MINERALIZATION OF ORGANIC PHOSPHORUS IN SOIL SIZE FRACTIONS UNDER DIFFERENT VEGETATION COVERS IN THE NORTH OF RIO DE JANEIRO⁽¹⁾

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SUMMARY

In unfertilized, highly weathered tropical soils, phosphorus (P) availability to plants is dependent on the mineralization of organic P (Po) compounds. The objective of this study was to estimate the mineralization of total and labile Po in soil size fractions of > 2.0, 2.0-0.25 and < 0.25 mm under leguminous forest tree species, pasture and "capoeira" (secondary forest) in the 0-10 cm layer of a Red-Yellow Latosol after 90 d of incubation. The type of vegetation cover, soil incubation time and soil size fractions had a significant effect on total P and labile P (Pi and Po) fraction contents. The total average Po content decreased in soil macroaggregates by 25 and 15 % in the > 2.0 and 2.0-0.25 mm fractions, respectively. In contrast, there was an average increase of 90 % of total Po in microaggregates of < 0.25 mm. Labile Po was significantly reduced by incubation in the > 2.0 (-50 %) and < 0.25 mm (-76 %) fractions, but labile Po increased by 35 % in the 2.0-0.25 mm fraction. The Po fraction relative to total extracted P and total labile P within the soil size fractions varied with the vegetation cover and incubation time. Therefore, the distribution of P fractions (Pi and Po) in the soil size fraction revealed the distinctive ability of the cover species to recycle soil P. Consequently, the potential of Po mineralization varied with the size fraction and vegetation cover. Because Po accounted for most of the total labile P, the P availability to plants was closely related to the mineralization of this P fraction.

Index terms: Bowman's extraction method, leguminous forest tree species, pasture, capoeira (secondary forest).

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RESUMO: *MINERALIZAÇÃO DAS FRAÇÕES ORGÂNICAS DE FÓSFORO EM CLASSES DE AGREGADOS DO SOLO, SOB DIFERENTES COBERTURAS VEGETAIS DO NORTE FLUMINENSE*

Em solos tropicais altamente intemperizados, não fertilizados, a disponibilidade de P para as plantas é dependente da mineralização de compostos orgânicos com P (Po). O objetivo deste trabalho foi estimar a mineralização de Po total e de lábil em classes de agregados (>2,0, 2,0-0,25 e < 0,25 mm) de solos sob leguminosas florestais, pastagem e capoeira de um Latossolo Vermelho-Amarelo na profundidade de 0-10 cm, após incubação por 90 d. A cobertura vegetal, o tempo de incubação e as classes de agregados do solo tiveram efeito significativo sobre os teores das frações de P total e P lábil (Pi e Po). Houve reduções médias de Po total nos macroagregados do solo: 25 e 15 % nas frações > 2,0 mm e 2,0-0,25 mm, respectivamente. Ao contrário nos microagregados (< 0,25 mm), ocorreu aumento médio de 90 %. Para o Po lábil, houve acentuada redução com o tempo de incubação nas frações > 2,0 mm (-50 %) e < 0,25 mm (-76 %), mas aumento de 35 % na fração 2,0-0,25 mm. Nas classes de agregados dos solos, a proporção da fração Po em relação ao P total e P lábil total extraídos variou com a cobertura vegetal e o tempo de incubação. Desse modo, a distribuição das frações de P (Pi e Po) nas classes de agregados dos solos revelou distinta capacidade das coberturas vegetais em reciclar o P do solo. Em consequência, o potencial de mineralização de Po no solo variou com a classe de agregados e a cobertura vegetal. Como o Po predominou na composição do P lábil total, a disponibilidade de P para as plantas estaria estreitamente relacionada à mineralização dessa fração de P.

Termos de indexação: método Bowman, leguminosas florestais, pastagem, capoeira.

INTRODUCTION

Phosphorus (P) is one of the most limiting nutrients for the productivity of the majority of crops grown on the highly weathered soils of tropical environments (Novais & Smyth, 1999). Particularly, in soils under forest cover and pasture, organic forms of P (Po) may represent 15 to 37 % of total extracted P, and 41 to 87 % of the total labile P is in organic form (Cunha et al., 2007). Under these conditions, Po becomes a major source of P to plants through decomposition and mineralization of the labile Po fraction, which is easily mineralized, thereby contributing to the availability of P to plants (Gatiboni, 2003; George et al., 2006).

Soil Po is derived from microorganism, plant or animal residues and can be recycled by the microbial biomass or stabilized in the mineral phase of the soil (Ohel et al., 2004). As proposed by McGill & Cole (1981), mineralization can be divided into two processes: 1) biochemical mineralization in which inorganic phosphorus (Pi) is released from organic compounds through phosphatase exoenzymes and regulated by P demand; and 2) biological mineralization, which is the release of Pi from organic materials during carbon (C) oxidation by soil organisms and regulated by the energy demand.

The content of Po in soil is closely associated with the capacity of organic C accumulation in the soil (Nizigueba & Bünemann, 2005). Therefore, mechanisms that are involved in the accumulation of organic C in the soil could be applied to Po to a certain extent. Among these mechanisms, the physical protection of soil organic matter through occlusion within aggregates or in small pores has been considered an important mechanism for reducing the bioavailability and accessibility of soil organic matter by microorganisms and soil enzymes (Gama-Rodrigues et al., 2010). Furthermore, according to the aggregate size classes, the C of organic compounds (OCs) has different levels of lability. The OCs associated with macroaggregates (> 250 mm) are more labile and represented largely by light organic matter, while OCs associated with microaggregates are more recalcitrant and mainly represented by the more stable fraction (Six et al., 2002; Bronick & Lal, 2005; Cadisch et al., 2006). Based on this mechanism of physical protection of OCs in soil, the Po distribution in the different size fractions could also show different degrees of lability in Po fractions and, consequently, a differentiated mineralization potential. Therefore, the Po mineralization rate would tend to be higher in macroaggregates and lower in microaggregates. In contrast, as Po accumulation in the soil is influenced by the type of vegetation cover (Cunha et al., 2007; Zaia et al., 2008), the distribution of P fraction in the size fractions and their mineralization potential could also be influenced by the type of land use.

The aim of this study was to estimate the mineralization potential of total and labile Po in soil size fractions under leguminous trees, pasture and *capoeira* (secondary forest).

MATERIAL AND METHODS

Description of sampling areas and soil sampling

Soil samples were collected from a (Tb)-kaolinitic Red-Yellow-Latosol in an undulated relief with a slope

of approximately 35 cm m⁻¹ under 12-year-old monoculture plantations of Mimosa caesalpiniifolia (sabiá) and Acacia auriculiformis spaced 3 x 2 m in 1,500 m² plots, on the Fazenda Carrapeta in Conceição de Macabú, State of Rio de Janeiro (RJ) (21º 37' S, 42° 5' W). Adjacent to the leguminous tree plantations, soil samples were collected from degraded pasture areas with the predominance of molasses grass (Melinis minutiflora), coastal sand paspalum (Paspalum maritimum) and Brazilian satintail (Imperata brasiliensis), with low forage production and no phosphate fertilization. Soil samples were also collected from a secondary Atlantic forest fragment with species in different successional stages (*capoeira*). Both vegetation covers were approximately 40 years old. All plots of vegetation covers were adjacent to each other, forming a continuous landscape. The pasture stocking rate was 0.5 animal/ha.

Four composite soil samples were collected for each vegetation cover, and each composite sample was composed of 15 random single samples collected by opening mini-trenches with depths of 0-10 cm. The composite samples were packed in polyethylene bags. Soil samples were air dried and pressed through 2 mm sieves. Chemical and physical characterizations of the soil (Table 1) were obtained according to the methods described by the Brazilian Agricultural Research Corporation (Embrapa, 1999) as follows: pH in water; P and potassium (K) extractable by Mehlich-1 with P being determined by colorimetry and K by flame photometry; exchangeable calcium (Ca), magnesium (Mg) and aluminum (Al) by 1 mol L⁻¹ KCl with Ca and Mg being determined by atomic absorption spectrophotometry and Al by titration with 0.025 mol L⁻¹ NaOH; total nitrogen (N) by the Kjeldahl method; organic C by oxidation with 1.25 mol L⁻¹ K₂Cr₂O₇ in acidic medium (Anderson & Ingram, 1996); particle-size analysis by the pipette method; and total P by nitric-perchloric acid digestion (Bataglia et al., 1983).

Fractionation in soil size fractions

Wet sieve analysis (Rita et al., 2011) was used to quantify and measure the distribution of water-stable aggregates using 100 g of pre-moistened soil. Soil was pre-moistened with a sprayer and left to stand for 4 h to prevent disintegration during mechanical mixing of the soil with water. The soil was then transferred to a set of sieves (mesh 2.0 and 0.25 mm). The soil samples were vertically shaken for 15 min in a Yoder apparatus (amplitude 4 cm, 32 oscillations/min). The samples were removed from the sieves by a jet from a plastic wash bottle, transferred to previously weighed Petri dishes and dried in an incubator at 105 °C for 24 h. As a result, three size fractions were obtained: > 2.0, 2.0-0.25 and < 0.25 mm.

Soil moisture was determined by weighing 20 g of sample in an aluminum crucible of known weight, the sample incubated at 105 °C until reaching constant weight and maintained in a desiccator thereafter. The weights of the samples dried at 105 °C were used to calculate the percentage of each aggregate size fraction by the following equation:

% of aggregates = (weight of aggregates in the size fraction/weight of the initial sample) x 100

Determination of the organic P fraction

The Bowman extraction method was used to quantify total Po compounds (Bowman, 1989). Inorganic P (Pi) was determined after clarification of acidic and alkaline extracts with activated charcoal (Guerra et al., 1996). In the acidic and alkaline extracts Pi, total P (PT) and Po were determined, and the total Po was calculated as the sum of Po in all extracts as follows:

Po acid = PT acid (digested) - Pi acid (extracted) Po alkaline = PT alkaline (digested) - Pi alkaline (extracted)

Po $_{total}$ = Po $_{acid}$ + Po $_{alkaline}$

Table 1. Chemical and physical characterization of soils under leguminous trees, pasture and capoeira (secondary forest)

Property	Acacia auriculiformis	Mimosa caesalpiniifolia	Pasture	Capoeira	
pH (H ₂ O)	4.8	4.5	4.7	4.2	
C (g kg ⁻¹)	37.9	37.6	35.8	37.5	
N (g kg ⁻¹)	0.8	1.5	1.5	1.3	
P (mg dm ⁻³)	2.8	2.4	2.0	2.9	
Pt (mg dm ⁻³) ⁽¹⁾	451	472	574	437	
K ⁺ (cmol _c dm ⁻³)	0.13	0.14	0.06	0.09	
Ca^{2+} (cmol _c dm ⁻³)	1.09	0.64	0.08	0.09	
Mg^{2+} (cmol _c dm ⁻³)	0.33	0.32	0.28	0.08	
Al ³⁺ (cmol _c dm ⁻³)	0.27	0.82	0.95	1.35	
Areia (g kg ⁻¹)	739	632	636	540	
Silt (g kg ⁻¹)	76	77	79	74	
Clay (g kg ⁻¹)	185	291	285	386	

⁽¹⁾ Pt = total P by nitric-perchloric acid digestion.

Labile Po (rapid mineralization) was determined by extraction with 0.5 mol L^{-1} NaHCO₃ (Bowman & Cole, 1978). In addition, activated charcoal was used for clarification of the extracts for Pi determination. Labile Po was determined by the difference between PT and Pi. The Pi concentrations in all extracts and digestion were determined by the method of Murphey & Riley (1962).

Mineralization of organic P

Soil samples and each aggregate size class samples were weighed (20 g), placed in small plastic pots, moistened to 60 % field capacity and placed in a BOD incubator at a constant temperature of 40 °C for 90 d (T1) for the Po mineralization test. Every 3 d, the moisture was restored to the initial level of incubation (T0). The amount of mineralized total Po and labile Po during the incubation time was estimated according to Acquaye (1963), based on the following equations:

 $Po_{min} = Po_{T1}-Po_{T0}$ P-Po_{min} = [(Po_{T1} - Po_{T0})/Po_{T0}] x 100

where $Po_{min} (mg kg^{-1})$ is the potentially mineralized $Po; Po_{T1} (mg kg^{-1})$ is the Po content determined after a 90 d incubation; $Po_{T0} (mg kg^{-1})$ is the Po content measured at time zero (before incubation); and P-Po_{min} (%) is the proportion of Po_{min} in Po_{T0} .

Negative values indicate reduction in Po amount with incubation indicating occurrence of mineralization of the fraction.

Statistical analysis

For the incubation assay, data for each vegetation cover with comparison of incubation times and contents of Pi and Po in soil size fractions were subjected to variance analysis in a completely randomized design with four replications in a factorial design of four vegetation covers, two incubation times and three size fractions as variation sources. The Tukey's test at 5 % was used to compare the means. Each cover was considered a non-randomized treatment.

RESULTS AND DISCUSSION

Organic P (Po) mineralization in soil

A significant effect of vegetation cover and incubation times on total soil contents of Pi and Po fractions was observed. Soil incubation led to an increase in the total Pi average content of 24 %, and a decrease in the total Po average content of 16 %. Nevertheless, the effect of incubation times on total Po differed according to the vegetation covers. There was a decrease in the Po levels in soils under *Acacia auriculiformis* and capoeira, but an increase in soil under pasture. Moreover, no significant changes were observed in soil under *Mimosa caesalpiniifolia* (Table 2). Soils under vegetation covers differed significantly in total Po in comparison to total Pi after both incubation times. At time zero (T0), the highest and lowest total Po contents were found in soil under *Acacia auriculiformis* and pasture, respectively. In contrast, soils under *Mimosa caesalpiniifolia* and capoeira showed the highest and the lowest contents of total Po, respectively, after 90 d of incubation (T1).

Rates of P (Pi + Po) recovered by Bowman's extraction method (Table 2) relative to the total soil P measured by the nitric-perchloric acid digestion (Table 1) at T0 were 66, 65, 40, and 60 %, whereas rates after T1 were 70, 70, 60, and 57 % for soils under *Acacia auriculiformis*, *Mimosa caesalpiniifolia*, pasture and capoeira, respectively.

The effect of vegetation cover and incubation time on the labile Pi and Po fraction contents was different (Table 2). On average, the increase in labile Pi was not significant with incubation time. Only soil under pasture differed significantly in the contents of this fraction. In contrast, labile Po showed a significant average increase of 18 %. However, the effect of incubation time was different for each vegetation cover. Labile Po significantly increased in soil under Acacia auriculiformis and decreased in soil under pasture. In soils under *Mimosa caesalpiniifolia* and capoeira, there were no significant differences in labile Po contents. Furthermore, differences in labile Po between the vegetation covers varied for each incubation time. At T0, only soil under Acacia auriculiformis was significantly different from the other cover types with the lowest content of labile Po. After T1, the labile Po contents were highest under capoeira and Mimosa caesalpiniifolia, followed by Acacia auriculiformis, with an intermediate value and pasture with the lowest content.

Pi was the major component of the total P (Pi + Po) composition in soils under each vegetation cover and after both incubation periods (Table 3). In general, the average Po was 26.7 and 19.8 % of total P extracted after T0 and T1, respectively. After T0, the greater proportion of Po was found in the soil under Acacia auriculiformis, and the soil under Mimosa caesalpiniifolia showed the highest proportion of this fraction after incubation for 90 d. The decrease in Po proportion in the soil under Acacia auriculiformis and capoeira with incubation would be due to its reduced content in this fraction (Table 2). However, although there was an increase in Po content in the soil under pasture, its ratio to total P did not change, but the Po content in the soil under Mimosa caesalpiniifolia remained unchanged after both incubation periods, resulting in a similar ratio of this fraction in the total P composition.

In contrast to the total P (Pi + Po) fraction, the labile Po fraction predominated over the labile Pi of the total labile P in the soil (Table 3). On average, the labile Po represented 70.1 and 65.8 % of the total labile P extracted after T0 and T1, respectively. At T0, the

			Labile						
Cover	Pi		Ро		Pi		Ро		
	TO	T1	T0	T1	T0	T1	T0	T1	
	mg kg ⁻¹								
A. auriculiformis	$191.0 \ Bb^{(1)}$	258.9 Ba	107.8 Aa	56.1 Cb	$2.0Aa^{\left(2\right)}$	2.2 Aa	2.3 Bb	3.9 Ba	
M. caesalpiniifolia	225.8 Ab	244.7 Ba	81.7 Ba	84.2 Aa	1.8 ABa	2.2 Aa	5.2 Aa	5.9 Aa	
Pasture	182.6 Bb	272.8 Aa	46.9 Db	73.9 Ba	1.0 Bb	2.0 Aa	3.2 ABa	2.1 Cb	
Capoeira	199.6 ABa	211.6 Ca	60.8 Ca	36.0 Db	1.5 ABa	2.1 Aa	4.9 ABa	6.3 Aa	
Average	199.8 b	247.0 a	74.3 a	62.5 a	1.6 b	2.1 a	3. 9 a	4.6 a	

Table 2. Contents of total inorganic (Pi), total organic (Po), labile inorganic (labile Pi) and organic (labile Po) phosphorus of soils under leguminous trees, capoeira and pasture at incubation time zero (T0) and after 90 d of incubation (T1)

⁽¹⁾ For Total P forms, averages followed by the same uppercase letters in the column (comparing covers in each incubation time) and averages followed by the same lowercase letters in the row (comparing each cover at different times) are not significantly different by Tukey's test at 5 %. ⁽²⁾ For Labile P forms, averages followed by the same uppercase letters in the column (comparing covers in each incubation time) and averages followed by the same lowercase letters in the row (comparing each cover at different times) are not significantly different by Tukey's test at 5 %.

Table 3. Percentage distribution of total inorganic (Pi), total organic (Po), labile inorganic (PiL) and organic (PoL) phosphorus contents of soils under leguminous trees, pasture and capoeira at incubation time zero (T0) and after 90 d of incubation (T1)

Cover	Pi/Pt	Po/Pt	Pi/Pt	Po/Pt	PiL/ PLtotal	PoL/ PLtotal	PiL/ PLtotal	PoL/ PLtotal
	Т	0	Т	1	Т	0	r	Γ1
				0	%			
A. auriculiformis	$63.8 c^{(1)}$	36.2 a	82.2 b	17.8 c	$46.2 a^{(2)}$	53.8 b	35.5 b	64.5 b
M. caesalpiniifolia	73.4 b	26.6 b	74.9 d	25.6 a	$25.2 \mathrm{b}$	74.8 a	27.0 с	73.0 a
Pasture	79.5 a	20.5 c	78.7 с	21.3 b	24.6 b	75.4 a	49.0 a	51.0 c
Capoeira	76.6 ab	23.4 bc	85.4 a	14.6 d	23.6 b	76.4 a	25.3 с	74.7 a
Average	73.3	26.7	80.2	19.8	29.9	70.1	34.2	65.8

⁽¹⁾ For total P forms, averages followed by the same letter in the column do not differ statistically by Tukey's test at 5 %; ²⁾ For Labile P forms, averages followed by the same letter in the column do not differ statistically by Tukey's test at 5 %.

lowest content of labile Po was measured in soil under *Acacia auriculiformis*, and the labile Po levels under other covers were not significantly different. The soil under pasture had the lowest contents of labile Po after soil incubation.

Mineralization of Po in soil size fractions

The soil size fraction distribution varied according to the cover type, and the highest and lowest aggregate percentages were observed in the 2.0-0.25 and < 0.25 mm fractions, respectively (Table 4). Macroaggregates represented approximately 95 % of the size fractions in every cover. Among covers, leguminous trees and capoeira showed the highest aggregate values in the > 2.0 mm fraction (Table 5). In turn, pasture had a higher percentage of aggregates in the 2.0-0.25 mm fraction. These results suggested that different vegetation covers significantly affect the stability and formation of soil aggregates (Roldán et al., 2005). Consequently, the accumulation of soil organic matter and nutrients are closely associated with vegetation covers (Rita et al., 2011).

The vegetation cover, incubation time and soil aggregate size classes had significant effects on the contents of total P and labile P fractions (Table 5). The aggregate size classes differed in their ability to retain the fractions of total P (Pi and Po) according to their vegetation cover and incubation time (Table 5). In general, smaller amounts of Pi were found in the < 0.25 mm aggregates after T0 and T1. Under all vegetation covers and in all aggregate classes, total Pi contents increased with incubation. However, the same was not observed for total Po. In general, the < 0.25 mm size fraction had the lowest and highest levels of total Po after T0 and T1, respectively. There was an average decrease of 25 % in the total Po in the

Size fraction	A. auriculiformis	M. caesalpiniifolia	Pasture	Capoeira	
mm		%			
> 2	$27.0 \ \mathrm{Ba}^{(1)}$	25.6 Ba	16.1 Bb	27.7 Ba	
2.0-25	69.0 Ab	70.4 Ab	79.8 Aa	66.9 Ab	
< 0.25	3.2 Ca	2.7 Ca	3.3 Ca	3.7 Ca	

Table 4. Distribution of stable soil size fractions in water from soils under leguminous trees, pasture and capoeira

⁽¹⁾ Averages followed by the same uppercase letters in the column (comparing size fraction in each cover) and averages followed by the same lowercase letters in the row (comparing different covers in each size fraction) are not significantly different by Tukey's test at 5 %.

Table 5. Contents of total organic (Po), labile inorganic (labile Pi) and organic (labile Po) phosphorus in different size fractions and vegetation covers at incubation time zero (T0) and after 90 d of incubation (T1)

		Total				Labile				
Cover Siz	Size fraction	Pi		Ро		Pi		Ро		
		T0	T1	T0	T1	TO	T1	TO	T1	
	mm		mg kg ⁻¹							
A. auriculiformis	> 2	$333.4{\rm Aa}^{(1)}$	321.0 Aa	52.3 Bb	62.3 Ba	$3.5 \mathrm{Ba^{(2)}}$	4.8 Aa	10.8 Ba	2.2 Bb	
	2.0-25	236.4 Ba	278.8 Ba	57.6 Ab	66.7 Ba	2.2 Ba	2.0 Ba	6.3 Ca	4.0 Ab	
	< 0.25	218.8 Bb	280.2 Ba	23.6 Cb	72.4 Aa	11.8 Aa	2.5 Bb	19.6 Aa	1.2 Cb	
M. caesalpiniifolia	<i>u</i> > 2	207.9 Bb	279.7 Ba	99.9 Aa	58.0 Ab	5.5 Ba	3.0 Ab	10.4 Ba	5.4 Ab	
	2.0-25	300.4 Aa	300.8 Aa	77.5 Ba	37.2 Bb	8.1 Aa	3.0 Ab	4.6 Cb	5.3 Aa	
	< 0.25	219.3 Bb	293.5 ABa	40.3 Ca	25.2 Cb	5.5 Ba	2.0 Bb	14.7 Aa	2.0 Bb	
Pasture	> 2	191.0 Ab	301.2 Ba	52.0 Aa	50.5 Ba	1.4 Cb	2.0 Aa	4.4 Aa	2.6 Ba	
	2.0-25	222.5 Ab	393.9 Aa	24.0 Ba	16.4 Cb	2.4 Ba	2.0 Aa	2.0 Ba	2.1 Ba	
	< 0.25	144.7 Bb	210.5 Ca	22.0 Bb	69.2 Aa	4.0 Aa	1.0 Bb	4,5 Aa	3.9 Aa	
Capoeira	> 2	214.9 Ab	351.6 Aa	72.6 Aa	35.7 Bb	4.3 Ba	3.0 Ab	11.3 Aa	8.1 Ab	
	2.0-25	233.3 Aa	263.5 Ba	29.1 Bb	39.2 Ba	6.6 Aa	2.8 Ab	1.9 Bb	8.5 Aa	
	< 0.25	186,0 Bb	274.0 Ba	33.4 Bb	61.4 Aa	2.0 Cb	2.8 Aa	2,2 Bb	2.8 Ba	

⁽¹⁾ For total P forms, means followed by the same uppercase letters in the column (comparing size fraction in each cover and time) and means followed by the same lowercase letters in the row (comparing time in the size fraction of each cover) are not significantly different by Tukey's test at 5 %; ⁽²⁾ For Labile P forms, means followed by the same uppercase letters in the column (comparing size fraction in each cover and time) and averages followed by the same lowercase letters in the row (comparing time in the size fraction of each cover) are not significantly different by Tukey's test at 5 %.

> 2.0 mm aggregates and an average decrease of 15 % in the total Po in the 2.0-0.25 mm aggregates, as opposed to the 90 % average increase in the < 0.25mm aggregates. The soils under leguminous trees showed clear differences in the contents of total Po affected by the incubation times in all soil size fractions. Soil under Acacia auriculiformis had increased Po levels in all size fractions, especially in the < 0.25 mm aggregates. In the soil under Mimosa caesalpiniifolia, however, Po was reduced in all size fractions, particularly in the 2.0-0.25 mm aggregates. In the soil under pasture, the effect of incubation time varied with the size fractions as follows: in the > 2.0mm aggregates, no significant change in Po levels was observed: and in the 2.0-0.25 and < 0.25 mm aggregates, the Po levels were reduced and increased,

respectively. Moreover, in the soil under capoeira, Po was reduced in the > 2.0 mm aggregates but increased in the other size fractions.

In general, the average content of labile Pi was higher in the < 0.25 mm aggregates at T0 (Table 5). After T1, however, the contents of labile Pi were highest in the > 2.0 mm aggregates. No significant variation in labile Pi after different incubation times was observed in this size fraction, except for the soil under *Acacia auriculiformis*. However, Pi decreased in the smaller size fraction with incubation time. In contrast, the average content of labile Po was lowest in the 2.0-0.25 mm aggregates at T0. In general, there was a significant average reduction in labile Po with incubation in the > 2.0 (-50 %) and < 0.25 mm (-76 %) aggregates, but a 35 % increase in labile Po occurred in the 2.0-0.25 mm aggregates. Generally, in soils under leguminous trees, incubation reduced the labile Po contents. However, a decrease was only observed in the > 2.0 mm aggregates under capoeira, which contrasted the increase in the other size fractions. Pasture cover did not show significant changes in labile Po in all size fractions.

The ratio of total Po fraction relative to total extracted P (Po/PT_{extracted}) as well as the ratio of labile Po fraction relative to the total extracted labile P (PoL/ $PT_{extracted}L$) in soil size fractions varied with vegetation covers and incubation times (Table 6). The highest average ratio of total Po occurred in the > 2.0 and < 0.25 mm aggregates at T0 and T1, respectively. The 2.0-0.25 and < 0.25 mm aggregates had the lowest average ratio of labile Po before and after soil incubation, respectively.

The variation in total Pi contents with incubation times showed the effectiveness of the Bowman method (Bowman, 1989) to extract previously non-accessible Pi compounds. In contrast, a decrease in total Po would demonstrate its possible mineralization and consequent release of Pi in the soil solution, and an increase in total Po with incubation time would also show that stable compounds of this fraction became accessible to the extraction method. The recovery rate (Pi + Po) of the Bowman method in relation to total soil P by digestion may be influenced by the experimental conditions (incubation times, temperature and humidity), soil and vegetation cover

types, and the rates may vary between 50 and 87 %(Cunha et al., 2007; Nunes, 2011; Zaia et al., 2012). Similar reasoning can be applied to the bicarbonateextracted labile P in which the Pi fraction is considered as the amount of P available to the plant, and the Po fraction is considered as the amount of potentially mineralizable P (Bowman & Cole, 1978). A decrease in labile Pi following soil incubation would indicate that the minerals in soil colloids stabilized this fraction, transforming labile P to stable P. This process was evident by the decrease of the average Pi content in the 2.0-0.25 and < 0.25 mm aggregates (Table 6). In soils where no P was applied, Goncalves et al. (1989) observed a small, non-systematic variation of the recovered P extracted by resins, Bray-1 and Mehlich according to incubation. In a study by Araújo et al. (2004), positive or negative variations of labile Pi and Po were found in fractions extracted by bicarbonate from soils incubated for 120 d. The authors attributed the effect on buffering or on the increase of the labile fraction to the moderately labile P fractions (NaOH-P). Using the ignition method, Acquave (1963) reported that mineralization or increased Po is significantly influenced by soil physicochemical properties, soil fertilization, temperature and humidity, and incubation time.

In this study, the use of the incubation-extraction technique determined the basal Po mineralization. In contrast to nitrogen (N) mineralization, however, the net mineralization of Po cannot be measured by the

Cover Siz	ize fraction	Pi/Ptotal		Po/P	total	PiL/PLtotal		PoL/total	
	ize fraction	TO	T1	TO	T1	TO	T1	TO	T1
	mm				<i>q</i>	%			
A. auriculiformis	> 2	$86.4 Aa^{(1)}$	83.7 Ab	13.6 Bb	16.3 Ba	24.5 Bb(2)	68.4 Aa	75.6 Aa	31.6 Bb
	2.0-25	80.1 Ba	80.6 Ba	19.9 Aa	19.4 Aa	26.5 Bb	33.7 Ba	73.9 Aa	66.5 Ab
	< 0.25	90.2 Aa	79.5 Bb	9.6 Bb	20.5 Aa	37.4 Ab	67.8 Aa	62.6 Ba	32.2 Bb
M. caesalpiniifolia	ia > 2	67.5 Cb	82.8 Ca	32.5 Aa	17.2 Ab	34.6 Ba	35.8 Ba	65.4 Ba	64.2 Aa
	2.0-25	79.5 Bb	89.0 Ba	20.5Ba	11.0 Bb	63.8 Aa	36.4 Bb	36.2 Cb	63.6 Aa
	< 0.25	84.5 Ab	92.1 Aa	15.5 Ca	7.9 Cb	$27.3 \ \mathrm{Cb}$	50.8 Aa	72.7 Aa	49.2 Bb
Pasture	> 2	78.5 Bb	85.6 Ba	21.5 Aa	14.5 Bb	23.9 Cb	43.5 Ba	76.1 Aa	56.5 Bb
	2.0-25	89.9 Ab	96.0 Aa	10.1 Ba	4.0 Cb	55.3 Aa	49.2 Ab	44.7 Cb	50.8 Ca
	< 0.25	86.8 Aa	75.3 Cb	13.2 Bb	24.7 Aa	46.9 Ba	20.7 Cb	53.2 Bb	79.3 Aa
Capoeira	> 2	74.7 Cb	90.8 Aa	25.3 Aa	9.2 Cb	27.6 Ca	26.8 Ba	72.4 Aa	73.2 Aa
	2.0-25	88.8 Aa	87.0 Ba	11.2 Ca	13.0 Ba	77.4 Aa	24.3 Bb	22.6 Cb	75.7 Aa
	< 0.25	84.8 Ba	81.7 Cb	15.2 Bb	18.3 Aa	47.2 Ba	49.3 Aa	52.8 Ba	50.7 Ba

Table 6. Percentage distribution of total inorganic (Pi), organic (Po), labile inorganic (PiL) and organic (PoL) phosphorus contents of soil size fractions under leguminous trees, pasture and capoeira at time zero (T0) and after 90 d of incubation (T1)

⁽¹⁾ For total P forms, means followed by the same uppercase letters in the column (comparing size fraction in each cover and time) and means followed by the same lowercase letters in the row (comparing time in the size fraction of each cover) are not significantly different by Tukey's test at 5 %; ⁽²⁾ For Labile P forms, means followed by the same uppercase letters in the column (comparing size fraction in each cover and time) and averages followed by the same lowercase letters in the row (comparing time in the size fraction of each cover) are not significantly different by Tukey's test at 5 %.

incubation-extraction technique. Once Pi is released from Po, it does not accumulate in the soil solution because it is adsorbed by the mineral phase of the soil (Bünemann et al., 2007). Furthermore, small changes in Po net values are masked by the large amount of total Po present in the soil.

Consequently, mineralization of basal Po can be defined as the mineralization of soil organic matter in soil that with no recent input of fresh organic matter (Ohel et al., 2004), thus representing the potential basal Pi fraction released from soil Po to soil solution (Ohel et al., 2001). Simultaneously, remineralization can occur during the incubation period due to the recycling of microbial P as a result of death and predation of microorganisms and indicates mineralization of the newly synthesized Po (Randhawa et al., 2005).

The distribution of P fractions (Pi and Po) in soil size fractions revealed the distinctive ability of the evaluated vegetation covers in recycling soil P. The average content of labile Pi in all size fractions was higher than the content of the P fraction in the soil as a whole, indicating the variation in P availability within the soil size fraction. In an analogous situation for the labile Po, its distribution and high contents in the soil size fractions indicated differences in the ability of vegetation covers to accumulate Po in the soil. Similar to OC, accumulated Po in the smaller size fractions of soil would be physically protected from the action of soil microorganisms. However, this does not indicate that the predominant forms of Po in the smaller soil size fractions are mostly recalcitrant as typically occurs with forms of OC (Six et al., 2002; Cadisch et al., 2006). It should also be considered that many phosphate groups that make up the P organic forms in the soil influence its mineralization rate. The diester forms of soil Po are more labile and easily mineralized than monoesters, and diester Po compounds are important sources of P to plants, thereby playing an important role in the P cycle ecosystem (Makarov et al., 2002). Therefore, future studies on the distribution of Po compounds and its mineralization potential in different soil size fractions could elucidate the influence of different types of land use on the accumulation of Po in soil and the P availability to plants in highly weathered soils.

CONCLUSION

The distribution of P fractions (Pi and Po) in the soil size fractions revealed the distinctive ability of vegetation covers to recycle soil P. As a result, the mineralization potential of Po in the soil varied with the soil size fraction and the vegetation cover. Because Po was the predominant form in the total labile P composition, P availability to plants was closely related to the mineralization of labile Po.

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LITERATURE CITED

- ACQUAYE, D.K. Some significance of soil organic phosphorus mineralization in the phosphorus nutrition of cocoa in Ghana. Plant Soil, 19:65-80, 1963.
- ANDERSON, J.D. & INGRAM, J.S.I. Tropical soil biology and fertility: A handbook of methods. 2.ed. Wallingford, UK CAB International, 1996. 171p.
- ARAÚJO, M.S.B.; SCHAEFER, C.E.G.R. & SAMPAIO, E.V.S.B. Frações de fósforo após extrações sucessivas com resina e incubação, em Latossolos e Luvissolos do semi-árido de Pernambuco. R. Bras. Ci. Solo, 27:985-1002, 2004.
- BATAGLIA, O.C.; FURLANI, A.M.C.; TEIXEIRA, J.P.F.; FURLANI, P.R. & GALLO, J.R. Métodos de análise química de plantas. Campinas, Instituto Agronômico de Campinas, 1983. 48p. (Boletim Técnico, 78)
- BOWMAN, R.A. A sequential extraction procedure with concentrated sulfuric acid and diluted base for soil organic phosphorus. Soil Sci. Soc. Am. J., 53:326-366, 1989.
- BOWMAN, R.A. & COLE, C.V. Transformation of organic phosphorus substrates in soil as evaluated by $NaHCO_3$ extraction. Soil Sci., 125:95-101, 1978.
- BRONICK, C.J. & LAL, R. Soil structure and management: A review. Geoderma, 124:3-22, 2005.
- BÜNEMANN, E.K.; MARSCHNER, P.; McNEILL, A.M. & McLAUGHLIN, M.J. Measuring rates of gross and net mineralization of organic phosphorus in soils. Soil Biol. Biochem., 39:900-913, 2007.
- CADISCH, G.; MUTUO, P.; MERCADO, A.; HAIRIAH, K.; NYAMUGAFATA, P.; BOYE, A. & ALBRECHT, A.
 Organic matter management in tropical agroforestry systems: Soil quality, soil C storage and soil atmosphere gas exchange. In: GAMA-RODRIGUES, A.C.; BARROS, N.F.; GAMA-RODRIGUES, E.F.; FREITAS, M.S.M.; VIANA, A.P.; JASMIN, J.M.; MARCIANO, C.R. & CARNEIRO, J.G.A., eds. Sistemas Agroflorestais: Bases científicas para o desenvolvimento sustentável. Campos dos Goytacazes, UENF, 2006. p.275-290.
- CUNHA, G.M.; GAMA-RODRIGUES, A.C.; COSTA, G.S. & VELLOSO, A.C.X. Fósforo orgânico em solos sob florestas montanas, pastagens e eucalipto no Norte Fluminense. R. Bras. Ci. Solo, 31:667-671, 2007.

- EMPRESA BRASILEIRA DE PESQUISA AGROPECUÁRIA -EMBRAPA. Centro Nacional de Pesquisa do Solo. Manual de métodos de análises de solo. Centro Nacional de Pesquisa de Solos (CNPS). 2.ed. Rio de Janeiro, 1999. 212p.
- GAMA-RODRIGUES, E.F.; NAIR, P.K.R.; NAIR, V.D.; GAMA-RODRIGUES, A.C.; BALIGAR, V.C. & MACHADO, R.C.R. Carbon storage in soil size fractions under two cacao agroforestry systems in Bahia, Brazil. Environ. Manage., 45:274-283, 2010.
- GATIBONI, L.C. Disponibilidade de formas de fósforo do solo às plantas. Santa Maria, Universidade Federal de Santa Maria, 2003. 247p. (Tese de Doutorado)
- GEORGE, T.S.; TURNER, B.L.; GREGORY, P.J.; CADE-MENUM, B.J. & RICHARDSON, A.E. Depletion of organic phosphorus from Oxisols in relation to phosphatase activities in the rhizosphere. Eur. J. Soil Sci., 57:47-57, 2006.
- GONÇALVES, J.L.M.; NOVAIS, R.F.; BARROS, N.F.; NEVES, J.C.L. & RIBEIRO, A.C. Cinética de transformação de P-lábil em não-lábil, em solos de cerrado. R. Bras. Ci. Solo, 13:13-24, 1989.
- GUERRA, J.G.M.; ALMEIDA, D.J.; SANTOS, G.A. & FERNANDES, M.S. Conteúdo de fósforo orgânico em amostras de solos. Pesq. Agropec. Bras., 31:291-299, 1996.
- MAKAROV, M.I.; HAUMAIER, L. & ZECH, W. Nature of soil organic phosphorus: An assessment of peak assignments in the diester region of 31P-NMR spectra. Soil Biol. Biochem., 34:1467-1477, 2002.
- McGILL, W.B. & COLE, C.V. Comparative aspects of cycling of organic C, N, S and P through soil organic matter. Geoderma, 26:267-286, 1981.
- MURPHEY, J. & RILEY, J.P. A modified single solution method for the determination of phosphate in natural waters. Anal. Chem. Acta, 27:31-36, 1962.
- NIZIGUEBA, G. & BÜNEMANN, E.K. Organic phosphorus dynamics in tropical agroecosystems. In: TURNER, B.L.; FROSSARD, E. & BALDWIN, D.S., eds. Organic phosphorus in the environment. Wallingford, CAB International, 2005. p.243-268.
- NOVAIS, R.F. & SMYTH, T.J. Fósforo em solo e planta em condições tropicais. Viçosa, MG, Universidade Federal de Viçosa, 1999. 399p.

- NUNES, D.A.D. Mineralização de fósforo orgânico em solos sob leguminosas florestais, floresta secundária e pastagem. Campos dos Goytacazes, Universidade Estadual do Norte Fluminense, 2011. 59p. (Dissertação de Mestrado)
- OHEL, F.; FROSSARD, E.; FLIESSBACH, A.; DUBOIS, D. & OBERSON, A. Basal organic phosphorus mineralization in soils under different farming systems. Soil Biol. Biochem., 36:667-675, 2004.
- OHEL, F.; OBERSON, A.; SINAJ, S. & FROSSARD, E. Organic phosphorus mineralization studies using isotopic dilution techniques. Soil Sci. Soc. Am. J., 65:780-787, 2001.
- RANDHAWA, P.S.; CONDRON, L.M.; DI, H.J.; SINAJ, S. & MCLEANAGHEN, R.D. Effect of green manure addition on soil organic phosphorus mineralization. Nutr. Cycling Agroescosyst., 73:181-189, 2005.
- RITA, J.C.O.; GAMA-RODRIGUES, E.F.; GAMA-RODRIGUES, A.C.; POLIDORO, J.C.; MACHADO, R.C.R. & BALIGAR, V.C. C and N content in density fractions of whole soil and soil size fraction under cacao agroforestry systems and natural forest in Bahia, Brazil. Environ. Manage., 48:134-141, 2011.
- ROLDÁN, A.; SALINAS-GARCÍA, J.R.; ALGUACIL, M.M. & CARAVACA, F. Changes in soil enzyme activity, aggregation and C sequestration mediated by conservation tillage practices and water regime in a maize field. Appl. Soil Ecol., 30:11-20, 2005.
- SIX, J.; CONANT, R.T.; PAUL, E.A. & PAUSTIAN, K. Stabilization mechanisms of soil organic matter: Implications for C-saturation of soils. Plant Soil, 241:155-176, 2002.
- ZAIA, F.C.; GAMA-RODRIGUES, A.C. & GAMA-RODRIGUES, E.F. Formas de fósforo no solo sob leguminosas florestais, floresta secundária e pastagem no Norte Fluminense. R. Bras. Ci. Solo, 32:1191-1197, 2008.
- ZAIA, F.C.; GAMA-RODRIGUES, A.C.; GAMA-RODRIGUES, E.F.; MOÇO, M.K.S.; FONTES, A.G.; MACHADO, R.C.R. & BALIGAR, V.C. Carbon, nitrogen, organic phosphorus, microbial biomass and N mineralization in soils under cacao agroforestry systems in Bahia, Brazil. Agrofor. Syst., 86:197-212, 2012.