5.7	17	5.0	11	8.7	0.1	0.1	0.8	0.1	0.2	0.3
I20		43	44	41	43	43	43	4.2	4.5	3.8	8.0	4.3	5.0	0.1	0.2	0.3	0.1	0.1	0.2
I40		41	43	42	42	38	40	7.8	8.7	11	2.9	6.2	7.2	0.1	0.3	0.3	0.1	0.1	0.2
Mean		41ns	42	40	41	41		7.2ns	5.7	9.9	5.7	7.9		0.1B	0.2B	0.5A	0.2B	0.2B	

Within each depth and P fraction, means followed by the same capital letter in rows and tiny letter in columns are not statistically differently ($p > 0.05$).

ns: not significant; IT, initial treatments Annual rates; 50B, 50 band; 100B, 100 band; 50SB, 50 superficial broadcast; 100SB, 100 superficial broadcast.

4.3.3. Non-labile P fractions ($P_{i0.5Hid}$, $P_{o0.5Hid}$ and $P_{Residual}$)

Overall, the non-labile P increased in the soil surface and was affected by both initial P fertilizer treatments and annual rates and P placements (Table 2). This P pool represented from 60 to 85 % of the total P in the soil with lower levels observed at initial 0 and annual 0A (Figure 3 – A1). The $P_{i0.5Hid}$ was not affected by any treatment at all soil layers, not discussed here. However, the $P_{o0.5Hid}$ was affected by the interaction of initial P fertilizer treatments \times annual rates and placements in the soil surface (0-10 cm) and by annual P fertilizer alone in the 10-20 cm soil layer (Table 3). In the 0-10 soil layer, the lowest concentration of $P_{o0.5Hid}$ was observed at 0A under all initial P fertilizer treatments. Under 0A, 50B and 50SB, the lowest concentration of non-labile P were observed at S, 0, and I40, respectively. Yet, the highest concentration was observed at I20 under 100B (Table 6). In addition, in the 10-20 cm the highest concentration of P was observed at 100B. The $P_{Residual}$ was affected by both initial P fertilizer treatments and annual rates and P placements but just at 0-10 cm (Table 3), with the lowest concentration observed at initial 0 and 0A (Table 6).

In general, after five years, the total P was affected by both initial P fertilizer treatments and annual rates and placements in the soil surface (0-10 cm) and by annual rates and placements in the 10-20 and 20-40 cm soil layers (Table 2). At 0-10 cm the lower level of total P was observed at 0 initial P fertilizer, and for annual rates and placements the highest concentration was observed at 100B and 100SB (Figure 3 – B1). However, in both deeper soil layers (10-20 and 20-40 cm) the highest total P was observed at 100B (Figure 3- B2 and B3).

Table 6. Non-labile P fractions ($P_{i0.5Hid}$, $P_{o0.5Hid}$ and $P_{Residual}$) at three soil sampling depths (0-10, 10-20 and 20-40 cm) for four initial P fertilizer treatments (surface-S, incorporated to 20 cm-I20, incorporated to 40 cm-I40 and control-0), and five annual rates and placement treatments (50-band, 100-band, 50- superficial broadcast, 100- superficial broadcast and control). Samples collected after five years of double-crop soybean and corn.

Hadley phosphorus fractions / annual rates and placements / depth																			
		$P_{i0.5Hid}$ (mg kg⁻¹)					$P_{o0.5Hid}$ (mg kg⁻¹)					$P_{Residual}$ (mg kg⁻¹)							
		0-10 cm																	
IT		0A	50B	100B	50SB	100SB	Mean	0A	50B	100B	50SB	100SB	Mean	0A	50B	100B	50SB	100SB	Mean
0		40	42	45	45	43	43ns	20Ca	35ABb	49Ab	33Ba	33Ba	34	209	281	296	301	298	277b
S		44	51	55	45	52	49	6.2Cb	48Aa	52Ab	27Ba	41ABa	35	296	304	301	304	296	301a
I20		42	46	48	39	47	45	9.6Cab	43Ba	68Aa	20Cab	33Ba	35	295	328	303	306	332	312a
I40		42	49	50	46	55	48	24Ba	38Aab	44Ab	16Cb	28ABa	30	291	319	311	299	322	308a
Mean		42ns	47	49	44	49		15	41	53	24	34		272B	308A	303A	303A	313A	
		10-20 cm																	
0		41	39	47	50	49	45ns	10	13	26	13	15	15ns	273	263	276	281	277	274ns
S		44	49	46	36	44	44	5.5	7.3	18	8.0	8.7	9.4	267	285	279	358	281	294
I20		37	39	43	33	36	38	11	19	24	21	11	17	286	293	298	292	284	290
I40		42	42	43	44	36	41	4.6	16	21	6.6	20	13	283	290	294	306	283	291
Mean		41ns	42	45	41	41		7.5C	14B	22A	11BC	13B		277ns	283	287	309	281	
		20-40 cm																	
0		35	43	42	42	38	40ns	12	4.1	7.8	6.9	11	8.3ns	293	298	290	293	292	291ns
S		43	38	38	40	40	40	5.0	5.7	17	5.0	11	8.7	283	279	292	291	294	288
I20		43	44	41	42	43	43	4.2	4.5	3.8	8.0	4.3	5.0	279	287	298	304	284	290
I40		41	42	38	39	41	40	7.8	8.7	11	2.9	6.2	7.2	293	298	298	293	293	295
Mean		41ns	42	40	41	40		7.2ns	5.7	9.9	5.7	8.0		287ns	288	294	295	291	

Within each depth and P fraction, means followed by the same capital letter in rows and tiny letter in columns are not statistically differently ($p > 0.05$). ns: not significant; IT, initial treatments Annual rates; 50B, 50 band; 100B, 100 band; 50SB, 50 superficial broadcast; 100SB, 100 superficial broadcast.

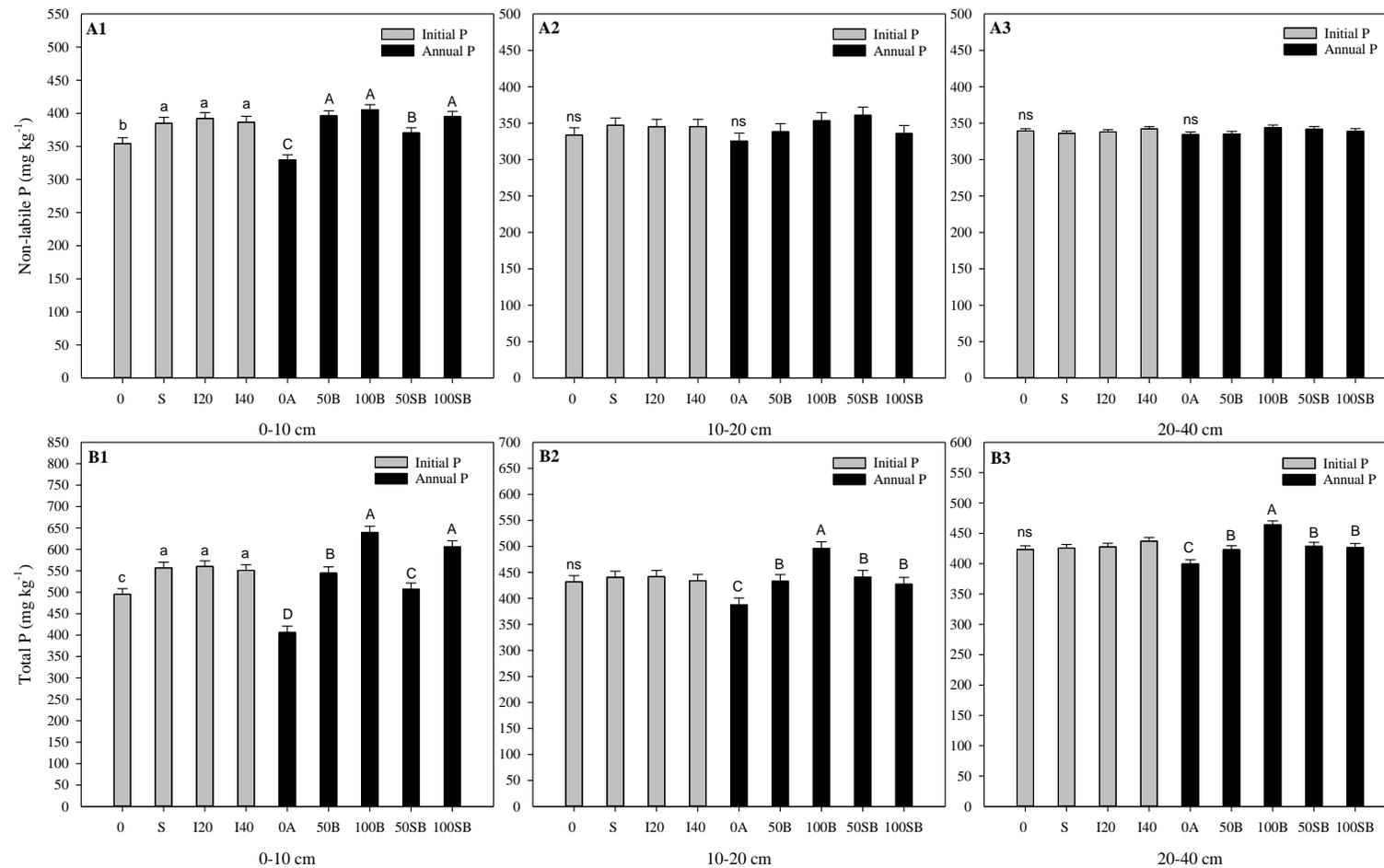


Figure 3. Soil non-labile P (A) and total P (B) for three soil sampling depths as affected by for four initial P fertilizer treatments (surface-S, incorporated to 20 cm-I20, incorporated to 40 cm-I40 and control-0), and five annual rates and placement treatments (50-band, 100-band, 50-surficial broadcast, 100-surficial broadcast and control) after five years of double-crop soybean and corn in a no-till system. Within each depth and P fractions, means followed by the same uppercase letter (annual treatments) and lowercase letter (annual rates and placements) are not statistically different ($p < 0.05$). ns: not significant

4.4. DISCUSSION

4.4.1. Labile P fractions (P_{AER} , P_{iBic} and P_{oBic})

Five years after the transition from the extensive pasture to no-till system under P fertilizer management (initial placements and annual rates and placements) have promoted significant changes in the levels of the labile P pool. Our study showed higher level of labile P in the soil surface compared to other studies under no-till system (Rodrigues et al., 2016, Herrera et al., 2016, Teles et al., 2017; Soltangheisi et al., 2018), however, showed the same behavior with decrease of labile P in deeper layers. Yet, the highest amount of labile P (P_{AER} , P_{iBic} and P_{oBic}) means high levels of available P in the surface soil (0-10 cm) and low for the subsoil (10-20 and 20-40cm).

The highest level (~19%) was found for the annual 100B under initial I20, what was attributed to the specific localization of the higher rate plus the incorporation of P fertilizer to the 20 cm in the soil. In Oxisols, with high amount of P fixing components, when P fertilizer is banding in the plant row with minimum disturbance of the soil as promoted by NT, it can contribute to reduce the high P sorption reactions, and that may have contributed to increasing the labile P levels in the soil surface, what is supported by literature (Rheinheimer and Anghinoni, 2001, Rheinheimer et al., 2003). In addition, the incorporation of the initial P fertilizer to 20 cm could partially saturate the soil adsorption sites, and thus P accumulation from annual band rate have improved the levels of P labile. As decreasing P adsorption, more P from the fertilizer can remain in the labile form (Rodrigues et al., 2016).

Moreover, the presence of low molecular weight compounds from organic matter near the surface from the crop residues might block P adsorption sites increasing the P availability (Hue, 1991; Rheinheimer and Anghinoni, 2001; Guppy et al., 2005), what partially explains the no differences between the annual 100B and SB100 under 0, S and I40 initial P fertilization placement. Also, the differences found under annual fertilization 50B and 100SB compared to the 0A is due to the addition of initial P fertilizer that remains in the soil after five years. Still, the initial P fertilization, S and I20, contributed to the highest amount of labile P for 100B since the banding application is about 5-10 cm of the soil profile.

The highest levels of labile P pool (10-12%) founded for the 100B compared to the all other annual P additions at 10-20 and 20-40 soil depths, may be attributed to the continuous banding application which providing less contact between soil and the fertilizer and

consequently enhanced labile P pool. Moreover, the initial P fertilizer application also contributed to enhance labile P compared to the control (0) in deeper soil layer (20-40 cm).

The P_{AER} for 100B and 100SB annual P application contributed to more than 50% of readily-available P than 50B and 50SB and almost 90% more than 0A (no P fertilization). The no differences between the rate 100 as band or broadcast means that P dynamics was similar each other. However, higher concentration was observed for the 100B in 10-20 cm. In addition, in deeper soil layer under all initial P fertilizer managements the amount of P was higher under 100B due deeper applications.

For the P_{iBic} , the highest concentration was founded at 100B under S and I20 with the same behavior for the same treatments found in the labile P pool in the soil surface as well the no differences between the 100B and 100SB. In addition, in the 10-20 and 20-40 cm soil depth the P_{iBic} presented similar behavior founded for the P_{AER} in the 10-20 cm soil depth. However, the higher amount of P in this fraction was found at initial P application I40, attributed to the remaining available P from initial fertilizer application after 5 years. In general, the amount of P_{oBic} was higher in the soil surface than deeper layers. Obviously, NT system has contributed to the maintenance of crop residue on the soil surface and consequently increasing the amount of SOM and Po (Gatiboni et al., 2007). The highest level of P_{oBic} in the soil surface and 10-20 cm is due more to the partial immobilization of P from fertilizer for both initial and annual fertilizer application.

4.4.2. Moderately labile P fractions ($P_{i0.1Hid}$, $P_{o0.1Hid}$ and P_{HCl})

The higher rates of annual P fertilizer for all soil layers (0-10, 10-20 and 20-40) increased the levels of moderately labile P pool with no differences between 100 band and broadcast. In addition, the amount of $P_{o0.1Hid}$ under 0A annual rates increased with the incorporation of initial fertilizer for correction of the soil profile and still remains in the soil later. However under 50SB it was observed the opposite due the cumulative P fertilizer application. Also, the higher concentration of $P_{o0.1Hid}$ founded for 100B under I20 in the soil surface and 100B in the 10-20 cm soil depth have an explanation similar to that found for the labile P pool for which soil depth. In addition, for the $P_{o0.1Hid}$ in the 10-20 soil depth the continuous localization plus the higher rate of the P fertilizer increased the amount of this fraction in the soil. Thus, the organic P in this fraction can be drain with continuous high P fertilizer application, what is also supported by literature (Conte et al., 2003).

The differences of levels founded to the P_{HCl} for all soils layers is a consequence of the localization of calcium added to soil, since the source of P fertilizer used have 10% of Ca and also was applied lime in three occasions during the experiment. This fraction was a very small amount of P in the moderately labile P pool in this strongly weathered soil (Pavinato et al., 2009, Rodrigues et al., 2016) since this fraction is linked to apatite, other moderately soluble Ca-P compounds or negatively charged oxide surfaces (Hedley et al., 1982), what is very low in this soil.

4.4.3. Non-labile P fractions ($P_{i0.5Hid}$, $P_{o0.5Hid}$ and $P_{Residual}$)

This non-labile P represents the largest pool of the total P due to strong soil P-buffering capacity of well weathered soils like this Brazilian Oxisol, which more significantly decrease available P over the time under soluble sources in long-term cumulative fertilizer application. This behavior is due to the high MPAC value (826 mg kg^{-1}), and lower amorphous/crystalline Fe ratio (FeOxalate/FeDCB,) 0.05 (0.041), which indicates that in this soil the crystalline oxides are predominant (~71% of clay in the soil profile) and confirmed by the types of mineral predominant in X-ray diffraction (Table 1 and Fig. 1). The higher P adsorption capacity and consequently reduction of labile and moderately labile P pools and poor mobility of P, is a consequence of the soil constitution, predominantly of kaolinite, gibbsite, goethite and hematite.

Lower non-labile P founded for the annual control (0A) under no initial fertilizer application in the soil surface and this has an explanation similar to that found for the $P_{o0.1Hid}$ in the moderately labile fraction. In addition, the highest concentration of organic moderately labile P for I20 under 100B in the soil surface and 100B in the 10-20 cm soil depth have an explanation similar to that found for labile P fraction in the same soil layers. Thus, the lower concentrations for residual with no initial and annual P fertilizer application and the higher concentrations with both initial and annual P fertilization emphasizing the higher capability to accumulate P in more recalcitrant fractions of this soil due its great MPAC.

The interactions of initial and annual treatments in the labile P at the soil surface and the effects of annual fertilization applications for 10-20 and 20-40 soil layer may had affected the highest concentration for the total P presents similar behavior for 100B and 100SB and 100B for soil surface and 10-20 and 20-40 cm soil depths, respectively. Therefore, inorganic P mainly controlled P availability when phosphate fertilizer is annually applied in the soil, which makes up a larger proportion of total P.

4.5. CONCLUSIONS

In tropical soils, in which Al and Fe oxide-hydroxide and 1:1 clay dominate the clay fraction, initial and annual phosphate placements under no-till system resulted in a large amount of legacy P accumulated in the soil, with more expressive increases in labile and moderately labile P in soil surface. Otherwise, annual P addition had more effect on P dynamics in the soil subsurface.

In general, the initial phosphate placements increased the labile, non-labile and total P pools in soil surface. In addition, annual rates and placements affected all P fractions in all layers of the soil profile analyzed. Hence, under initial P fertilization incorporated to 20 cm (I20) banding 100 kg P₂O₅ ha⁻¹ (100B) annually increased more expressively the labile P fraction in the soil surface, although under the other initial placements the 100B had the same effect that 100SB in moderately labile. Yet, the 100B annual placement increased the labile and moderately labile P pools in deeper soil (10-20 and 20-40 cm), non-labile in the soil surface and total P in all soil layers.

Thus, P accumulation from the initial (P remains in the soil after five years) and annual P fertilizer application in the labile P pools could help, at list in part, to reduce high current dependency and large inputs of inorganic P fertilizer in tropical no-till systems.

REFERENCES

- Brookes, P.C., and D.S. Powlson. 1981. Preventing phosphorus losses during perchloric acid digestion of sodium bicarbonate soil extracts. *J. Sci. Food Agric.* 32:671–674.
- Ciampitti, I.A., Picore, L.I., Rubio, G., Garcia, F.O., 2011. Pathways of Phosphorous Fraction Dynamics in Field Crop Rotations of the Pampas of Argentina. *Soil Sci. Soc. Am. J.* 75, 918–926.
- Condon, L.M., Spears, B.M., Haygarth, P.M.B., Turner, L., Richarddson, A.E., 2013. Role of legacy phosphorus in improving global phosphorus-use efficiency. *Environ. Dev.* 8, 147-148.
- Condon, L.M., Goh, K.M., Newman, R.H., 1985. Nature and distribution of soil phosphorus as revealed by a sequential extraction method followed by ³¹P nuclear magnetic resonance analysis. *J. Soil Sci.* 36, 199-207.
- Cordell, D., White, S., 2013. Sustainable phosphorus measures: strategies and technologies for achieving phosphorus security. *Agronomy.* 3, 86-116.

- Conte, E., Anghinoni, I. Rheinheimer, D.D.S., 2003. Phosphorus accumulation fractions in a clayey oxisol in relation to phosphate doses under no-tillage. *Rev. Bras. Cienc. Solo* 27, 893–900.
- Cross, A.F., Schlesinger, W.H., 1995. A literature review and evaluation of the Hedley fractionation: applications to the biogeochemical cycle of soil phosphorus in natural ecosystems. *Geoderma*, 64, 197–214.
- De Oliveira, O.C., De Oliveira, I.P., Alves, B.J.R., Urquiaga, S., 2004 Boddey, R.M. Chemical and biological indicators of decline/degradation of *Brachiaria* pastures in the Brazilian Cerrado. *Agric. Ecosyst. Environ.* 103, 289-300.
- Dick, W.A., Tabatabai, M.A., 1977. Determination of orthophosphate in aqueous solutions containing labile organic and inorganic phosphorus. *J. Environ. Qual.* 6, 82-85.
- Dhillon, J., Torres, G., Driver, E., Figueiredo, B., Raun, W.R., 2017. World Phosphorus Use Efficiency in Cereal Crops. *Agron. J.* 109, 1670-1677.
- EMBRAPA, 2013. Sistema brasileiro de classificação de solos, Terceira ed. Brasília, Distrito Federal.
- EMBRAPA, 1997. Manual de métodos de análise de solo, segunda ed. Rio de Janeiro, Rio de Janeiro.
- EMBRAPA, 1999. Brazilian Agricultural Research Corporation. <http://www.embrapa.br/en/busca-de-publicacoes/-/publicacao/555307/area-e-distribuicao-especial-de-pastagens-cultivadas-no-cerrado-brasileiro/> (accessed 26 November 2018).
- Fontes, M.P.F., Weed, S.B., 1996. Phosphate adsorption by clays from Brazilian Oxisols: relationships with specific surface area and mineralogy. *Geoderma.* 72, 37-51.
- Gatiboni, L.C., Kaminski, J., Rheinheimer, D.S., Flores, J.P.C., 2007. Biodisponibilidade de formas de fósforo acumuladas em solo sob Sistema de plantio direto (Bioavailability of soil phosphorus forms in no-tillage system). *Rev. Bras. Ciênc. Solo (Braz. J. Soil Sci.)* 31, 691-699.
- Guppy, C.N., N.W. Menzies, P.W. Moody, and F.P.C. Blamey. 2005. Competitive sorption reactions between phosphorus and organic matter in soil: a review. *Soil Research* 43: 189-202.
- Jackson, M.L., 1979. *Soil Chemical Analysis Advanced Course*, second ed. Madison, Wisconsin.

- Hedley, M.J., Steward, W.B., Chauhan, B.S., 1982. Changes in inorganic and organic soil phosphorus fraction induced by cultivation practices and laboratory incubation. *Soil Sci. Soc. Am. J.* 46, 970-976.
- Herrera, W.F.B., Rodrigues, M., Teles, A.P.B., Barth, G., Pavinato, P.S., 2016. Crop yield and soil phosphorus lability under soluble and humic-complexed phosphate fertilizers. *Agron. J.* 108, 1692-1702.
- Hue, N.V. 1991. Effects of organic acids/anions on P sorption and phytoavailability in soils with different mineralogies. *Soil Sci.* 152:463–471.
- Rheinheimer, D.S.; Anghinoni, I.; Conte, E.; Kaminski, J., Gatiboni, L.C., 2003. Dessorção de fósforo avaliada por extrações sucessivas em amostras de solo provenientes dos sistemas plantio direto e convencional. *Ci. Rural.* 33, 1053-1059.
- Rheinheimer, D.S., Anghinoni, I., 2001. Distribuição do fósforo inorgânico em sistemas de manejo de solo. *Pesq. Agropec. Bras.* 36, 151-160.
- Kämpf, N., Scheinost, A.C., Schulze, D.G., 2000. Oxide minerals, in: Sumner, M. E. (Eds.), *Handbook of soil science*. University of Georgia, Georgia, pp. 125-168.
- Linquist, B.A., Singleton, P.W., Cassaman, K.G., 1997. Inorganic and organic phosphorus dynamics during a build-up decline of available phosphorus in an ultisol. *Soil Sci.* 162, 254-264.
- Littell, R.C., G.A. Milliken, W.W. Stroup, R.D. Wolfinger, and O. Schabenberber. 2006. SAS for mixed models. 2nd ed. SAS Inst., Cary, NC.
- Lopes, A.S., Guimarães Guilherme, L.R., 2016. Chapter One - A career perspective on soil management in the Cerrado region of Brazil. *Adv. Agron.* 137, 1-72.
- Murphy, J., Riley, J.P., 1962. A modified single solution method for the determination of phosphate in natural waters. *Anal. Chim. Acta.* 27, 31 –36.
- Pavinato, P.S., Merlin, A., Rosolem, C.A., 2009. Phosphorus fractions in Brazilian Cerrado soils as affected by tillage. *Soil tillage Res.* 105, 149-155.
- Raij, B., Quaggio, J.A., Silva, N.M., 2008. Extraction of phosphorus, potassium, calcium, and magnesium from soils by an ionexchange resin procedure. *Commun. Soil Sci. Plant Anal.* 17, 547-566.
- Reijnders, L. 2014. Phosphorus resources, their depletion and conservation, a review. *Resour. Conserv. Recycl.* 93, 178-187.
- Rodrigues, M., Pavinato, P.S., Withers, P.J.A., Teles, A.P.B., Herrera, W.F.B., 2016. Legacy phosphorus and no tillage agriculture in tropical oxisols of the Brazilian savanna. *Sci. Total Environ.* 542, 1050-1061.

- Sanchez, P.A., Palm, C.A., Buol, 2003. Fertility capability soil classification: a tool to help assess soil quality in the tropics. *Geoderma*. 114, 157-185.
- Soil Survey Staff., 2014. *Keys to Soil Taxonomy*, twelfth ed. Washington, District of Columbia.
- Soltangheisi, A., Rodrigues, M., Coelho, M.J.A., Gasperini, M., Sartor, L.R., Pavinato, P.S., 2018. Changes in soil phosphorus lability promoted by phosphate sources and cover crops. *Soil tillage res.* 179, 20-28.
- Sousa, D.M.G., Lobato, L., 2002. *Cerrado: correção do solo e adubação*. EMBRAPA Cerrados, Planaltina, Brazil.
- Teles, A.P.B., Rodrigues, M., Bejarano Herrera, W.F., Soltangheisi, A., Sartor, L.R., Withers, P.J.A., Pavinato, P.S., 2017. Do cover crops change the lability of phosphorus in a clayey subtropical soil under different phosphate fertilizers?. *Soil Use Manage.* 33, 34-44.
- Tiecher, T., Rheinheimer, D.S., Kaminski, J., Calegari, A., 2012. Forms of inorganic phosphorus in soil under different long term soil tillage systems and winter crops. *Braz. J Soil Sci.* 36, 271-281.
- Tiessen, H., Moir, J.O., 1993. Characterization of available P by sequential extraction, in: Carter, M.R., (Ed.), *Soil sampling and methods of analysis*. Canadian Soc. of Soil Sci. Lewis Publ., Boca Raton, Florida. pp. 75–86.
- USEPA - United States Environmental Protection Agency, 1971. *Methods of chemical analysis for water and wastes*. Environmental Protection Agency, Cincinnati, Ohio.
- Vance, C.P., Unde-Stone, C., Allan, D.L. 2003. Phosphorus acquisition and use: critical adaptation by plants for securing a nonrenewable resource. *New Phytol.* 157, 423-447.
- Vu, D.T., Tang, C., Armstrong, R.D. 2008. Changes and availability of P fractions following 65 years of P application to a calcareous soil in a Mediterranean climate. *Plant Soil.* 304, 21-33.
- Witheres, P.J., Rodrigues, M., Soltangheisi, A., Carvalho, T.S., Guilherme, L.R.G., Benites, V.M., Gatiboni, L.C., Sousa, D.M.G., Nunes, R.S., Rosolem, C.A., Andreote, F.D., Oliveira Jr, A., Coutinho, E.L.M., Pavinato, P.S., 2018. Transitions to sustainable management of phosphorus in Brazilian agriculture. *Nature*. 8, 2537.

5. FINAL CONSIDERATIONS

Understand the effects of P fertilizer placements on long-term P accumulation in soils under different management systems is necessary for a more sustainable P-use management. Thus, the general objective of this study was to evaluate the effect of long-term P fertilizer placement (initial P fertilization - phosphating, band, deep-band, and broadcast) in three different management systems (conventional, reduced-tillage, and no-tillage) on changes in soil P pools (labile, moderately labile, and non-labile) and soil P legacy accumulation. For this study were used three adequately established field trials under consistent long-term P fertilizer management in three different soils and agricultural systems.

The first study (Chapter 2), demonstrated the changes in soil P pools (labile, moderately labile and non-labile) and legacy soil P accumulation after long-term P fertilization (12-years) with an initial soil P buildup and annual P applications in an Oxisol with adequate level of initial soil test P in Brazilian cerrado. Results showed a significant effect of P application on labile P fractions with annual rates of 60, 90 and 120 kg P₂O₅ ha⁻¹, while effects of the initial P application were less significant. Also, The rate of 97 P₂O₅ ha⁻¹ as annual base was the one responsible for optimum yield of both crops soybean and cotton. Thus, P accumulation from annual rates in the labile fractions will help to improve the soil legacy P availability and represent a profitable strategy to reduce large inputs of initial (phosphating) inorganic P fertilizer in tropical crop production systems

The second study (Chapter 3), demonstrated the influence of long-term P fertilizer placement (10-years) on soil P pools (labile, moderately labile, and non-labile) and legacy P accumulation under a long-term corn-soybean rotation in a Mollisol with low level of initial soil test P in North-central Kansas, US. Broadcast P fertilizer placement increased the amount of labile and moderately labile P in the topsoil (0-7.5 cm) and had a greater level of P fertilizer use efficiency compared to deep-band placement. However, the deep-band contributed to increase the labile and non-labile P pools at the 7.5-15 cm, but only for sampling in the row position. With reduced tillage, organic carbon amount increased in the soil surface (0-7.5 cm) that may have affected the soil P dynamics. Placing P fertilizer exclusively in one strategy over many years can promote depletion of P pools from some soil layers and row locations.

The third study (Chapter 4) evaluated the effects of P fertilizer placement (initial phosphating and annual rates and placement of P over the first 5 years), in the transition of pasture to double-crop of soybean/corn under no-till crop production, on changes in the soil P

pools (labile, moderately labile and non-labile) and legacy P accumulation, in an Oxisol with very low initial soil test P in Brazilian cerrado. Results showed effect of P application on labile P fractions in soil surface with the adoption of annual rates and placement, especially under 100 kg ha^{-1} of P_2O_5 as broadcast or sowing band under all initial P application. The broadcast of 100 kg ha^{-1} of P_2O_5 also influenced all other less labile P fractions and soil depths. Thus, labile P accumulation from the initial phosphating (P remains in the soil after five years) and annual P fertilizer application could help (at least in part) to reduce current high dependency and large inputs of P fertilizer in no-till systems.

Based on our findings, initial P fertilization (phosphating) was not effective to improve crops efficiency and soil P availability in long-term for Oxisols in Brazil. Annual fertilization by band-placement in a conventional tillage system and broadcast or band-placement in no-till were even effective to improve soil P availability. Still, broadcast and band-placement were similarly efficient to improve crops yield and soil P availability in long-term for Mollisols in Kansas, US. However, more investigation is recommended about the role of legacy P under a combined P fertilizer placement strategy, such as deep-band and broadcast placements under reduced-tillage in Mollisols and band and broadcast placement under no-tillage in Oxisols, to increase the available P in the soil profile while maintaining or increasing plant P uptake.

APPENDIX

APPENDIX A. Supplementary material relative to chapter 2

Supplementary Material - PHOSPHATE PLACEMENT AND RATES INFLUENCING SOIL LEGACY PHOSPHORUS IN A LONG-TERM COTTON/SOYBEAN ROTATION IN BRAZILIAN CERRADO

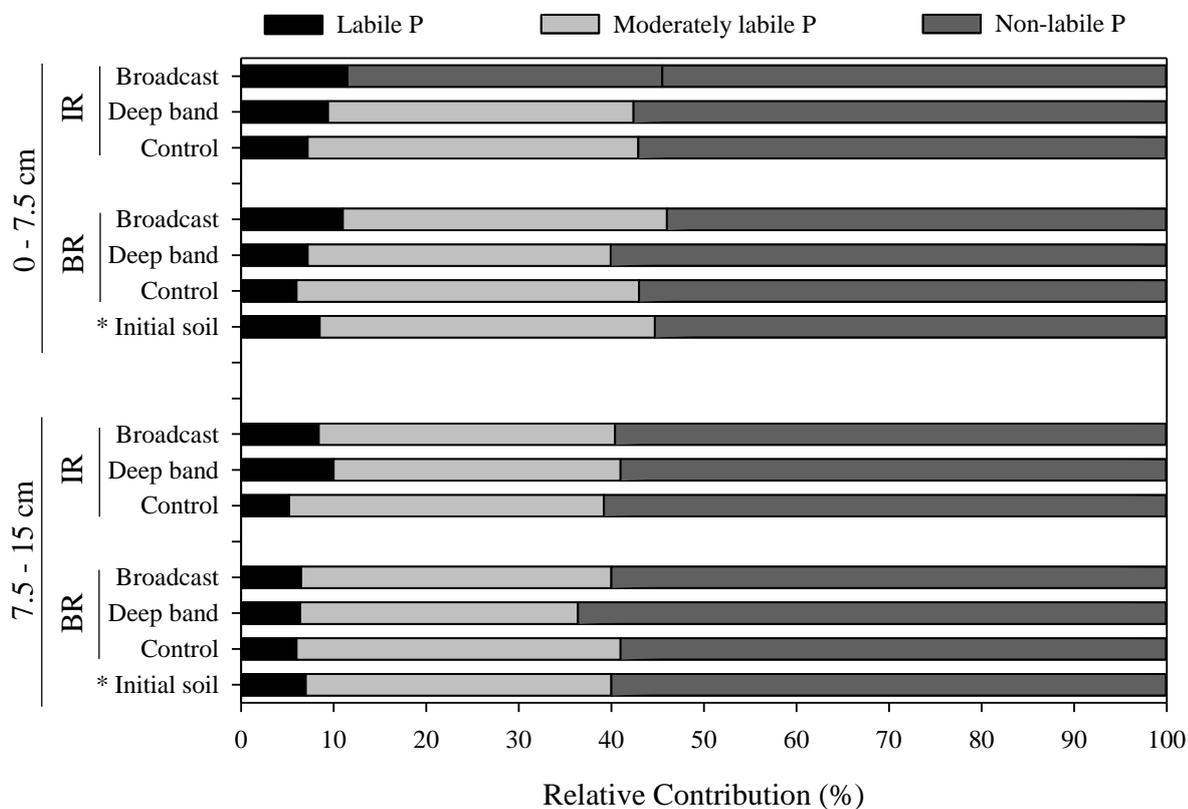
Supplementary Table 1. Summary of the analysis of variance (ANOVA) for P fractions at two soil depths after twelve-years cultivation period as affected by initial and annual rates of P fertilizer.

Fixed effect	Hedley P fractions									
	P _{AER}	P _{iBic}	P _{oBic}	P _{i0.1Hid}	P _{o0.1Hid}	P _{HCl}	P _{i0.5Hid}	P _{o0.5Hid}	P _{Residual}	P _{Total}
	----- p > F -----									
	<u>0 - 20 cm depth</u>									
Initial rates (IR)	0.038	0.041	0.604	0.013	0.413	0.031	0.779	0.015	0.767	0.749
Annual rates (AR)	<0.001	<0.001	0.012	<0.001	0.083	<0.001	0.345	<0.001	0.083	<0.001
IR × AR	0.887	0.710	0.097	0.063	0.578	0.910	0.831	0.162	0.180	0.124
	<u>20 - 40 cm depth</u>									
Initial rates	0.018	0.076	0.212	0.122	0.009	0.517	0.166	0.796	0.103	0.059
Annual rates	<0.001	<0.001	0.059	<0.001	<0.001	0.007	0.359	0.163	0.809	0.006
IR × AR	0.320	0.140	0.261	0.216	0.288	0.171	0.147	0.200	0.959	0.513

P_{AER}, Inorganic P extracted by anion exchange resin; P_{iBic}, Inorganic P extracted by 0.5 M NaHCO₃; P_{oBic}, Organic P extracted by 0.5 M NaHCO₃; P_{i0.1Hid}, Inorganic P extracted by 0.1 M NaOH; P_{o0.1Hid}, Organic P extracted by 0.1 M NaOH; P_{HCl}, Inorganic P extracted by 1 M HCl, P_{i0.5Hid}, Inorganic P extracted by 0.5 M NaOH; P_{o0.5Hid}, Organic P extracted by 0.5 M NaOH; P_{Residual}, Residual P; P_{Total}, P Total.

APPENDIX B. Supplementary material relative to chapter 3

Supplementary Material - LONG-TERM FERTILIZER PLACEMENT IN A CORN-SOYBEAN ROTATION AFFECT SOIL PHOSPHORUS FRACTIONS AND LEGACY



Supplemental Fig. S1. The relative contribution of labile, moderately labile and non-labile P fractions at two soil sampling depths and two sampling positions (between-row – BR; and in-row - IR) as affected by P fertilizer treatments (deep-band, broadcast, and control) after ten years of a corn-soybean rotation.

Supplemental Table S1. Abbreviations

H+Al - exchangeable acidity
K - exchangeable potassium
Mg - magnesium
STP - soil test P
Ca - calcium
Na - sodium
CEC - cation exchange capacity
Fe - extractable iron
Zn - zinc
Cu - cooper
Mn - manganese
TOC - total organic carbon
TON - total organic nitrogen
IR - in row
BR - between rows
P - phosphorus
MPAC - maximum P adsorption capacity
Pi - inorganic phosphorus
Po - organic phosphorus
AER - anion exchange resin
P _{AER} - P extracted by AER
P _{Bic} - Pi extracted by 0.5 M NaHCO ₃
P _{O_{Bic}} - Po extracted by 0.5 M NaHCO ₃
P _{i0.1Hid} - Pi extracted by 0.1 M NaOH
P _{O0.1Hid} - Po extracted by 0.1 M NaOH
P _{HCl} - Pi extracted by 1 M HCl
P _{i0.5Hid} - Pi extracted by 0.5 M NaOH
P _{O0.5Hid} - Po extracted by 0.5 M NaOH
P _{Residual} - residual P extracted by H ₂ SO ₄ + H ₂ O ₂ + MgCl ₂ digestion
P _{tBic} - total P extracted by autoclave digestion
P _{t0.1Hid} - total P extracted by autoclave digestion
P _{t0.5Hid} - total P extracted by autoclave digestion
LP - labile P (P _{AER} plus the P _{Bic} (Pi and Po))
MLP - moderately labile P ((P _{0.1Hid} (Pi and Po) plus the P _{HCl})
NLP - non-labile P (P _{0.5Hid} (Pi and Po) plus the P _{Residual})
PAE - P agronomic efficiency
PRE - P recovery efficiency
