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Diffusion and uptake of phosphorus, and root development of corn seedlings, in three contrasting subtropical soils under conventional tillage or no-tillage

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Abstract Soil mineralogy and management can have substantial effects on phosphorus (P) diffusion in soil and hence on root morphology. The aim of this study was to assess these effects in soils differing widely in P-sorbing components. Samples of a Rhodic Paleudult (RP), a Rhodic Hapludox (RH), and a Humic Hapludox (HH) under conventional tillage (CT) or no-tillage (NT) were placed in Petri dishes to assess P diffusion. Performance and root morphology in corn grown on soil samples subjected to three different P fertilization treatments (no fertilizer, surface, and deep fertilizer application) were examined. Phosphorus diffusion was slower in the HH than in the other soils by effect of its high P adsorption capacity because of its mineralogy. Soil management had no effect on P diffusion. Fertilization with P only affected plant-related variables in RH: surface fertilization increased root biomass, and deep fertilization shoot biomass and proportion of thick roots. Soil management affected plants growing on HH and RP: the plants grown under NT were taller (RP) and developed more fine roots (HH) than those under CT. Fertilization with P had little effect on plant growth in the goethite-rich soil (HH) or the one with high content in available P (RP). Our results suggest that management system in these soils has therefore greater influence on fertility than fertilization treatment.

Keywords Phosphorus adsorption · Phosphorus availability · Phosphorus diffusion · Root density

Introduction

The phosphorus (P) concentration in the soil solution is generally low because the phosphate ion is typically involved in many soil reactions. Thus, P is less mobile and bioavailable than other macronutrients (Hinsinger 2001), which is particularly apparent in crops grown on strongly weathered soils (Raghothama and Karthikeyan 2005; DeLong et al. 2013). This poor mobility requires using effective soil management systems and fertilization practices to increase the efficiency and bioavailability of P fertilizers (Barrow and Debnath 2014).

Phosphate in the vicinity of roots moves mainly through diffusive flux in the soil solution, from which it is absorbed by plants (Barber 1995; Santner et al. 2014). Phosphorus diffusive flux and availability are influenced by the soil moisture content, bulk density, clay content, mineralogy, P concentration in the soil solution, and microbial activity (Barber 1995; Hinsinger 2011; Liu et al. 2014). The high P adsorption capacity of iron and aluminum oxides and hydroxides in strongly weathered soils helps to maintain low levels of available phosphate (Johnson and Loeppert 2006; DeLong et al. 2013; Fink et al. 2014) and reduces its diffusive flux to the soil solution, thereby hindering or delaying approach of this anion to plant roots (Raghothama and Karthikeyan 2005).

Plants are capable of detecting available P in soil and of regulating growth and the architecture of their root system in response (Lopes-Bucio et al. 2003; Cahill et al. 2010). For

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instance, root length increases with increasing P availability (Malamy 2005; Flavel et al. 2014). However, a high P availability in soil can reduce growth of fine roots—and the explored soil volume as a result (Lopes-Bucio et al. 2003; Hinsinger 2011)—while increasing root diameter (Sánchez-Rodríguez et al. 2014). These effects can lead to irreversible losses in crop yield under adverse conditions such as water stress (Gregory 2006).

Most farmers use fertilizers by topdressing in order to increase the economic efficiency of agricultural machinery (Olibone and Rosolem 2010). This fact, together with the slow diffusion of P in soil, contributes to increase the concentration of P in the surface layer of soil; as a result, root systems must develop within a thin soil layer (Costa et al. 2009, 2010). Besides, there is an increased risk of P losses by erosion and the subsequent water eutrophication (Shigaki et al. 2007). Soil management also influences P mobility in soil. Thus, Sá et al. (2013) found corn yields higher in an Oxisol under no-tillage (NT) when fertilizers were applied on the soil surface in comparison to furrow application; by contrast, Prado and Fernandes (2001) observed the opposite.

The primary aim of this work was to assess the influence of soil properties and management system (conventional tillage or no-tillage) on P diffusion and growth of corn seedlings in three subtropical soils to which fertilizer P was applied either on the surface or below the seeds.

Material and methods

Soil samples

Samples of three subtropical soils including a Rhodic Paleudult (RP), a Rhodic Hapludox (RH), and a Humic Hapludox (HH) (Soil Survey Staff 2010) under no-tillage (NT) or conventional tillage (CT) of the 0–10-cm depth were collected for study from southern Brazil. The physical, chemical, and mineralogical properties of the soils are described in detail elsewhere (Fink et al. 2016) and summarized in Table 1. The contents in extractable P, K, Ca, and Mg were all above the critical levels for development of annual plants, and the pH was classified as "high" for RP, "medium" for RH, and "low" for HH according to CQFS-RS/SC (2004), with no significant differences in this respect between NT and CT samples.

In vitro phosphorus diffusion

Soil samples were placed in Petri dishes 1 cm deep×5 cm in diameter. A total of 72 dishes per soil were used: 2 management systems×3 experimental blocks (corresponding to three field replicates)×3 dishes (2 replicates+1 control without P addition)×4 sampling times (see Fig. 1). Four 8×20 mm sheets of anion exchange resin (AER; AMI-7001) were placed at the four quadrants on the lateral wall of each Petri dish (Fig. 1). Then, a dose of 68 mmol P kg^{-1} as $NH_4H_2PO_4$ was added at a depth of 0.5 cm in the center of each dish. This P dose corresponds to the amount of fertilizer added in 2 cm of crop furrow for a dose of 135 kg P_2O_5 ha^{-1}. Soil moisture was maintained at about 80 % field capacity. After 7, 14, 21, and 28 days of P application, the AER sheets were carefully removed, washed with deionized water to remove soil, and shaken with 10 mL of 0.5 mol HCl L^{-1} in an end-over-end shaker for 2 h to determine the concentration of P in the resulting solution according to Murphy and Riley (1962).

On day 28, soil samples from the central zone (CZ; 1.3-cm diameter) and outer zone (OZ) of the Petri dishes (Fig. 1) were

Table 1 Physical, chemical, and mineralogical properties of soils under conventional tillage (CT) and no-tillage (NT)

Property	Rhodic Paleudult		Rhodic Hapludox		Humic Hapludox	
	NT	CT	NT	CT	NT	CT
Clay (g kg^{-1})	205±5	225±5	571±1	586±3	634±4	631±4
Fe_d (g kg^{-1})	11.3±0.7	12.5±0.4	88.3±1.9	89.8±1.6	69.6±1.8	72.5±1.4
Gt (g kg^{-1})	6.4±0.4	6.8±0.3	26.6±0.8	27.1±0.7	86.1±2.4	89.8±1.8
Hm (g kg^{-1})	8.8±0.6	9.4±0.5	62.0±1.9	63.3±1.7	12.9±0.4	13.4±0.3
$pH_{1:1}$	6.3±0.2	5.9±0.1	5.2±0.0	5.1±0.2	5.4±0.1	4.7±0.3
Ca ($cmol_c$ kg^{-1})	3.6±0.2	2.9±0.1	7.2±0.8	6.7±0.5	7.6±1.8	5.6±1.2
Mg ($cmol_c$ kg^{-1})	2.2±0.1	2.1±0.1	3.2±0.1	3.7±0.1	3.3±0.8	2.4±0.4
K (mg kg^{-1})	146±16	126±20	247±35	213±18	207±29	130±29
P (mg kg^{-1})	68.4±11.8	41.1±3.1	24.2±4.4	22.9±2.5	14.3±1.5	5.9±2.6
P_{AER} (mg kg^{-1})	38.9±5.0	38.4±5.8	18.3±3.3	17.0±1.8	13.2±1.8	5.1±1.7
OC (g kg^{-1})	16.8±1.7	11.8±0.8	19.7±0.4	14.2±1.0	42.5±1.3	31.8±1.0

Mean±standard error for all variables

Clay content as determined with the pipette method; Fe_d Fe in pedogenic iron oxides as determined according to Mehra and Jackson (1960); *Gt* goethite; *Hm* hematite; *$pH_{1:1}$* pH in a 1:1 soil/water suspension; *Ca, Mg, K,* and *P* contents as assessed according to Tedesco et al. (1995); *P_{AER}* P extracted by an anion exchange resin (AER); *OC* organic C as assessed by dry combustion
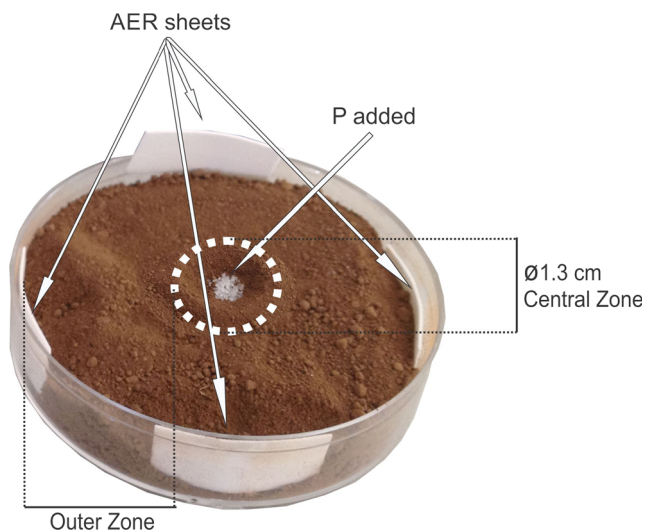
Fig. 1 Petri dish containing soil and four equally spaced sheets (8× 20 mm) of anion exchange resin (AER) in contact with the walls and P fertilizer in the central zone

collected and air-dried after removing the AER sheets. Then, recovery of previously added P was assessed by extracting P in the central and outer zones with one sheet of AER in a 1:10 soil/water suspension that was shaken for 16 h. Finally, the amount of P in the AER sheets was measured as described above.

Plant growth

Soil samples were placed in cylindrical 175-cm³ pots, 10 cm tall and 5 cm in diameter, and subjected to three different fertilization treatments, namely (1) control (no P fertilizer), (2) surface application (P fertilizer on the soil surface), and (3) deep application (P fertilizer 4 cm below the seeds). The P dose used in the surface and deep fertilization treatments was equivalent to 145 kg $P_2O_5\,ha^{-1}$, which is the regionally rec-ommended dose for a corn (*Zea mays*) yield of 9 ton ha^{-1} (CQFS-RS/SC 2004). Corn seeds were germinated on filter paper saturated with deionized water at 20 °C for 72 h, after which two germinated seeds were placed at a depth of 3 cm in each pot. The pots were then placed in a growth chamber with controlled photoperiod (12 h day^{-1}), temperature (21 °C day/ 18 °C night) and relative humidity (75 %). Only one corn plant per pot (experimental unit) was maintained after seed-ling emergence. The pots were periodically weighed and watered to maintain soil moisture near 80 % field capacity, and plant height was measured on a periodic basis.

Thirty days after germination, plants were cut above the soil surface to measure fresh biomass. Shoot plants were dried in an oven at 60 °C for at least 72 h to measure shoot biomass. Shoots were then ground, and digested with nitric and perchloric acids (Zasoski and Bureau 1977), to determine Ca, Mg, K, and P by atomic absorption spectrophotometry.

Soil with roots was removed from the pots and split into two layers according to depth: 0–5 and 5–10 cm. Each layer was washed with water to remove soil and then carefully distribut-ed onto a 20×30 cm acrylic plate to avoid overlap for scanning on an EPSON V700 PHOTO scanner equipped with a dual-lens system. Root density, surface area, and average diameter were determined with WinRHIZO® (Regent Instruments, Inc., Quebec, Canada). Fresh root biomass was then weighed, and roots were dried similarly to shoots for weighing.

Data analysis

The results were subjected to factorial analysis of variance (ANOVA) based on soil type (RP, RH, and HH), management system (NT and CT), and fertilization treatment (control, sur-face, and deep fertilization). When significant interaction be-tween soil type and management system or fertilization treat-ment was observed, the effect of soil management and fertil-ization treatment was compared separately in each type of soil (Figs. 3, 4, and 5; Table 2). Means were compared via Tukey's test at $p<0.05$. Pearson's correlation coefficients for P diffu-sion ($n=6$) and other variables ($n=36$) were also calculated. All statistical analyses were performed with the software Statistical Analysis System (SAS) for Windows.

Results and discussion

Phosphorus diffusion

Figure 2 shows the time course of P diffusion to the AER membranes in the three soils. Diffusion decreased in the fol-lowing sequence: RP (71.4 ± 2.8 µmol m^{-2})>RH (5.5 ± 1.1 µmol m^{-2})>HH (0.1 ± 0.1 µmol m^{-2}). Also, it seems to be related with the soil goethite content, which was 6.6, 26.8, and 88.3 g kg^{-1} in RP, RH, and HH, respectively. This result is consistent with the typically increased surface area and P ad-sorption capacity of goethite relative to hematite (Torrent et al. 1994). In contrast to HH, P diffusion in RH and RP under both management systems changed markedly with time (Fig. 2) until equilibrium was reached after about 14 days in RP and 21 days in RH. The increased contents in Fe oxides (goethite, hematite, and maghemite) of RH relative to RP resulted in increased P adsorption and hence in reduced P mobility, in the former soil.

No significant differences in P diffusion between NT and CT were found even though the soils under NT exhibited increased organic C contents (Table 1) that could have delayed P adsorption (Afif et al. 1995) and enhanced P diffusion.

Total P recovery from the central (CZ) and outer zones (OZ) of the Petri dishes at day 28 (Fig. 3) was 100 % in RP, 99 % in RH, and 80 % in HH. Again, the high goethite content of HH reduced P desorption relative to RH, which contained

Table 2 Corn root growth in the 0–5-cm soil layer in comparison to the 5–10-cm layer [(value for the 0–5 cm layer)/(value for the 5–10 cm layer)] in relation to soil management and P fertilizer placement

Treatment	Root density			Surface area			Average diameter		
	NT	CT	Tukey's test	NT	CT	Tukey's test	NT	CT	Tukey's test
Rhodic Paleudult (%)									
Control	0.66	0.78	ns	0.61	0.59	ns	0.93	0.84	ns
Surface	1.18	0.68		0.82	0.56		0.75	0.83	
Deep	0.67	0.42		0.54	0.40		0.92	0.95	
Tukey's test	NS			NS			NS		
Rhodic Hapludox (%)									
Control	0.70	0.48	b	0.65	0.44	b	0.93	0.92	b
Surface	1.10	0.71	a	0.98	0.62	a	0.90	0.88	b
Deep	0.45	0.30	b	0.44	0.35	b	1.00	1.20	a
Tukey's test	NS			A	B		NS		
Humic Hapludox (%)									
Control	0.76	0.90	b	0.66	0.72	b	0.87	0.81	ns
Surface	1.08	1.56	a	0.88	1.24	a	0.81	0.80	
Deep	0.74	1.00	b	0.64	0.82	b	0.87	0.82	
Tukey's test	NS			NS			A	B	

Uppercase letters compare soil management system, and lowercase letters P fertilization method, via Tukey's test ($\alpha=0.05$)

NT no-tillage, *CT* conventional tillage, *Control* no P fertilization, *Surface* P fertilizer applied to the soil surface, *Deep* P fertilizer applied 5 cm below seeds.

mainly hematite. Phosphate is adsorbed onto goethite (110) faces (90 % of crystal surfaces) as a binuclear complex (Parfitt et al. 1975); by contrast, only a few faces in hematite can form a binuclear complex (Barrón et al. 1988), phosphate being adsorbed largely as a mononuclear complex. Phosphate binding to iron oxides is stronger in binuclear complexes than in mononuclear complexes (Borggaard et al. 2005). Unexpectedly, P recovery from RH was higher under CT than under NT. As expected, CZ had higher P contents than OZ at day 28 (Fig. 2). The [(P in CZ)/(P in OZ)] ratio was approximately 2, 3, and 12 for RP, RH, and HH, respectively. This result suggests tha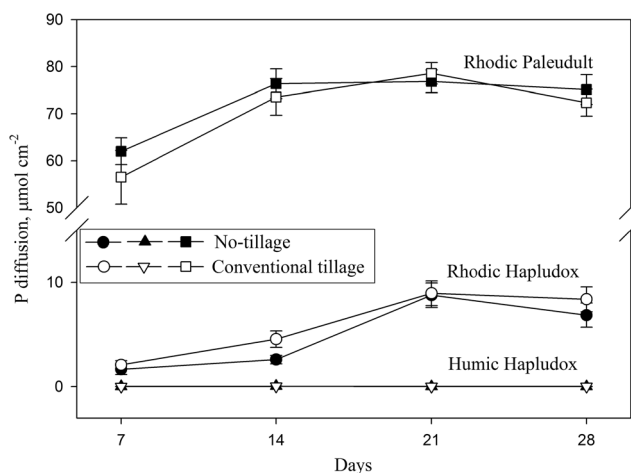t most P added to HH was adsorbed by the CZ soil (see Figs. 1 and 3). Based on the foregoing, P in fertilizer granules placed far from corn seeds is probably unavailable to plants in soils rich in Fe oxides (especially in those where goethite predominates).

Plant growth

Plants grown on RP were significantly taller, and exhibited greater shoot and root dry biomass, than plants grown on RH and HH (Table S1, Fig 4). Thus, the lower content in
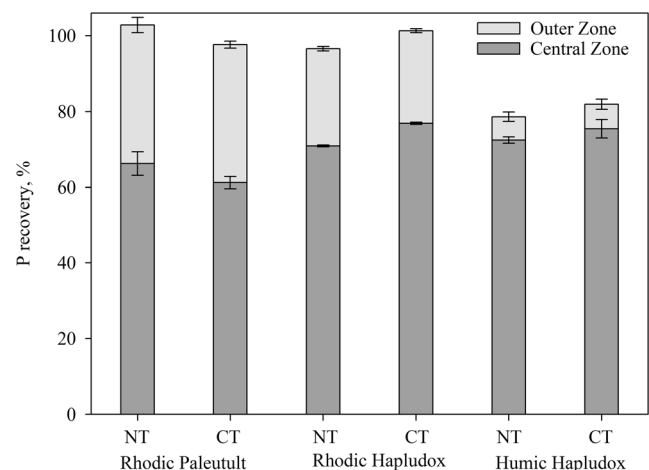

Fig. 2 Time course of P diffusion to the AER in the three soils under conventional tillage (CT) and no-tillage (NT)

Fig. 3 Percent recovery of added P (±standard error) in the central and outer zones of Petri dishes at day 28 in the three types of soil under conventional tillage (CT) and no-tillage (NT)
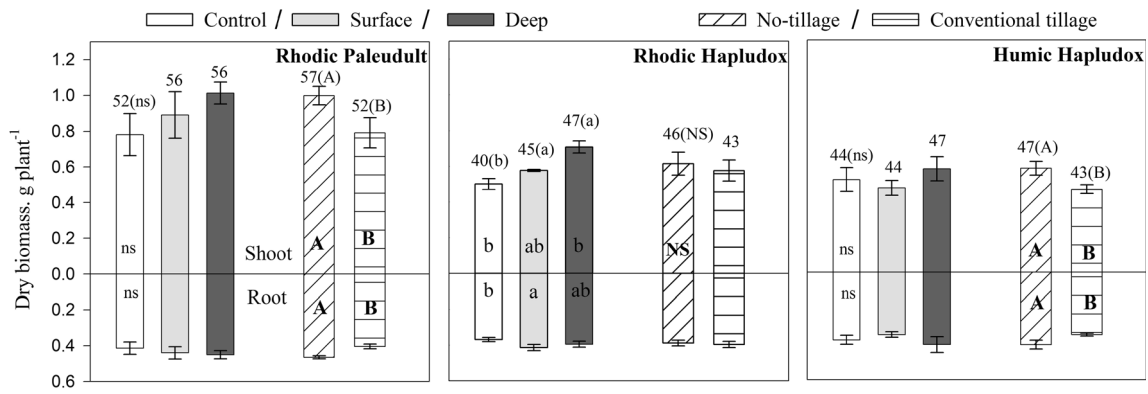
Fig. 4 Shoot and root dry biomass (*bars*), and height (*values* in cm above bars), of plants grown on the three soils as a function of P fertilization treatment (control (no P fertilizer added), surface P fertilization, and deep pedogenic Fe oxides (Fe_d) of RP apparently resulted in increased fertilization (fertilization below seeds)) and soil management system. *Uppercase letters* compare soil management systems, and *lowercase letters* P fertilization treatments, via Tukey's test ($\alpha = 0.05$)

pedogenic Fe oxides (Fe_d) of RP apparently resulted in increased P availability (Fink et al. 2014) and also better development of roots and shoots.

Phosphorus (deep and surface) fertilization increased plant height only in RH; deep fertilization increased shoot dry biomass, and surface fertilization root dry biomass, relative to the control plants (no P fertilization) (Fig. 4). On the other hand, soil management only affected plant height, and root and dry biomass, in HH and RP, the plants grown under NT exhibiting higher values of the three variables than those under CT (Fig. 4).

Soil P availability at the time of sampling had a substantial effect on plant height ($r = 0.91$; $p = 0.012$), shoot dry biomass ($r = 0.99$; $p < 0.001$), and root dry biomass ($r = 0.92$; $p = 0.009$). This result is consistent with that of Nadden et al. (2012), who found P uptake to start soon after radicles emerged and to be governed by P availability. Also, Noack et al. (2012) found vegetal residues accumulating on soil—soils under NT in our case—to account for an increased proportion of readily available P. These results suggest that, as previously noted by Costa et al. (2010) and Souza et al. (2014), soil P availability is developed over time by soil management—mainly in soils with high contents in Fe oxides.

Plant nutrition

No symptoms of nutritional deficiency were observed in any plants except those grown on HH. Nutrient contents in plant shoots ranged from 1.7 to 3.5 mg kg^{-1} for P, 4.6 to 5.6 mg kg^{-1} for Ca, 3.2 to 6.7 mg kg^{-1} for Mg, and 14.2 to 26.5 mg kg^{-1} for K, all of which except those for K are adequate for plant growth according to Jones Jr et al. (1991).

The P concentration was affected by the interaction between soil and fertilization treatment (Table S2). Plants grown on RP and RH stored more P when the fertilizer was placed below the seeds than when applied to the surface or not applied (control treatment). The RP soil, which had the highest P availability (68 mg kg for NT and 41 mg kg for CT samples) and the lowest goethite and Fe oxide contents (Table 1), allowed a considerable P diffusion (Fig. 2). Therefore, corn plants grown on this soil were able to easily uptake P (P that was in the soil before P fertilization and, in addition, P coming from the P fertilizer in two of the three treatments) without the limitations that plants grown on the HH soil found (see Fig. 5, P stored in shoot in each soil). As commented above, HH had the highest goethite content, which reduced P diffusion in soil
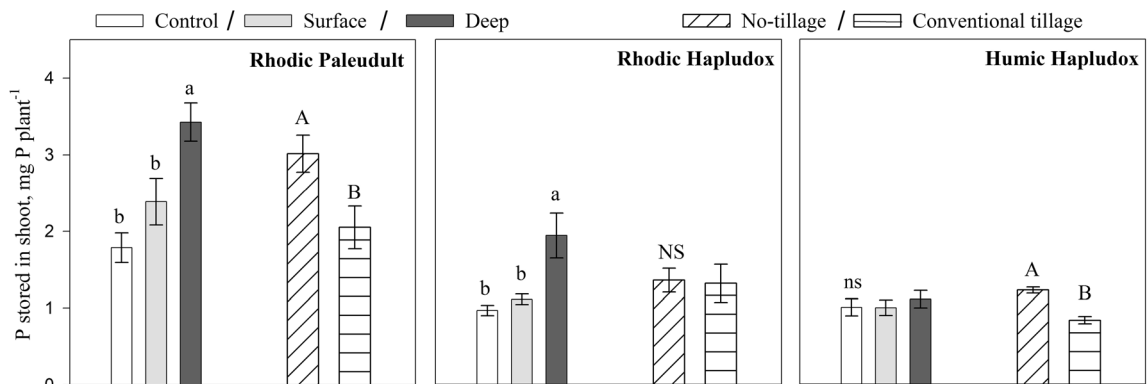

Fig. 5 Phosphorus in plant shoots as affected by P fertilization (control (no P fertilizer added), surface P fertilization, and deep fertilization (fertilization below the seeds)) and soil management system. *Uppercase letters* compare soil management methods, and the *lowercase letters* P fertilization treatments, via Tukey's test ($\alpha = 0.05$)

(Figs. 2 and 3) and, as a result, P bioavailability. These reasons could explain why differences due to P fertilization in plants grown on RP and HH soils were not significant in comparison with those grown on RH (P availability between RP and HH soils and lower goethite content than the HH soil). The plants grown on RP and HH contained more P under NT than under CT (Fig. 5) by effect of the increased P availability under NT (Table 1). Phosphorus uptake was correlated with available P in soil ($r=0.95$; $p=0.003$), the amount of P diffusing to AER ($r=0.89$; $p=0.018$), Fe_d ($r=-0.80$; $p=0.056$), and root dry biomass ($r=0.70$; $p<0.001$).

Root growth

Fewer roots developed in the 0–5-cm (surface) layer than in the 5–10-cm (deeper) layer; also, root dry biomass in the surface layer was higher in the plants grown on RH and HH than in those grown on RP (Table S3). Only in RH were the differences significant as a result of the effect of soil management and fertilization treatment on root dry biomass in the surface layer. Therefore, plants grown under NT developed more roots in the surface layer than those grown under CT; also, plants grown on deep-fertilized soil developed more roots in the 5–10-cm layer than those grown on surface-fertilized soil (Fig. 6). One reason might be the better structure of the NT relative to the CT soil (because of the higher organic C content of the NT soil) allowing a better root development in the initial plant growth stages (when roots are mostly in the 0–5-cm layer). Moreover, in soils under NT, an increase in microbial activity occurs (Souza et al. 2014), which can solubilize considerable amounts of adsorbed P in acid soils (Liu et al. 2014)—mainly in the surface layer. On the other hand, phosphate fertilization can also increase the microbial abundance, especially fungal abundance (Li et al. 2015)— providing more P to plants.

Root density ranged from 6 to 11.9 cm cm^{-3} and surface area from 0.51 to 1.09 cm^2 cm^{-3}, both being affected by soil type and fertilization treatment; overall, the plants grown on

Table 3 Root length as function of root diameter class as affected by fertilization treatment in the Rhodic Hapludox and by soil management in the Humic Hapludox

Diameter	Length by diameter class (%)					
	0.1–0.2 mm		0.2–0.5 mm		0.5–2.0 mm	
Depth (cm)	0–5	5–10	0–5	5–10	0–5	5–10
Rhodic Hapludox						
Control	61.0 a	56.5 ns	31.4 ns	31.3 ns	7.6 b	12.2 a
Surface	59.2 ab	56.9	34.8	31.7	5.9 b	11.4 ab
Deep	54.9 b	57.2	34.9	33.3	10.2 a	9.5 b
Humic Hapludox						
NT	62.8 A	51.7 A	30.7 B	36.0 NS	6.4 B	12.2 B
CT	57.3 B	46.9 B	34.9 A	36.9	7.7 A	16.2 A

Uppercase letters compare soil management system, and lowercase letters P fertilization method, via Tukey's test ($\alpha=0.05$)

NT no-tillage, *CT* conventional tillage, *Control* no P fertilization, *Surface* P fertilizer applied to the soil surface, *Deep* P fertilizer applied 5 cm below seeds

HH had the highest root density and surface area (Table S4). Plants grown on RH and HH explored a greater soil volume— and had a higher root density and surface area as a result—in the layer where the fertilizer was added (Table 2). Specifically, the root density of plants grown on deep-fertilized RH was approximately 37 % greater in the surface layer than in the deep layer. In addition, deep fertilization in RH resulted in plants developing thinner roots in the subsurface layer than in the surface layer. Phosphorus uptake was correlated with root density ($r=0.52$; $p<0.001$) as the likely result of the increased soil volume explored. In fertilized zones (Flavel et al. 2014) or zones with enhanced P availability (Li et al. 2012), greater soil exploration by roots has been observed. This suggests that the strong plastic behavior of roots related to P availability can be an important mechanism to increase fertilizer efficiency.
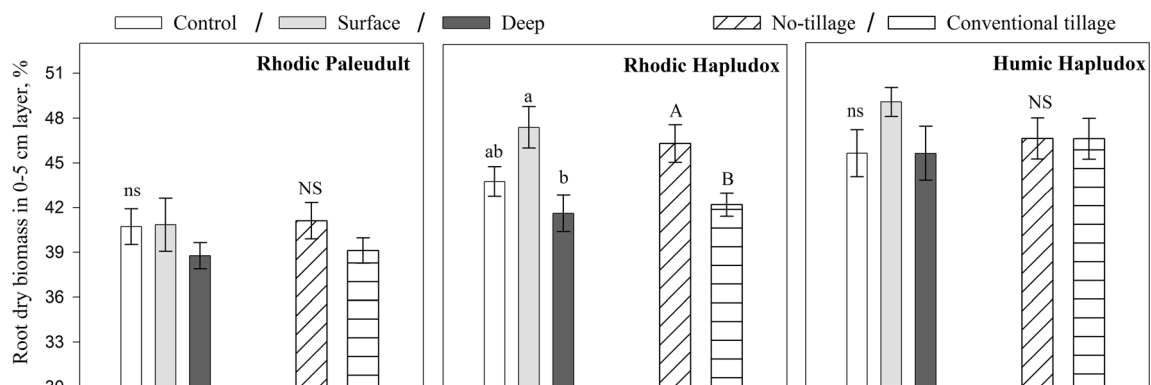

Fig. 6 Root dry biomass in the 0–5-cm layer relative to total root dry biomass (roots in 0–5-cm layer+roots in 5–10-cm layer) of plants grown on the three types of soil as affected by P fertilization treatment (control (no P fertilizer added), surface P fertilization, and deep (fertilization below the seeds)) and soil management system. *Uppercase letters* compare soil management methods, and *lowercase letters* P fertilization treatments, via Tukey's test ($\alpha=0.05$)

Root morphology

Plants grown on RP soil exhibited an increased proportion of fine roots relative to the other soils at both depths, whereas those grown on HH tended to develop a higher proportion of thick roots (0.2–0.5 and 0.5–2.0 mm) (Tables 5S and 6S). Because RP contained greater amounts of available nutrients (Table 1), these results are consistent with those of Lopes-Iglesias et al. (2014), who found a lower proportion of fine roots where the soil solution was rich in nutrients.

The roots of plants growing on RH were affected by fertilization treatment; thus, deep fertilization resulted in a low proportion of fine roots in the 0–5-cm layer and a low proportion of thick roots in the 5–10-cm layer relative to surface fertilization (Table 3). Lynch (2007) previously found corn to develop its main roots in zones of low P availability and secondary roots to grow preferentially in zones of high P availability.

Soil management only affected root distribution in plants growing on HH, the proportion of fine roots (0.1–0.2 mm) being higher under NT and that of thicker roots under CT (Table 3). Indeed, the proportion of fine roots was significantly correlated with P uptake ($r = 0.59$; $p < 0.001$).

Conclusions

Phosphorus diffusion was correlated with the nature and content in pedogenic iron oxides, which enhance P adsorption and decrease P diffusion—and hence P uptake. Goethite seemingly restricted P diffusion and uptake more efficiently than did hematite. No effect of P fertilization on shoot dry biomass was observed in plants grown on the soil with the highest P adsorption capacity (Humic Hapludox) or P availability (Rhodic Paleudult). The plants grown on the Rhodic Hapludox exhibited an increased root density and root surface area, and a decreased proportion of thick roots, in the layer where P fertilizer was applied. No-tillage in the Humic Hapludox, which had a high goethite content, increased growth of fine roots relative to conventional tillage.

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