The Effects of Different Harvest Intensities on the Distribution of Soil Phosphorus and Nutrient Stocks in a Brazilian Oxisol

Marcella Leite de Campos Menegale

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Reading Committee:
Robert B. Harrison, Chair
Scott Holub
Irae Amaral Guerrini

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Abstract

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Chair of the Supervisory Committee:
Dr. Robert B. Harrison
School of Environmental and Forest Sciences

Forest soils form an important reservoir in ecosystem nutrient and carbon budgets, which are crucial for sustained productivity of forests. Nutrient fluxes and transformations in forest soils are a result of a complex interchange between the atmosphere, plants, and soil. Consequently, soils are a critical source of plant nutrition, and integral to the recovery of ecosystems following natural or human disturbances. As essential plant nutrients, the concentrations of plant available nitrogen (N), phosphorous (P), and sulfur (S) are crucial controls on the net primary productivity of most forest sites. As the plants uptake these nutrients, their concentrations within the soil are reduced until they are replaced either by nutrient recycling or fresh inputs. Highly weathered tropical soils are characteristically deficient in essential plant nutrients. This is especially true of the macronutrients essential for plant growth and development. Organic matter (OM) plays a fundamental role in soil fertility, contributing to cation exchange capacity (CEC) and as a source of nutrients, while also stabilizing soil aggregates and increasing water holding capacity. Soil management practices can sustain adequate levels of OM while providing sufficient quantities of plant available P. The need for balancing high timber productivity while maintaining soil nutrient concentrations has led to the development of sustainable silvicultural practices. Retaining forest harvest residues on the soil surface can help to return nutrients to the soil, as well as contributing substantial quantities of OM, thus aiding in a more sustainable production system. This research project identified the different forms of P in the soil (labile P, moderately labile P and non-labile P), as well as their concentrations at depth (to approximately two meters) in an Oxisol planted with *Eucalyptus* in Brazil under different timber harvest intensities. The harvest regimes considered were (i) conventional stem-only harvest (all forest residues were maintained on the soil); (ii) whole-tree harvest (only litter was maintained on the soil – all slash, stemwood and bark were removed) and (iii) whole-tree harvest + litter layer removal. In addition, this research seeks to establish the relationship between the different forms of P and carbon (C) and nitrogen (N) in the soil. A reduction in all nutrient stocks was observed in the 0-20 cm layer for all treatments. For N, this reduction was 20% higher when harvest residues were removed from the soil, and 40% higher when no N fertilizer was applied. The maintenance of harvest residues on the soil reduces the loss of N, P, and S due to harvest by 120%, 50%, and 40%, respectively. Our study has also shown that organic and inorganic P forms in the soil can act as source or sink to the soil solution, depending on the mineralogical composition of the soil, fertilization and harvest management applied. From 2004 to 2016, it was observed a reduction in the relative contribution of non-labile P in the treatments for which all harvest residues were maintained. However, this fraction of P still accounts for more than 60% of total P present in the soil, followed by labile and moderately labile forms of

the nutrient. The highest stocks of total P were observed at 0-10cm. At the end of the first crop rotation of the experiment (2012), the treatments which all harvest residues were maintained on the soil without fertilization (ReM-F), all harvest residues were removed from the soil plus fertilization (ReR+F) and harvest residues were partially removed from the soil plus fertilization (ACR+F) presented the highest values for the nutrient.

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DEDICATION

This work is dedicated to my family.

CHAPTER 1.   INTRODUCTION

In this chapter we consider the importance of forest soils as reservoirs of carbon (C), nitrogen (N), sulphur (S), and phosphorus (P) in the soil-plant-atmosphere system. As plant macronutrients, N and P are needed in high quantities in order to maintain the productivity of forest ecosystems. The sustainable management of forest ecosystems balances the demands for high timber yields with environmental preservation. For example, maintaining levels of organic matter (OM) within the soil through good soil management practices can increase timber yields while reducing the need for synthetic fertilizer applications. Retaining forest harvest residues is a practice that raises the likelihood of medium to long-term soil fertility maintenance and productivity of wood in species such as *Eucalyptus*. The retention of harvest residues on site is a practice that aims to promote nutrient retention by maintaining the physicochemical properties of the soil. Additionally, the protective surface layer formed by the presence of harvest residues can reduce nutrient leaching through the soil profile and also helps to avoid disruption of the colloidal particles and soil losses due to erosion and raindrop impact. An improved knowledge of the abundance and diversity of soil P species and of the soil and land use factors governing the pathway to current soil P conditions will better inform soil management for timber production, water quality, and soil quality. The effects of forest management on soil C, P, and N are important tools for understanding the spatial and temporal variability in soil fertility and how it relates to tree growth. Soil management must maintain good physicochemical properties to continue to address the balance between protection of the environment and sustainable use of P in forest production.

1.1. The process of formation of Phosphorus in the soil

Phosphorus (P) is an essential nutrient for all life forms and is primarily conserved in sediments and soils (Brady and Weil, 2013). It plays a vital role in determining the productivity of terrestrial and aquatic ecosystems. It is an essential constituent of tissues and cells because it is required for the formation of nucleic acids and energy-carrying molecules, such as adenosine tri-phosphate (Taiz and Zeiger, 2010).

In soils, P cycling is a complex phenomenon that is strongly influenced by the nature of the inorganic and organic solid phases present, the forms and extent of biological activity occurring, as well as the chemistry of the soil solution and many other environmental factors such as temperature and soil moisture content (Parfitt, 1978).

The development of sustainable land management practices for all agro-ecosystems requires a fundamental understanding of the chemical, biological and physical processes in the soil that affect the availability of P to terrestrial plants (Sims and Sharpley, 2005). Walker and Syers (1976) stated that P is perhaps the key element in pedogenesis because of its great ecological significance. This is the one major element in soil organic matter (SOM), which must be supplied almost entirely by the parent material of unfertilized soils because of low atmospheric returns.

P in most soils originates from the weathering of apatite (Condron and Tiessen, 2005). As soils weather, concentrations of aluminum (Al) and iron (Fe) phosphates and organic forms of P increase. Soil solution P concentrations generally range from <0.01 mg P L^{-1} in infertile soils to 1 mg P L^{-1} in well-fertilized soils, and can be as high as 7 to 8 mg P L^{-1} in soils recently amended with organic residues or fertilizers (Sims and Sharpley, 2005).

Total P concentrations in soil vary widely from 100-3000 mg kg^{-1} (Harrison, 1987). Soil P pools include both organic and inorganic P species (Henriquez, 2002). Typically, between 30% and 65% of the soil pool is organic P, although soils high in OM can contain up to 90% organic P (Harrison, 1987). P continuously cycles between organic and inorganic forms within the soil. The inorganic P group can be separated into two parts: P coming from primary minerals, and adsorbed P (Condron and Tiessen, 2005).

Highly weathered soils adsorb large amounts of P and limit plant growth as a result (Vitousek et al., 2009). The degree of interaction between phosphate and soil minerals can be measured via the maximum phosphorus adsorption capacity (Pmax) or remaining P (Prem) (Olsen and Watanabe, 1957).

Biogeochemical theory predicts that soil total P declines as pedogenesis progresses (Sharpley, 1985). At the same time, there are changes in soil P chemistry, with a decline in primary mineral P and an accumulation of occluded and organic P as soil ages (Walker and Syers, 1976). Soil bacteria and fungi constitute the concentration of P in soil organisms. Soil microorganisms can mineralize organic P as well as immobilize phosphate from the soil solution (Sharpley, 1985). Similarly, plants absorb phosphate from the soil solution of organic material into the soil from litterfall, root decay, crop residues, animal excreta or dead soil organisms (Walker and Syers, 1976). These provide the starting point for soil organic P formation.

1.2. Occurrence of Phosphorus in tropical ecosystems

It is well documented in the literature that P is an important limiting nutrient in forest production in tropical ecosystems (Tamminen et al., 2012; Darch et al., 2016;

Rocha et al., 2016; Rodrigues et al., 2016; Mao et al., 2017). Limited availability of P fertilizers coupled with high ecosystem demands for P is one of the major global environmental challenges of the 21st century (Cordell and Neset, 2014). Hence, the turnover of organic P is widely considered fundamental in the maintenance of P supply in tropical forests (Condron and Tiessen, 2005).

The dynamics of P in the soil are associated with environmental factors that control the activity of the microorganisms which immobilize or release the orthophosphate ions, as well as their physicochemical and mineralogy properties. Hence, organic P transformations in soil are important in determining the overall biological availability of P, which in turn influences ecosystem productivity.

Low solubility of phosphate (PO_4^{3-}) makes P a relatively immobile nutrient. However, it is still subject to leaching and runoff losses in both its organic and inorganic forms (Rodrigues et al., 2016). Because of this mobility, the productivity of forests growing on old soils and soils in high leaching environments (i.e. wet tropics) is limited with respect to P (Gonçalves et al., 2002; Laclau et al., 2010). Forests growing on young soils or soils in dry environments (i.e. New Zealand) are less likely to be growth limited with respect to P (McDowell and Stewart, 2006; Brandtberg et al., 2010).

As a result of the influence of soil mineralogy on P solubility, different soil types show different behaviors with respect to P transformations. No correlation between total P, solution P, or P availability (Chapuis Lardy et al., 2002).

Young and moderately weathered soils such as Vertisols, Chernosols, and Neosols, contain P in the form of primary minerals, where it is mostly found in its organic or mineral forms, or weakly adsorbed to secondary minerals (Brady and Weil, 2013).

Inorganic forms linked to mineral fractions with high energy and to physically and chemically stable organic forms dominate highly weathered soils, such as Oxisols (Condron et al., 1985). These compounds are classified as labile and non-labile phosphates based on their higher or lower degree of stability (Condron et al., 1985). Labile P species are able to quickly replenish in the soil solution when absorbed by plants or microorganisms (Condron et al., 1985). Therefore, the concentrations of these most labile fractions are dependent upon the extent to which P containing parent material has weathered, soil mineralogy, soil texture, and OM content (Sharpley, 1985).

Physicochemical characteristics, biological activity and dominant vegetation cover are the primary drivers of weathering rates and soil properties that support high concentrations of labile P forms in the soil profile (Walker and Syers, 1976; Cross e Schlesinger, 1995). P released by weathering from P-bearing minerals, from P additions in plant residues, and from fertilizers is primarily retained in the clay fraction (Harrison, 1987). Thus, the efficient management of inorganic and organic P is essential for maintaining the environmental and economic sustainability of the timber production chain.

1.3. Addition of harvest residues on the soil and how it affects soil fertility

Globally, forest soils are among the most important reservoirs of C, P, and N (Vanguelova et al, 2010). In order to create a more sustainable ecosystem (meaning high productivity and environmental preservation), it is essential that the levels of OM in the soil be maintained through good soil management practices.

In Brazil, soil nutrient depletion is the results of high weathering rates and erosion, both resulting in low SOM concentrations (Zinn et al., 2002). In order to reduce the need for synthetic fertilizers and decrease erosion rates, it is extremely important to enhance nutrient accumulation through build up OM in the soil (Gonçalves et al., 2002). This is increasingly achieved by retaining forest harvest residues on the soil surface following harvest. The residual material deposited on the soil surface is able to return nutrients back to the soil and help with soil nutrient pool maintenance, increasing the availability of nutrients for plant uptake. In order to compensate for the lack of nutrients available in the soil, the application of synthetic fertilizers must be considered. The input of nutrients through synthetic fertilizers or OM additions help to "build-up" soil nutrient pools (Corbeels et al., 2005).

Highly productive soils contain large quantities of plant available nutrients and OM (Rodrigues et al., 2016). The presence of timber harvest residues on the soil surface benefits the soil-plant system by improving the physicochemical characteristics of the soil (i.e. uptake of nutrients, SOM content, soil structure, pH), presenting, consequently, a series of benefits for plants cultivated in the next rotations (Rocha et al., 2016). Soil quality improves with increased cation exchange capacity (CEC), aggregation, water-holding capacity, infiltration, microbial diversity and pH-buffering capacity (Lal, 2005; Xu et al., 2012). Hence, the presence of a "protector layer" on the soil surface helps to reduce nutrient leaching to the water table. In addition to that, it contributes to the reduction of incidence of invasive plants in the area (Rocha, 2014).

Essential nutrients for plant growth are deposited back to the soil during the decomposition of OM. This helps with nutrient cycling and with the improvement of nutrient pools and OM content in the soil (Corbeels et al., 2005). The plants cultivated in the area will then absorb these nutrients. Furthermore, the loss of nutrients associated with the removal of harvest residues from the soil may also result in site acidification and other physicochemical problems (Rocha, 2014). As theoretically stated, site acidification is a result of the removal of cations (Ca, Mg, K and Na) normally presented in the aerial parts of trees, especially foliage and branches (Maliondo, 1988).

Evidence points to a reduced need for the application of high doses of fertilizers in the soil when it already has considerable amounts of nutrients available for use by subsequent cultivations (Huang et al., 2013; Hubner, 2015). This consequently decreases the total energetic balance of the wood production process. In *Eucalyptus* plantations in Brazil, in trees of 7 years' age, the bark represents approximately 23% of the timber harvest residues. This material contains approximately 12, 41, 42 and 28% of N, P, K and Ca, respectively (Gonçalves et al., 2002). The production process of fertilizers is highly energetic, releasing large amounts of C to the atmosphere, thus generating a negative impact on the sustainability of timber production.

Additionally, the removal of residues from the soil surface can decrease site productivity (Rocha et al., 2016b). Besides releasing nutrients back to the soil, harvest residues provide a protective layer to the surface. A layer of harvest residues decreases the erosive impact of rainfall, avoiding disaggregation of soil particles, enhancing soil structure, and reducing leaching rates (Gonçalves et al., 2002; Huang et al., 2013; Rodrigues et al., 2016; Rocha et al., 2016). Preserving the soil structure is essential for the normal flux of water through the macropores and micropores in the soil, as well as good soil aeration. Moreover, most importantly, the deposition of harvest residues on the soil helps to keep adequate levels of soil moisture and temperature, and influences OM decomposition rate and the presence of microorganisms (Gonçalves et al., 2002).

While a conventional timber harvest generally removes only merchantable bole wood, harvesting biomass for energy production removes all forms of woody biomass (i.e. woody debris, branches and stumps) resulting in a greater loss of biomass and nutrients as well as more severe habitat alteration (Peng et al., 2008).

Comparing the production systems in Brazil and the United States, Zinn et al. (2002) found that such variations in macronutrient contents are a result of the weathering process and SOM decomposition rate (influenced by climatic and environmental factors), regardless of the type and cycle of the species cultivated in each region. In Brazil, the low availability of nutrients during the initial growth of *Eucalyptus* can negatively influence the development of plants, with a consequent impact on the final production of timber (Rocha, 2014). In this sense, the use of harvest residues as soil coverage helps to maintain the soil moisture content, resulting in the decrease of the SOM decomposition rate, influencing the quantity of nutrients stored in the soil and available to plants (Gonçalves et al., 2002).

In *Eucalyptus*, the highest concentration of N and K (potassium) occur in the leaves, bark, and branches respectively. For P, calcium (Ca), and magnesium (Mg) bark has the highest concentrations followed by leaves and branches respectively (Laclau et al., 2000). Gonçalves et al. (2002) found that the bark represents about 12 to 14% of bole total mass, and presents high concentrations of Ca and P, at *Eucalyptus* plantations in

Brazil. In addition, in a 7-year old *Eucalyptus grandis* plantation, the bark represents 23% of the total harvest residues, and presents 12%, 41%, 42% and 28% of N, P, K and Ca, respectively (Nambiar and Brown, 1997).

Rocha (2014) found that *Eucalyptus* plantations managed under a minimum tillage system have the same timber productivity as those managed under an intensive cultivation system with the addition of fertilizers. The primary advantage of maintaining forest residues on the soil is to produce short-term gains in productivity and to reduce nutrient removal. The study also concluded that the removal of forest residues reduces the oxidizable organic C from the surface layer of the soil by 50%, and 75% of this reduction happens in the labile fractions.

Jones et al. (2011) analyzed the changes in C and N stocks in the forest floor within the mineral soil (to a depth of 0.3 m) between pre-harvest and mid-rotation (stand age 15 years) measurements in a *Pinus radiata* plantation forest in the central North Island, New Zealand. They utilized three harvest residue management treatments: residue plus forest floor removal (FF), residue removal (whole-tree harvesting; WT) and residue retention (stem-only harvesting; SO). The authors concluded that the removal of harvest residues from the sites is likely to reduce C and N pools in the forest floor. In addition, they suggest that forest floor should be retained in order to avoid adverse impacts on topsoil layer fertility.

In a long-term productivity experiment conducted in North Wales, UK, by Walmsley et al. (2009), the alterations in soil characteristics and their effects were tested using different harvest treatments (whole-tree harvest and bole-only harvest) on 23-year-old second rotation stands of Sitka spruce (*Picea sitchensis*). The authors found that the whole-tree harvest treatment is responsible for the depletion of three to four times greater quantities of N, P and K than the conventional bole-only harvest in the first rotation. They also observed higher soil moisture in the whole-tree harvest treatment, with no indication of a decrease in OM contents nor increase in soil acidity.

In a similar experiment, Devine et al. (2012) evaluated N leaching rates during 6 and 8-year post-harvest periods on two Douglas-fir sites (Fall River and Matlock Sites) in Western Washington/USA. The plots were divided into bole-only and whole-tree harvest treatments and presence/absence of 5 years of competing vegetation control. N leaching patterns among treatments at these two sites suggest that differences in soil N content and C:N ratio, post-harvest vegetation regrowth, and harvest residues influenced the amount of N leached. The presence of harvest residues can possibly return N to the soil and contribute to the increase of N pools in the soil.

Huang et al. (2015) studied the influence of vegetation cover in coastal wetland soils in China, and found that OM accumulation and decomposition play an important role in regulating potential P dynamics in the soils, now being considered as a key factor to increasing P availability to plants.

In agricultural systems, Sharpley et al. (1992), at various sites in Texas and Oklahoma, demonstrated the relationship between soil cover, soil erosion and, consequently, total P losses from natural runoff events. The study also concluded that, as soil cover increased, the percentage of the total P that was present in bioavailable forms also increased.

SOM content is closely related to the availability of P in the soil. As aforementioned, most of the P present in the soil is not readily available for plants.

Higher quantities of SOM resulting from the maintenance of harvest residues on the soil surface contribute to P availability for plants, since the addition of organic substances to the soil serves as a C source that enhances microbial biomass and phosphatase activity, helping with the transformation of occluded P into a readily available form suitable for plant uptake.

1.4. Forms of Phosphorus in the soil

The quantitative distribution of the different organic and inorganic forms of P in the soil is highly variable and dependent upon the intensity of factors that form the soil, with weathering being one of the main determinant factors for the quantity of each form of P present in the soil (Gatiboni et al., 2013). Walker and Syers (1976) established the relationship between the fractions of P and the process of soil formation. They concluded that, based on the exportation of P by living organisms in the soil and on the losses of the systems, the concentration of total-P in the soil decreases so that the more weathered the soil, the lower its total-P content.

Sequential chemical extractions illustrate the general strength of P adsorption to soil materials, as well non-labile P pools (Hall, 2008). Soil management and type of vegetation are factors that highly influence the forms of P in the soil, especially the organic forms, due to the fact they are directly related to biological activity in the soil (Conte et al., 2003).

The use of cultivation methods that advocate the maintenance of crop residue on the soil may lead to many benefits for plants. It may also lead to benefits for future cycles because it provides a higher OM content in the soil, which allows for nutrient cycling.

Moreover, this can lead to an increase in the availability of P to plants, due to better soil exploration, and its ability to mobilize the insoluble inorganic P (Prescott, 2010).

Using techniques which allow for the determination of the different fractions of P in the soil, various soil management systems may be evaluated, along with their contributions to the soil-plant system. With this, it is possible to gain a broader understanding of the transformations of P in the soil by quantifying not only the labile P, but also the fractions of lower lability between the labile P and total P in the soil (Gatiboni et al., 2013).

One widely-used method to determine the different fractions of P in the soil was described by Hedley et al. (1982). The main advantage of this method is the simultaneous identification of inorganic and organic forms of P, which can be grouped according to their lability in the soil. These fractions of P are arbitrary, due to the difficulty in defining and measuring labile and moderately labile P (Novais and Smith, 1999). The reason is that, depending on the absorption rate of P by the plant, the fraction of P can be classified as labile and non-labile.

The fractionation scheme of Hedley et al. (1982) focuses on P fractions that are dynamic in the soil-plant system and later variations, which included additional extractions to improve the extraction efficiency of the organic P fractions. Briefly, the Hedley et al. (1982) fractionation scheme extracts P with resin in bicarbonate, $NaHCO_3$, NaOH, sonication + NaOH and HCl followed by digestion of the residue with H_2SO_4 and H_2O_2. This fractionation method is a valuable tool to indicate the relative importance of biological processes to soil P in soils of different weathering (Cross and Schlesinger, 1995).

The techniques used to determine the different fractions of P in the soil, as with any other analytical method using extractable solutions, are subjected to errors. There are some limitations inherent to overmanipulation of a single soil sample, due to sequential extraction (Rodrigues et al., 2016). Similarly, P in the soil is classified in two groups, organic P and inorganic P, depending on the nature of the compost to which it is connected. However, the identification of different fractions of P between these two groups is complicated due to the large number of chemical reactions the nutrient can suffer as well as the resultant composts (Condron and Newman, 2011).

Gatiboni et al. (2013) observed that one limitation of Hedley's method for determining the organic and inorganic fractions of soil P is the lack of standardization. As P has a complex dynamic in the soil and is highly variable depending on the soil conditions, climate, management, etc., other authors have extensively modified the fractionation schemes in order to account for the differences. For this reason, it is very difficult to quantitatively compare the fractions of P obtained in different studies. However, since all fractionations and their variations use the sequential extraction as the starting point as well as strong extractors, it is possible to relatively compare the dynamic of labile and recalcitrant fractions among different studies.

The extractable P in the soil is used to determine the need for its supplementation through fertilization, and it represents a P availability index for plants (Cunha et al., 2007). However, this index does not consider the organic and inorganic fractions less available in the soil (Beck and Sanches, 1994). In forest ecosystems, the availability of P in the soil does not reflect the accumulation of this element in the biomass, nor the quantity of P cycled annually (Novais and Smith, 1999).

Fractionation techniques aim to identify the preferable fractions of P retained in the soil, their occurrence, and available forms to plants (Gatiboni et al., 2013). Thus, the study of fractions of P is extremely important in the management of fertilizer application on the soil, the consideration of soil type and of environmental conditions.

The main factors influencing the mineralization rate of harvest residues are the quantity and quality of the substrate (Paul and Clark, 1996). The release of P to the soil solution is controlled by the mineralization rate of SOM, and this is dependent upon the soil microbial activity (Barber, 1984). The release rate of nutrients from the decomposition of harvest residues depends on the form in which these nutrients are found in the decomposing material (Giacomini et al., 2003).

Most of the P fractions in the soil are susceptible to environmental conditions, and are characterized as mobile P pools (Lukkari et al., 2007). Studies suggest that only OM can explain the variation in the different forms of the nutrient in the soil (Huang et al., 2015; Rodrigues et al., 2016), which means, OM accumulation and decomposition are the key factors to increasing soluble forms of P in the soil. Therefore, the accumulation of harvest residues on the soil surface is a viable option for increasing SOM and thus explaining the variations of the different forms of P through the profile.

Zin et al. (2015) evaluated the chemical properties and the different inorganic forms of P in acid sulfate rice soils of Brunei. The authors observed that among the P fractions, the order of distribution for inorganic P forms in the soil was Fe-P > Al-P > reductant soluble-P > loosely bound P > Ca-P. Management practices that reduce the impact of Fe and Al in soil solutions might improve P availability in low-P soils.

A recent review examined how well sequential P fractionation reflected the impact of land use and management on soil P (Negassa and Leinweber, 2009). Numerous medium-term and long-term studies of sequentially-extracted P suggest that practices with little or no P input deplete both labile and moderately labile fractions in temperate, subtropical and tropical soils. In a similar experiment, Oliveira et al. (2015) observed that the conversion of native grasslands to pine plantations led to a decrease in OM quality and organic P depletion.

Hence, the distribution of the different forms of P in temperate soils is closely related to a number of abiotic and biotic related processes (Stutter et al., 2015). They seem to govern the accumulation of different soil P species and the variability in forms and concentrations within land use groups.

1.5. N:P and C:P ratio in tropical soils

Across tropical soils with similar chemical composition, correlations are frequently found between total P and C and N, indicating the overall P limitation on biological production. An accumulation of C and N is observed as soil develops, which results in increasing N:P and C:P ratios and strengthening P limitation through time (Chapuis Lardy et al., 2002).

The cycling of P and C in soils is tightly coupled because the bioavailability of C drives ecosystem processes that in turn affects sequestration and supply of soil P, with associated change in soil P species (Bradford et al., 2008; Kirkby et al., 2013). This C:P macronutrient coupling is especially important to understanding the increase in soil organic C storage for benefits to soil biodiversity, aggregate stability, and climate policy.

The coadsorption of phosphate and SOM on hydrous Fe and Al (hydr)oxides has been extensively studied in the field of soil chemistry (Sibanda and Young, 1986). Harter and Foster (1975) suggested that SOM and phosphate were adsorbed in soil by different mechanisms, and that phosphate may be adsorbed by direct bonding to OM in which the phosphate anion replaces the organic hydroxyls. Spohn and Kuzyakov (2013), on the other hand, observed that humic acid (HA) and fulvic acid (FA) are unable to compete effectively with phosphate. The contradictory results found in the literature stimulate further studies to clarify the competitive adsorption between SOM and phosphate on metal (hydr)oxides.

Cleveland et al. (2005) presents evidence that P limitation on the decomposer community limits OM turnover in most tropical forests. On two adjacent sites with different P concentrations, respiration response to the addition of C was highly dependent on P availability only in the P-poor soil. Thus, at low P availability, respiration/decomposition was restricted. The authors observed that such limitations might offset the expected faster respiration and the lower C storage of warmer tropical soils.

Tropical soils may conserve more C under increased OM inputs because P is limiting decomposition. Such P limitation will also affect other microbial processes such as the transformation of N. While most temperate ecosystems, which are limited by N supply, may store added mineral or atmospheric N for long periods, a P limited tropical forest could rapidly loose substantial amounts of N oxides upon N fertilization (Hall and Matson, 2003). The noticeable limitations of tropical soils not only affect plant productivity, but also decomposition and immobilization processes, and therefore the storage and cycling of C and N.

N inputs into tropical forest ecosystems are also coupled with the P cycle. For example, Binkley et al. (2003) observed double the rates of N_2 fixation associated with *Facaltaria moluccana* upon P additions to soils. Moreover, phosphate plus the resultant N (fixation) inputs increased tree growth, whereas N fertilization alone had no appreciable effect on rates of N_2 fixation. Kirkby et al. (2013) demonstrated that inorganic N and P availability were required to sequester plant residue C into stable soil pools, and in turn, that P was sequestered alongside C. Stutter et al. (2015) observed that organic P complexation increased in soils with greater C concentration. The authors demonstrated a positive relationship extending from arable soils in the area of ortho inorganic P dominance and low soil organic carbon (SOC) content through grassland to semi-natural soils with a dominance of organic P and large SOC contents.

CHAPTER 2. RESEARCH GOALS

Few studies have addressed the mechanisms involved in the interactions between different forms of soil phosphorous (P) (i.e. labile, moderately labile and non-labile P) and other nutrients within the soil (Kumaraswamy et al., 2014; Hubner et al., 2015; Rodrigues et al., 2016). It is also unclear how the addition of forest harvest residues and the efficient use of fertilizers might contribute to an increase in wood production.

Tropical soils are characteristically deficient in P due to their advanced weathering (Novais and Smith, 1999) and thus a limiting factor for plant growth. Chemical fertilizers derived from rock phosphate have played a key role in the expansion of agriculture and silviculture practices (Gonçalves et al., 2002; Laclau et al., 2003). While the exact size of global mineral P pools is disputed, it is a finite resource and requires conservative utilization to avoid depletion. Management practices that maintain soil organic matter (OM) concentrations and thus maintain plant available sources of P are contributing to a more sustainable use of forest resources (i.e. high timber productivity and lower environmental impacts) by limiting the need for synthetic fertilizer applications (Da Silva et al., 2013).

Retaining harvest residues on the soil surface has both economic and environmental benefits for the subsequent plantation (Jones et al., 2011). Harvest residues provide an inexpensive nutrient input by increasing soil organic matter (SOM concentrations and assistance in the cycling and bioavailability of other essential nutrients for plant growth (Turner and Lambert, 1986; Kumaraswamy et al., 2014; Rocha et al., 2016b).

Forest management can increase labile P stocks through the addition of harvest residues that effectively increase SOM (Maranguit et al., 2017). Increased SOM decreases adsorption and consequently increases nutrient availability to plants through mineralization. One should therefore consider the important role of SOM in the cycling of P between soil and plant pools. Additionally, understanding biogeochemical cycling of P throughout the soil profile, including its most common forms found at depth, can help us understand the process of biocycling of P (Rodrigues et al., 2016). More research about this topic could improve fertilizer use efficiency and limit negative environmental impacts associated with intensive forest management.

The focus of this research project is to identify the different forms of P (i.e. labile, moderately labile and non-labile P) within the soil profile, specifically in an Oxisol cultivated with *Eucalyptus* in Brazil. Soil P, C, S and N were measured to a depth of 2m under different timber harvest intensities in order to understand how forest management practices (i.e. harvest intensities and harvest residue retention) affect the availability of these four essential plant nutrients.

To address these objectives, this dissertation is divided into two studies presented in the following chapters:

Chapter 4 quantifies the labile, moderately labile, non-labile, and total fractions of P to a depth of 20cm in a Brazilian Oxisol cultivated with *Eucalyptus*.

Chapter 5 compares concentrations of P, S, C, and N in the soil to a depth of 2m in a *Eucalyptus* plantation following different harvest intensities. In this chapter, I specifically determine: 1) the stocks of available P, S, total N and oxidizable C in the soil;

and 2) estimates the distribution of P, S, C, and N in the soil and forest biomass pools as well as the inputs and outputs these nutrients for two crop rotations.

Finally, Chapter 6 synthesizes the information from Chapters 4 and 5 as to the effects of different harvest intensities on the distribution of soil P in a Brazilian Oxisol planted with *Eucalyptus* plantation, and the interactions of the different forms of P with other nutrients essential for plant growth. The challenges of sustainable forest production in highly weathered soils are discussed and alternative management practices that yield high quality wood products while balancing the efficient use of natural resources are proposed.

CHAPTER 3. MATERIAL AND METHODS

The goal of this chapter is to discuss points crucial to understanding the experiment and subsequent findings. First, a detailed description of the study area is presented, along with the management system which was implemented after the installation of the experiment. Second, the treatments applied in the experiment are described, depending on the presence or absence of harvest residues on the soil surface. Third, details are provided on the experimental design used in subsequent chapters. Finally, this chapter describes the methods of soil collection and analysis used in each chapter of this dissertation, as well as information related to the statistical analysis used in each section.

3.1. Study Site

The study was carried out at the Itatinga Forest Science Experimental Station of the University of São Paulo in Brazil (23°06’S lat and 48° 36’W long and 857 m above sea level) (Figure 3.1). The Köppen climate classification is humid subtropical, Cfa, with an average annual temperature of 19.4 C. In the coldest month (July), the temperatures average 15.6 C, and 22.3 C in the hottest month (January). The mean annual rainfall was 1319 mm, with 75% of the rainfall concentrated between October and March (Alvares et al., 2013).

The topography of the region is flat to undulating, and the soil is a very deep (>10m) Ferralsol (IUSS Working Group WRB, 2015; red-yellow Latosol - Brazilian Classification System, and Oxisols - USDA Soil Taxonomy) that developed on Cretaceous sandstone. The clay content ranges from 17% in the A1 horizon to 25% in deeper soil layers. The mineralogy was dominated by quartz, kaolinite and oxyhydroxides of Al and Fe with a low pH and small amounts of exchangeable cations (Table 3.1).

Table 3.1. Soil physical and chemical attributes of experimental site.

Depth	**Sandy**	**Silt**	**Clay**[1]	**pH**[2]	**CEC$_7$**[3]	**C**[4]	**N**	**P**[5]	**Exchangeable Cations**[4]			
									K	**Ca**	**Mg**	**Al**
cm	--------- g kg^{-1} ---------				mmol$_c$ kg^{-1}	g kg^{-1}		mg kg^{-1}	--------- mmol$_c$ kg^{-1} --------			
0 - 10	802	22	175	3.8	64.0	9.6	1.44	4	0.25	4.28	2.81	7.50
10 - 20	811	12	176	3.9	51.4	10.1	1.67	3	0.27	2.80	2.17	8.43
20 - 30	790	34	176	3.9	40.0	6.8	1.53	1	0.20	1.32	1.00	6.09
30 - 40	777	23	200	3.9	40.2	5.3	1.29	1	0.15	0.88	0.81	7.03
40 - 60	747	14	239	3.9	38.5	5.4	1.14	1	0.15	0.99	0.72	7.50
60 - 100	712	12	276	3.9	32.7	5.0	0.99	1	0.15	0.66	0.54	6.56
100 - 150	712	11	277	4.0	30.1	3.5	1.04	1	0.08	0.71	0.54	2.34
150 - 200	704	20	276	4.2	22.1	0.9	1.04	1	0.05	0.55	0.54	2.81

[1]Pipette method; [2] Determined in $CaCl_2$ 0.01 mol L^{-1} in soil/solution reason of 1:5; [3] Cation exchange capacity with soil at pH 7; [4]wet oxidation; [5]Extracted with exchange ion resin (Alvares et al., 2013).

The original vegetation of the site was Cerrado *stricto sensu* (Brazilian savannah) (Ribeiro et al., 1998). The site has been planted with *Eucalyptus* species since 1940. From 1940 to 1992, it was cropped with *Eucalyptus saligna* and managed by coppicing with clearcutting each 7 or 8 years. In 1992, the plantation was harvested and replanted with *Eucalyptus grandis*, which was harvested (clear-cutting) in 2004 when the study site was installed.

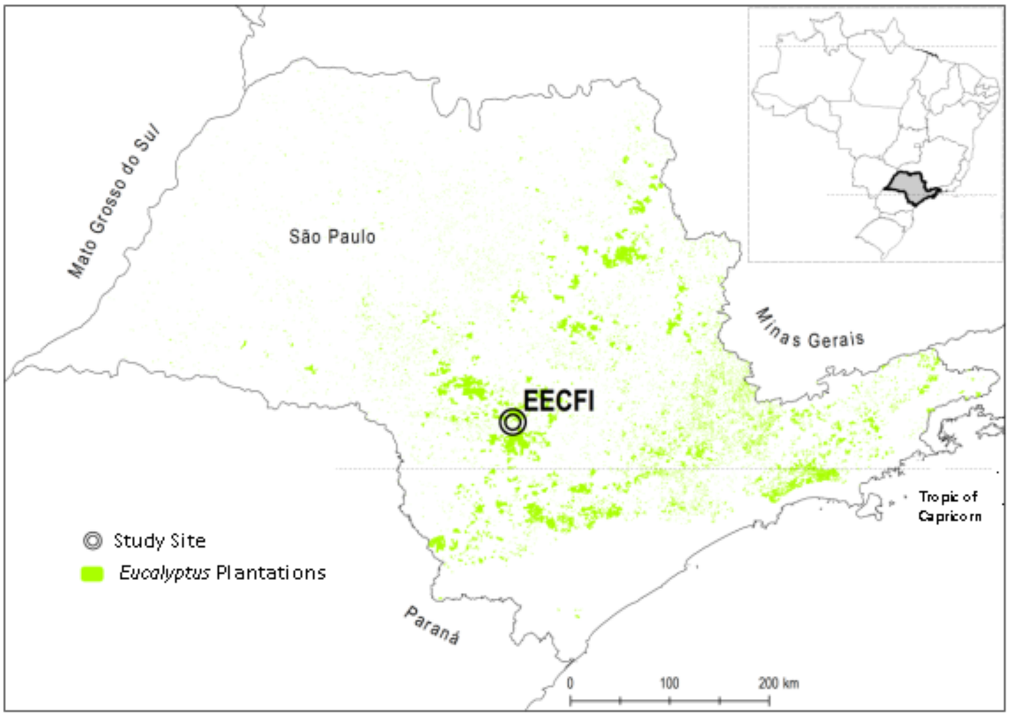

Figure 3.1. Experimental Area – Itatinga Forest Science Experimental Station.

3.2. Experimental Design

The study site was installed in 2004 (R1) and reinstalled in 2012 (R2) with three replicates of six treatments in a randomized block design. The plot sizes were 27 m x 18 m, with 81 trees per plot. The assessments were carried out in an inner plot of 15 m x 10 m (25 trees per plot) resulting in a 2 row buffer around the measurement plots. Six treatments were implemented with different management levels of forest residue removal and fertilizer applications (Table 3.2). The forest residues manipulated in this experiment include all of the organic residues remaining on the soil after wood harvesting of *E. grandis* plantations after 12 year's growth: leaves and branches less than 3 cm in diameter (canopy), bark and litter layer.

The treatments tested are shown in Table 3.2.

Table 3.2. Management of residues and nutrients applied in each treatment.

Treatment[1]	Forest residue[2]			Nutrients[3]				
	Canopy	**Bark**	**Litter layer**	**N**	**P**	**K**	**Ca**	**Mg**
				kg ha^{-1}				
ReM+F	M	M	M	130	44	125	480	120
ReM-F	M	M	M	-	-	-	-	-
ACR+F	R	R	M	130	44	125	480	120
ACR-N	R	R	M	-	44	125	480	120
ACR-P	R	R	M	10	-	125	480	120
ReR+F	R	R	R	130	44	125	480	120

[1] ReM+F = harvest residues maintained on the soil plus fertilization, ReM-F = harvest residues maintained on the soil without fertilization, ACR+F = harvest residues partially removed from the area plus fertilization, ACR-P = harvest residues partially removed from the area plus fertilization except P fertilizer, ACR-N = harvest residues partially removed from the area plus fertilization except N fertilizer, ReR+F = harvest residues removed from the area plus fertilization; [2] M = maintained on the soil, R = removed from the area; [3] N, P, K, Ca and Mg sources were ammonium sulphate, triple superphosphate, potassium chloride and limestone, respectively.

The treatments utilized in each experiment were designed to answer a specific question regarding the impact of management on soil and productivity. Each treatment and how it was carried out is described below.

3.2.1. Stocks of C, N, P and S in the soil

In order to determine the levels of C, N, P and S in the soil, five treatments were performed, with three replicates in a randomized block design. The treatments tested were as follows (Table 3.2):

1. ReM+F – Only stemwood was harvested; all of the forest residues (bark, canopy and litter layer from the previous rotation) were maintained on the soil after the clear-cutting, all nutrients were applied as fertilizer, and the soil was dressed with limestone;

2. ACR+F – The canopy (leaves and branches) and bark were removed after clear-cutting, but the litter layer was maintained; all nutrients were applied as fertilizer and the soil was dressed with limestone;
3. ACR-N – The canopy (leaves and branches) and bark were removed after the clear-cutting, but the litter layer was maintained; all nutrients except N fertilizer were applied and the soil dressed with limestone. However, a small quantity of N was applied to ensure tree survival;
4. ACR-P – The canopy (leaves and branches) and bark were removed after the clear-cutting, but the litter layer was maintained; all nutrients except P fertilizer were applied and the soil was dressed with limestone;
5. ReR+F - All of the forest residues (bark, canopy and litter layer from the previous rotation) were removed from the plot after the clear-cutting, all nutrients were applied as fertilizer, and the soil was dressed with limestone.

An unmanaged site adjacent to the experimental area and with similar soil characteristics was used as control treatments (Ref). Ref and the experimental area were identically managed until 1992. The experimental area was then harvested and Ref has continued to be left unmanaged since then. Ref presents trees of *Eucalyptus saligna* species remaining from 1940 (50m tall, on average) as well as a large regeneration of native species of the Cerrado biome (Brazilian savannah).

The ReR+F treatment was included in order to simulate a severe harvest condition where all forest residues were removed from the site, with fertilization being the only source of nutrients added to the soil. Although unusual, this harvest management system has already been used in some regions in Brazil, especially when biomass prices were high and all forest residues were removed with the objective of maximizing biomass production. Furthermore, by examining such an extreme treatment, this study brackets the range of harvest intensities actually utilized by different forest managers and allows us to consider an extreme treatment normally outside standard removal practices to bracket the forest management practices normally seen in Brazil.

3.2.2. Fractions of P in the soil

In order to determine the different fractions of P in the soil, five treatments were performed, with three replicates in a randomized block design. The treatments tested were as follows (Table 3.2):

1. ReM+F – All of the forest residues (bark, canopy and litter layer from the previous rotation) were maintained on the soil after the clear-cutting (only stemwood was harvested), all nutrients were applied as fertilizer and the soil was dressed with limestone;
2. ReM-F – All of the forest residues (bark, canopy and litter layer from the previous rotation) were maintained on the soil after the clear-cutting (only stemwood was harvested), no addition of fertilizer;
3. ACR+F – The canopy (leaves and branches) and bark were removed after the clear-cutting, the litter layer was maintained and all nutrients were applied as fertilizer and the soil was dressed with limestone;

4. ACR-P – The canopy (leaves and branches) and bark were removed after the clear-cutting, the litter layer was maintained, all nutrients except P fertilizer were applied and the soil was dressed with limestone;
5. ReR+F – All of the forest residues (bark, canopy and litter layer from the previous rotation) were removed from the plot after the clear-cutting, and all nutrients were applied as fertilizer and the soil was dressed with limestone.

An unmanaged site adjacent to the experimental area and with similar soil characteristics was used as a control treatment (Ref). Ref and the experimental area were identically managed until 1992. The experimental area was then harvested and Ref was left unmanaged afterward. Ref presents trees of *Eucalyptus saligna* species remaining from 1940 (5m tall, on average) as well as a large regeneration of native species of Cerrado biome (Brazilian savannah).

3.3. Field Procedures

After clear-cutting the 12-year-old *Eucalyptus grandis* plantation, the treatments (Table 3.2) were applied and the soil was prepared to 0.4 m depth by subsoiling with a ripper. The plots were planted with a single progeny of *E. grandis* Hill ex Maiden seedlings in June 2004 (one month after harvesting the previous plantation). The fertilizer that was applied is shown in Table 3.2. Additionally, 3.4 kg ha^{-1} of boron (B) and 30 kg ha^{-1} of *Fritted Trace Elements* (FTE) (9 % Zn, 1.8 % B, 0.8 % Cu, 2 % Mn, 3.5 % Fe, and 0.1 % Mo) were applied in every treatment. The fertilizer was applied as one base fertilizer application and two topdressing applications. The base fertilizer application was made on the same day as the planting, the triple superphosphate, the FTE, 10 kg ha^{-1} of N and K were added in a small pit to the side of each seedling, whereas the lime was applied to the whole area. The treatment ACR-N received a small application of N with the base fertilizer to ensure the survival and the initial development of plants. The topdressing application was applied as a side dressing at three and eight months after planting, with the application of N, K and B. The plantation was harvested at 8 years age and replanted one month later (November, 2012) with the reapplication of the same treatments in the same plots (Table 3.2).

During the clear-cutting harvest, the canopy and bark from the trees of each plot were maintained on the same plots. For this, each tree was felled with a chainsaw; the bark and branches were then removed manually. The seedlings in R2 were planted between the stumps of R1 without ripping. R2 was managed in the same way as R1 to evaluate the long-term effects of the residue removal and fertilizer application practices. The experimental site was maintained weed free throughout the two rotations.

3.4. Soil sampling and analysis

3.4.1. Stocks of C, N, P and S in the soil

Soil samples were taken before the application of the treatments in the first (2004) and second (2012) rotations at 0-10 and 10-20 cm. In 2016, samples were taken from the soil down to two meters (0-10, 10-20, 20-40, 40-60, 60-80, 80-100, 100-150 and 150-200cm) across three replications for each treatment. Each sample was composed of six subsamples taken from six points in the inner plot arranged in a diagonal design (Figure

3.2). For each plot, soil bulk density was calculated from soil samples taken from the walls of trenches with known volume rings.

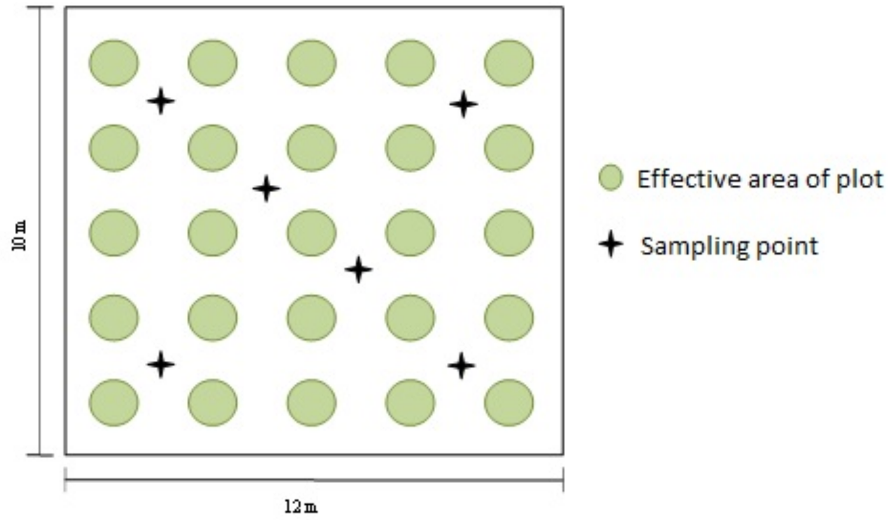

Figure 3.2. Arrangement of soil samples taken in each plot.

Samples were then air-dried at 45°C for three days and passed through a 2mm sieve for chemical analysis. Soil analyses were carried out for all treatments.

Total N was determined by dry combustion using Elemental Analyzer CHNS/2400 (Perkin Elmer) (Nelson and Sommers, 1996). Available P was determined by displacement using ion-exchange resins. Sulfur was displaced with $Ca(H_2PO_4)_2$ 0.01 mol L^{-1} solution and determined by turbidimetry using $BaSO_4$ (Van Raij et al., 2001). Oxidizable C was determined by wet oxidation (Walkley and Black, 1934).

3.4.2. Fractions of P in the soil

Soil samples were taken from 0-10cm and 10-20cm, with three replications for each treatment. Samples were taken in 2016, during the third rotation of the experiment.

Each sample was composed of six subsamples taken from six points in the plot arranged in a diagonal design (Figure 3.2). For each plot, soil bulk density was calculated from soil samples taken from the walls of trenches opened with known volume rings. Samples were then air-dried at 45°C for three days and passed through a 2mm sieve for chemical analysis

It is important to mention that the study initially intended to determine the fractions of P in the soil up to 2 meters depth. However, due to the low amounts relative to detection limits and difficulty in quantifying soluble fractions in the 20-200cm layers, these depths were not quantified. The high cost of the analysis and the short budget available in the project were also taken into consideration.

Available P was determined from the air-dried soil by displacement using ion-exchange resins (Van Raij et al., 2001). The sequential P fractionation was performed using methodology described by Hedley et al. (1982), with modifications by Condron et al. (1985). At each step, 10mL of extractant was added to 0.5 g soil in a 15 mL centrifuge tubes (1:20 soil: solution ratio) and the tubes shaken for 16 h at 25 C. This method comprises the sequential extractions of labile forms, partially labile and non-labile forms of P in the soil by separating them into organic and inorganic forms, approximately according to their relative availability for plant uptake.

The sequential extraction order and interpretation of the different forms of P removed by each extractant are given below, based on information from Tiessen and Moir (1993), Condron et al. (1985) and Gatiboni et al. (2007). It is important to mention that there is uncertainty regarding the modes of action with which these extractants act, as they vary according to the ecosystem in question and the soil analyzed (Condron and

Newman, 2011). Thus these are working definitions, not necessarily exact descriptions of specific P extracted.

a) Anion exchange resin — AER (P_{AER}) — extracts labile inorganic P readily diffused into solutions using a resin membrane (2.0 cm^2 in area).

b) 0.5 mol L^{-1} $NaHCO_3$ at pH 8.5 (P_{Bic}) — extracts labile inorganic P (Pi_{Bic}) weakly adsorbed on the surface of crystalline compounds, and labile organic P (Po_{Bic}) compounds with low recalcitrance.

c) 0.1 mol NaOH L^{-1} ($P_{HID\text{-}0.1}$) — removes moderately labile inorganic P ($Pi_{HID\text{-}0.1}$) strongly adsorbed onto Fe and Al and clay minerals, and moderately labile organic P ($Po_{HID\text{-}0.1}$) mainly associated with fulvic and humic acids adsorbed onto mineral and SOM surfaces.

d) 1.0 mol HCl L^{-1} (P_{HCl}) — extracts moderately labile inorganic P associated with apatite, other sparingly-soluble Ca-P compounds, or negatively charged oxide surfaces.

e) 0.5 mol NaOH L^{-1} ($P_{HID\text{-}0.5}$) — extracts more recalcitrant forms of inorganic P ($Pi_{HID\text{-}0.5}$) associated with Fe and Al and clay minerals and non-labile forms of organic P ($Po_{HID\text{-}0.5}$) associated with fulvic and humic acids inside aggregates.

f) Residual P ($P_{Residual}$) — obtained after the remaining soil was dried at 50 °C, milled and digested with $H_2SO_4 + H_2O_2$ in the presence of saturated $MgCl_2$.

Soil P fractions were grouped according to their lability, as predicted by the extractants. Labile P included P_{AER}, Pi_{BIC} and Po_{BIC}; moderately labile P included $P_{HID\text{-}0.1}$

(Pi and Po) and P_{HCl}, and non-labile P included $P_{HID\text{-}0.5}$ (Pi and Po) and $P_{Residual}$ (Cross and Schlesinger, 1995).

The advantage of Hedley's method over other methods described in the literature is that it helps quantify the availability of P in the short and long-term through the evaluation of the different soil layers in the profile with different degrees of relative availability to plants. This can also be used to track changes in the forms of P as a result of processes (i.e. forest management) that may alter the characteristics of soils.

3.4.3. Nutrient accumulation in the live biomass

In order to assess the accumulation of nutrients in the live biomass (canopy, bark, wood and coarse roots), 10 trees were felled in 2004, 10 trees per treatment were felled in 2012, and 3 trees per treatment was felled in 2016, using trees located in the border of each plot. Felled trees were separated into the following compartments: leaves, branches, stemwood (diameter > 3 cm at the thinner end) and stem bark. Coarse roots (diameter > 1 cm) were removed by excavation. Sub-samples were collected from all of the compartments and dried (65 C) until reaching a constant weight, and then the dry biomass of the compartments in each tree was proportionally calculated. To estimate the wood, bark biomass and stem volume of the plantation from the sampled trees, DBH and H were used as independent variables to adjust a model following the form of Schumacher and Hall (1933).

Total N was determined using the micro Kjeldahl method after sulphuric digestion. P was determined through colourimetry after nitric perchloric digestion, and S was determined through turbidimetry (Malavolta et al., 1989). Nutrient accumulation per hectare was determined by the sum of the product of biomass accumulation of each compartment and the nutrient concentrations.

3.5. Statistical analysis

3.5.1. Stocks of C, N, P and S in the soil and nutrient accumulation in the live biomass

Prior to statistical analysis, the data were tested for normality (Shapiro–Wilk) and homoscedasticity (Box-Cox). A two-way ANOVA test was performed to identify the differences between the factors considered in the experiment (soil depth and treatment). Equal sample size used was designed in order to obtain maximum power and robustness of the test. In the case of overall significant differences in the group means, LSD post hoc testing was performed to determine the differences between groups (Zar, 2010). Statistical tests were considered significant at $\alpha=0.05$. Data was analyzed using R statistical program version 3.0.1 (http://www.r-project.org/).

3.5.2. Fractions of P in the soil

The effects of the treatments on inorganic and organic P forms were examined by analysis of variance (ANOVA); using the harvest system method as a main factor, and soil depth as a subfactor. When treatment effects were significant, the differences were compared by least significant test (LSD, $p < 0.05$). Statistical tests were considered significant at $\alpha=0.05$. All statistical analyses were performed by R statistical program version 3.0.1 (http://www.r-project.org/).

CHAPTER 4. PARTITIONING OF ORGANIC AND INORGANIC PHOSPHORUS POOLS UNDER DIFFERENT HARVEST INTENSITIES

The focus of this chapter is to determine the concentrations of total and labile forms of P at depth in an Oxisol which was cultivated with *Eucalyptus* in Brazil over a 12-year-period. The cultivation was performed under different timber harvest intensities and different fertilizer applications. Recent research shows the importance of enhancing nutrient accumulation in the soil and building up its OM content in order to obtain higher timber production and a more sustainable production system. The residual material deposited on the soil surface cycles nutrients back into the soil where it becomes available for plant uptake. Fractionation techniques help in the identification of the preferable fractions of P retained in the soil, the occurrence of these fractions, and the different forms of P which are available to the plants. Based on the results, we conclude that organic and inorganic P forms in the soil act as both a source and sink to the soil solution depending on the mineralogical composition of the soil, fertilization, and harvest intensities applied. From 2004 to 2016, a reduction in the relative contribution of non-labile P in the ReM treatments was observed. However, this fraction of P still accounts for more than 60% of total P present in the soil, followed by labile and moderately labile forms of the nutrient. The highest stocks of total P were observed at 0-10cm. At the end of the first crop rotation of the experiment (2012), the ReM-F, ReR+F and the ACR+F treatments presented the highest values for the nutrient.

4.1. Results

4.1.1. Stability of soil P extracted during the fractionation process

Soil P fractions were grouped into three broad categories: labile P, moderately labile P, and non-labile P. The P_{AER} in combination with the P_{BIC} (organic and inorganic), falls within the category of labile P. The $P_{HID-0.1}$ (Pi and Po), added to P_{HCl} is categorized by moderately labile P. Finally, the sum of $P_{HID-0.5}$ (Pi and Po) and the P_{Resid} (Pi and Po) are categorized by stable P. The grouping of $P_{HID-0.5}$ in the non-labile fraction of P was necessitated by the change in the order of the extraction procedure (Condron et al., 1985). This is because the fraction corresponds to the P which is protected by intra-aggregates of the soil, which would only be considered of moderate lability in cases of exhaustion of the more labile forms of P.

In soil fertility, regardless of its chemical nature, P is divided according to the ease with which it reenters the soil solution (Parfitt, 1978). The labile, the moderately labile, and non-labile forms are extracted by fractionation. This is done in a sequence of extraction forces proportionate to the potential retention of P in the soil, starting from more soluble – and therefore theoretically more bioavailable fractions – and then the more recalcitrant forms, considered unavailable (Cross and Schlesinger, 1995).

Table 4.1. Soil P concentrations extracted sequentially by anion exchange resin (P_{AER}) bicarbonate in inorganic (Pi_{BIC}) and organic (Po_{BIC}) forms, Na hydroxide at 0.1 mol L^{-1} ($Pi_{HID0.1}$ and $Po_{HID0.1}$) chlorite acid (P_{HCl}) Na hydroxide at 0.5 mol L^{-1} ($Pi_{HID0.5}$ and $Po_{HID0.5}$) and P residual, in treatments with different timber harvest intensity and P fertilizer application.

	Labile forms of P			Moderately labile forms of P			Non-labile forms of P			
Treatment[1]	P_{AER}	Pi_{BIC}	Po_{BIC}	$Pi_{HID0.1}$	$Po_{HID0.1}$	P_{HCl}	$Pi_{HID0.5}$	$Po_{HID0.5}$	P_{RESID}	Ptotal
	mg kg^{-1}									
0-10 cm										
Installation of the experiment (2004)										
	5.9	13.6	0.9	4.9	41.6	0.19	11.4	3	96.4	178
End of first crop rotation of the experiment (2012)										
ReM+F	4.5 bc	8.4 b	2.5 a	3.7 b	32.1 b	0.16 ab	8.8 b	6.8 a	90	157 b
ACR+F	6.2 ab	12.3 ab	3.3 a	5.3 a	38.9 ab	0.09 b	8.3 b	6.6 a	101.5	182 a
ReR+F	5.2 abc	13.4 a	0.9 b	4.4 ab	30.5 b	0.14 ab	14.8 a	4.9 a	107.9	182 a
ACR-P	3.9 c	10.8 ab	1.6 ab	3.7 b	27.9 b	0.19 a	11.5 ab	5.2 a	98.1	163 b
ReM-F	6.8 a	12.8 a	0.6 b	2.3 c	48.7 a	0.13 ab	13.9 a	0.5 b	102.9	189 a
41 months after re-installation of the experiment (2016)										
ReM+F	5.1 a	12.2 a	1 ab	3 bc	43.3	0.19 c	13	10.0 a	96	184
ACR+F	4.1 ab	10.2 ab	0.8 bc	6.7 a	35.1	0.33 b	13.8	7.2 ab	104	182
ReR+F	2.6 c	7.2 b	1.2 a	2.9 bc	40.4	0.14 c	11.6	4.4 b	104.9	175
ACR-P	3.7 b	7.6 b	0.3 d	3.3 b	40.1	0.21 bc	11.1	5.1 b	106.9	178
ReM-F	5.2 a	12.9 a	0.7 c	1.6 c	39.7	0.57 a	12.5	6.4 ab	102.4	182
10-20 cm										
Installation of the experiment (2004)										
	2.4	4.1	3.9	3.4	22.8	0.43	10.6	1.1	89.8	139
End of first crop rotation of the experiment (2012)										
ReM+F	2.4	4.9	2.7 b	2.7 b	25.4	0.04	10.6	1.4 b	90.1	140 b
ACR+F	1.6	3.4	4.7 ab	3.2 ab	25.2	0.1	11	3.9 ab	91.9	145 a
ReR+F	2.5	5.3	3.8 ab	4.1 ab	26.9	0.28	9.8	4.9 a	90.9	148 a
ACR-P	2.6	3.5	5.6 a	6.1 a	24	0.07	11.9	4.5 ab	95	153 a
ReM-F	2.2	3.6	4.7 ab	3 b	29.7	0.07	8.3	0.1 c	89.2	141 ab
41 months after re-installation of the experiment (2016)										
ReM+F	2	3.9 b	3 c	7.4	23.7 b	0.09 b	11.1 b	2.4 c	97.5 b	151 b
ACR+F	1.4	2.1 c	4.2 b	5.3	20.5 bc	0.19 a	9.3 b	5 b	93.1 b	141 c
ReR+F	1.7	0.4 d	7.3 a	5.6	18 c	0.09 b	13 b	6.9 a	95.6 b	149 bc
ACR-P	1.8	2.6 c	7.0 a	3.9	30.6 a	0.16 a	9.6 b	6.6 ab	106.6 a	169 a
ReM-F	2	6.1 a	3.3 c	5	24.7 b	0.09 b	19.9 a	0.1 d	105 a	167 a

[1] ReM+F = harvest residues maintained on the soil plus fertilization, ACR+F = harvest residues partially removed from the area plus fertilization, ReR+F = harvest residues removed from the area plus fertilization, ACR-P = harvest residues partially removed from the area plus fertilization except P fertilizer, ReM-F = harvest residues maintained on the soil without fertilization; *Means followed by the same letter or not followed by letter do not differ by LSD test at 5 % of probability in each depth and period of installation of the experiment.

4.1.2. *Labile P fractions (P_{AER}, Po_{BIC} and Pi_{BIC})*

In 2012, the concentrations of P_{org} and P_{inorg} in the 0-10cm layer were significantly greater under ACR+F, ReR+F and ReM-F treatments ($p < 0.0001$) (Figure 4.1). The largest differences in P_{inorg} were also observed in these treatments. The concentrations of P_{org} were higher in the ReM+F and ACR+F treatments. At 10-20cm, no differences in the concentrations of P_{org} and P_{inorg} were observed between the treatments tested ($p < 0.0001$). For all treatments, no differences were observed in the concentration of P_{inorg}. The ACR+F, ACR-P and ReM-F treatments presented the greater concentrations of P_{org}.

In 2016, the concentrations of P_{org} and P_{inorg} in the 0-10cm layer were significantly greater in the ReM+F and ReM-F treatments ($p < 0.0001$) (Figure 4.1). The ReR+F and ACR-P treatments presented the lowest values for the aforementioned P fractions. The largest differences in P_{inorg} were also observed in the ReM+F and ReM-F treatments. The ReR+F treatment presented greater concentrations of P_{org} when compared to the other treatments, while ACR-P presented the lowest concentration. At 10-20cm, the highest concentrations of P_{org} and P_{inorg} were observed in the ACR-P and ReM-F treatments. The ReM-F treatment presented greater concentration of P_{inorg} when compared to the other treatments, while the ReR+F treatment presented the lowest concentration of P_{inorg}. The concentration of P_{org} was significantly greater under ReR+F and ACR-P treatments.

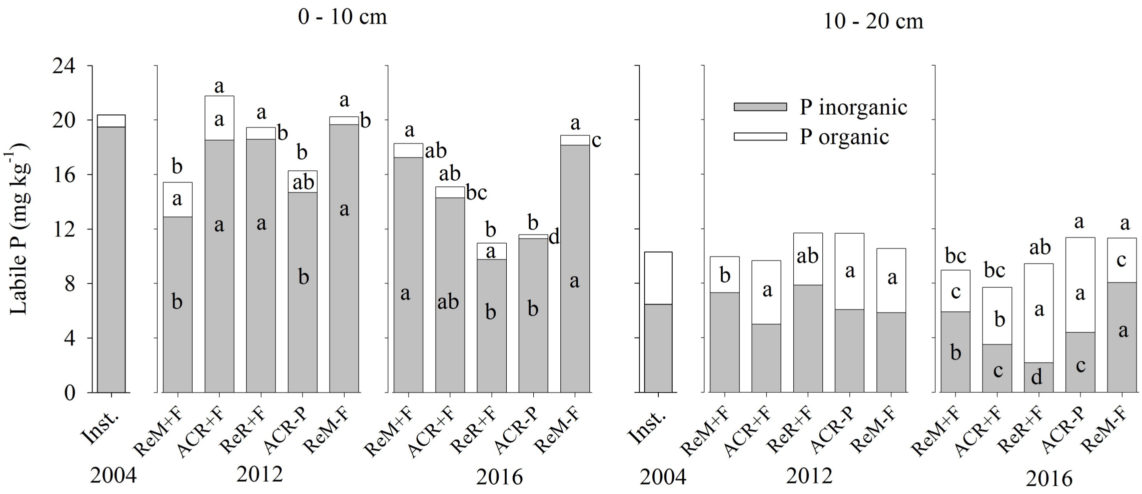

Figure 4.1. Concentration of labile P fractions in the soil by depth and different timber harvest intensity management$^{(*)}$ in two rotations of the experiment (from 2004 to 2016).

$^{(*)}$ ReM+F = harvest residues maintained on the soil plus fertilization, ACR+F = harvest residues partially removed from the area plus fertilization, ReR+F = harvest residues removed from the area plus fertilization, ACR-P = harvest residues partially removed from the area plus fertilization except P fertilizer, ReM-F = harvest residues maintained on the soil without fertilization.

4.1.3. Moderately labile P fractions ($Pi_{HID0.1}$, $Po_{HID0.1}$ and P_{HCl})

In 2012, moderately labile P fractions were high in the ReM-F treatment compared to the other treatments in the 0-10cm layer ($p < 0.0001$) (Figure 4.2). This same treatment presented the higher concentration of moderately labile P_{org}. In contrast, its inorganic form was the lowest observed among the treatments. The ACR+F treatment presented higher concentrations of the inorganic form of P. At 10-20cm, no differences in the concentrations of moderately labile P were observed between the treatments tested. The ACR-P treatment presented significantly higher concentrations of moderately labile P_{inorg}, while the concentration in the ReM-F treatment was at the lowest level.

In 2016, no differences were observed between the moderately labile forms of P in the 0-10cm layer ($p < 0.0001$) (Figure 4.2). The ReM+F treatment presented the higher concentration of P_{org}. Concentrations of P_{inorg} in all treatments were very low: values were less than 7 mg kg^{-1} in the ACR+F treatment and less than 5 mg kg^{-1} in the ReM-F treatment. At 10-20cm, the ACR-P treatment presented a slight increase in the moderately labile forms of P when compared to the other treatments. No differences between treatments were observed between the inorganic forms of moderately labile P, with values ranging from 3 to 6 mg kg^{-1}.

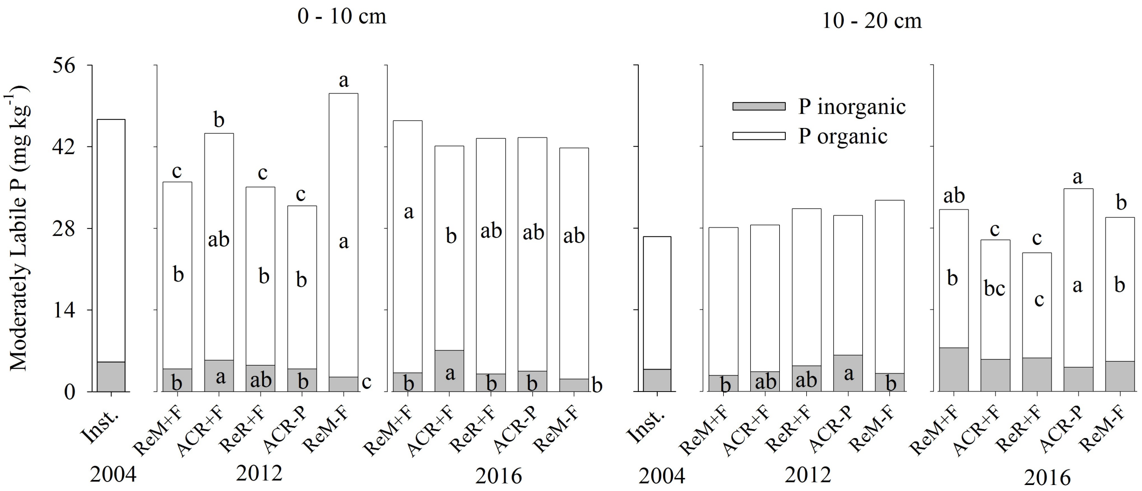

Figure 4.2. Concentrations of moderately labile P fractions in the soil with different timber harvest intensity management(*) in two rotations of the experiment (from 2004 to 2016).

(*) ReM+F = harvest residues maintained on the soil plus fertilization, ACR+F = harvest residues partially removed from the area plus fertilization, ReR+F = harvest residues removed from the area plus fertilization, ACR-P = harvest residues partially removed from the area plus fertilization except P fertilizer, ReM-F = harvest residues maintained on the soil without fertilization.

4.1.4. Non-Labile P fractions ($Pi_{HID0.5}$, $Po_{HID0.5}$ and P_{RESID})

The study noted that non-labile P constituted a sizeable fraction. In general, the non-labile P stocks in the 0-10 cm soil layer showed a significant increase over the 12 years of the experiment ($p < 0.0001$). In 2012, concentrations were greater in the ReR+F treatment at 0-10cm, while the ReM+F treatment presented the lowest value - with values ranging from 127.6 mg kg^{-1} and 105.6 mg kg^{-1}, respectively (Figure 4.3). The concentration of non-labile P_{inorg} was noticeably higher when compared to the other treatments, while the ReM-F treatment presented the lowest concentration of non-labile P_{inorg} ($p < 0.0001$). At 10-20cm, treatments did not significantly differ in regards to the concentration of non-labile P.

In 2016, the concentration of non-labile P fractions did not significantly differ between treatments in the 0-10cm layer, with values ranging from 118 mg kg^{-1} and 125 mg kg^{-1}. At 10-20cm, concentrations were greater in ACR-P and ReM-F treatments. The ReM-F treatment showed the greater concentration of non-labile P_{inorg}, while showing the lower concentration of non-labile P_{org}.

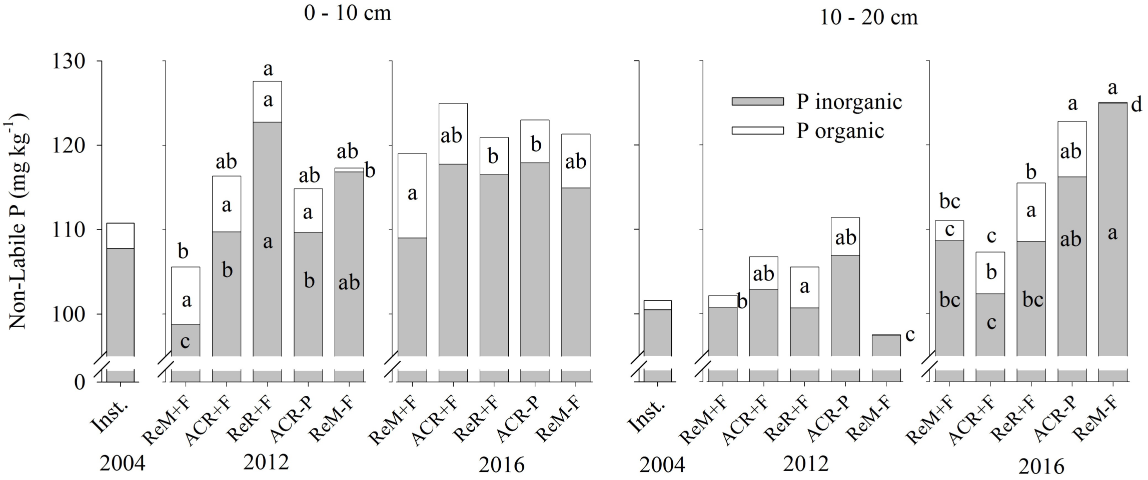

Figure 4.3. Concentrations of non-Labile P fractions in the soil with different timber harvest intensity management(*) in two rotations of the experiment (from 2004 to 2016).

(*) ReM+F = harvest residues maintained on the soil plus fertilization, ACR+F = harvest residues partially removed from the area plus fertilization, ReR+F = harvest residues removed from the area plus fertilization, ACR-P = harvest residues partially removed from the area plus fertilization except P fertilizer, ReM-F = harvest residues maintained on the soil without fertilization.

4.1.5. Total P stocks

Twelve years after the installation of this experimental site, differences between treatments were observed in the quantities of P partitioned between organic and inorganic P pools to a soil depth of 20cm ($p < 0.0001$) (Figure 4.4).

Prior to the installation of the experiment in 2004, the site contained 20.4 mg kg^{-1}, 46.69 mg kg^{-1} and 110.8 mg kg^{-1} of labile, moderately labile and non-labile P respectively, in the 0-10cm layer, with a total concentration of 177.89 mg kg^{-1} of P (Table 4.1). In 2012 (the end of first rotation of the experiment), the ReM-F, ACR+F and ReR+F treatments showed an increase of about 6%, 2.5% and 2% respectively, in total P stocks at 0-10cm when compared to the initial concentration of the nutrient before the installation of the experiment (Table 4.1 ; Figure 4.4). In 2016 (41 months after re-installation of the experiment), the total P stocks were observed to increase in almost all treatments in relation to the results obtained in 2012, with the ReM+F treatment showing an increment of 17%.

At 10-20 cm, in 2004, the concentrations of labile, moderately labile and non-labile P were 10.4 mg kg^{-1}, 26.63 mg kg^{-1} and 101.5 mg kg^{-1} respectively (Table 4.1). Total P stock was 138.53 mg kg^{-1}. In 2012 (end of first rotation of the experiment), a slight increase was observed in the total P stocks in all treatments, statistically different among each other ($p < 0.0001$) (Table 4.1 ; Figure 4.4). In 2016, an increase was also observed in total P stocks in some treatments when compared to the values observed in 2012. The ReM-F and ACR-P treatments showed an increase in total P stocks of 17% and 10%, respectively.

Considering the stocks of inorganic P in the soil in the 0-10 cm layer, after the first rotation of the experiment (2012) it is apparent that ReR+F and ReM-F treatments presented values higher than those initially obtained in 2004 - when the experiment was first implemented. The lowest values of total P were observed in the ReM+F treatment. After two rotations (2016), a small difference was observed in relation to the values initially obtained in 2004 ($p < 0.0001$). The ACR+F treatment presented the highest nutrient stocks, followed by the ReM-F treatment. In 2012, the highest values of organic P were obtained in the 0-10cm layer of treatments ACR + F, ReR + F and ReM-F. In 2016, the highest values were observed in the ReM + F treatment, followed by the ReM-F treatment.

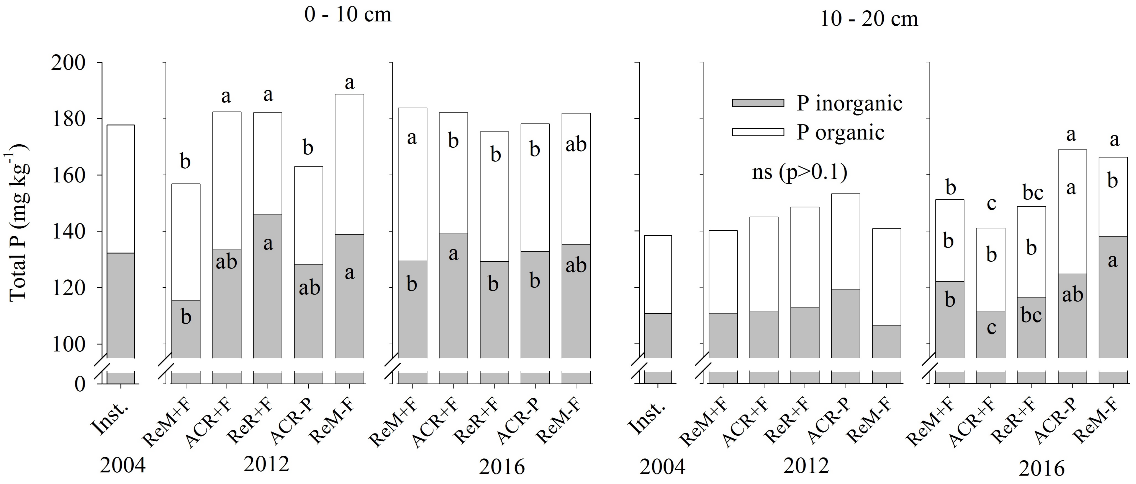

Figure 4.4. Concentrations of total P in the soil by depth and different timber harvest intensity management(*) in two rotations of the experiment (from 2004 to 2016).

(*) ReM+F = harvest residues maintained on the soil plus fertilization, ACR+F = harvest residues partially removed from the area plus fertilization, ReR+F = harvest residues removed from the area plus fertilization, ACR-P = harvest residues partially removed from the area plus fertilization except P fertilizer, ReM-F = harvest residues maintained on the soil without fertilization.

4.2. Discussion

4.2.1. The effects of forest harvest removal on soil P fractions

Long-term site productivity studies are particularly valuable for assessing the cumulative impacts of harvest management on soil chemical and physical characteristics as well as timber productivity (Mendham et al., 2014). In this study we focused on the P fractions found in the soil in a strategic region for wood production of Brazil where Oxisols have been depleted due to consecutive weathering, and fertilization is essential for plant growth.

The effect of forest harvest removal on the different P fractions found in the soil and their availability to plants is inconsistent between studies in the literature (Laclau et al., 2003; Rocha et al., 2016b; Maranguit et al., 2017). First, one should consider the soil physical and chemical characteristics, which are highly variable across the globe (Tamminen et al., 2012; Achat et al., 2015; Rocha et al., 2016). Secondly, the quality of the biomass left on the soil and its mineralization rate influence on the cycling of nutrients that will, in turn, be deposited back to the soil (Paul and Clark, 1996; Mack et al., 2014). Hence, this decomposition process is dependent upon the soil microbial activity (Barber, 1984) and the form in which these nutrients are found in the decomposing material (Giacomini et al., 2003).

A meta-analysis evaluating the impacts of harvest residues removal from the soil, revealed that consecutive removal of harvest residues significantly decreased total P stocks in soils in 6% to 10%(Achat et al., 2015). Kumaraswamy et al. (2014), on the other hand, evaluated the effects of harvest residue management on soil properties at 4 multi-rotation *Eucalyptus* plantations in India and found no differences on total P stocks according to the different harvest managements tested.

In this study, changes in the total P stocks were observed between rotation cycles and among the soil layers evaluated ($p < 0.0001$). The highest stocks of total P were observed at 0-10cm. At the end of the first crop rotation of the experiment (2012), the ReM-F, ReR+F and the ACR+F treatments presented the highest values for the nutrient. The treatments which harvest residues were partially or totally removed from the soil + fertilization presented similar results to the treatment which the soil received nutrients exclusively from the decomposition of the organic material left on the soil (Figure 4.4).

At 41 months after re-installation of the experiment (2016), total P stocks were higher in all treatments, not statistically different between each other ($p < 0.0001$). This can be explained by the fact that a significant proportion of the total P stocks is related to soil organic matter and hence, long-term availability of the nutrient is linked more closely to organic matter turnover than to the inorganic forms of P (Turner and Lambert, 1986; Brady and Weil, 2013; Turner and Lambert, 2016). The time it takes for nutrients to be deposited back to the soil during the decomposition process of the organic material on the soil should be taken in to consideration (Fleming et al., 2006).

The maintenance of forest harvest residues, such as bark, needles and leaves, has a large impact on P availability in forest sites; they represent around 57% to 70% of P accumulated in the soil biomass in *Eucalyptus* plantations (Laclau et al., 2003; Bazani et al., 2014; Santana et al., 2008; Rocha et al., 2016b). In turn, decomposition of these sources partially replace the labile fraction of P of the soil, reducing the demand for phosphorus fertilization.

Over the period of evaluation of this experiment, labile P in the 0-10cm layer presented a reduction between 10% to 36%, being most noticeable in the ReR+F and ACR-P treatments (-36% in each treatment) ($p < 0.0001$). Moreover, it was observed a reduction of approximately 35% in the inorganic forms of P in the labile fraction in the ReM+F treatment. Labile P at 10-20 cm remained the same, showing lower concentrations when compared to the stocks in the upper layer. Most of the P added by fertilization or by organic matter turnover has been accumulated in non-labile forms at ReR and ACR treatments (>50%). These results are consistent with other long-term experiments (Negassa and Leinweber, 2009; Rodrigues et al., 2015), demonstrating the relatively inefficiency of P inputs through fertilization in Brazilian Oxisols.

In 2016, on the other hand, an increase in the total P stocks and the inorganic forms of the nutrient in all the treatments was observed ($p < 0.0001$). The SOM and P availability at these plots are tightly coupled. Higher quantities of SOM on sites with harvest residues retained on the soil surface contribute to P availability for plants, since the addition of organic substances to the soil serves as a C source that enhances microbial biomass and phosphatase activity, increasing the transformation of occluded P into a readily available form suitable for plant uptake (Corbeels et al., 2005; Wang et al., 2010; Xu et al., 2012; Maranguit et al., 2017). Huang et al. (2015) studied the influence of vegetation cover in coastal wetland soils in China, and found that OM accumulation and decomposition play an important role in regulating potential P dynamics in the soils.

4.2.2. Relative contribution of the different forms of phosphorus in the soil

From 2004 to 2016, a reduction in the relative contribution of non-labile P in the ReM treatments was observed (Figure 4.5). However, this fraction of P still accounts for more than 60% of total P present in the soil, followed by labile and moderately labile forms of the nutrient (Figure 4.5). Moreover, organic P forms are predominant over the inorganic forms of the nutrient, especially in those treatments that have not received fertilization with P (ReM-F and ACR-P).

The reduction in the concentration of non-labile P in the treatments which harvest residues were maintained on the soil can be explained by the fact that the slow and gradual mineralization of organic material provides the release and redistribution of organic forms of P, which are more mobile through the soil profile and less susceptible to adsorption reactions. Moreover, they are able to maintain a continuous flux of different forms of C found in the soil, which compete with phosphate ions for the positive charges of inorganic colloids of the soil. They are also able to complex Al^{3+} and Fe^{3+} ions, forming stable and complex hydrosoluble compounds, reducing non-labile P stocks, and consequently, increasing available forms of P for root absorption (Taiz and Zeiger, 2010; Rheinheinmer and Anghinoni, 2001).

Medium-term and long-term studies of sequentially-extracted P suggest that both labile and moderately labile P fractions in temperate, subtropical, and tropical soils are depleted when little to no P is added after harvest (Negassa and Leinweber, 2009; Oliveira et al., 2015; Stutter et al., 2015). This suggests that land use management and soil P stocks are tightly coupled.

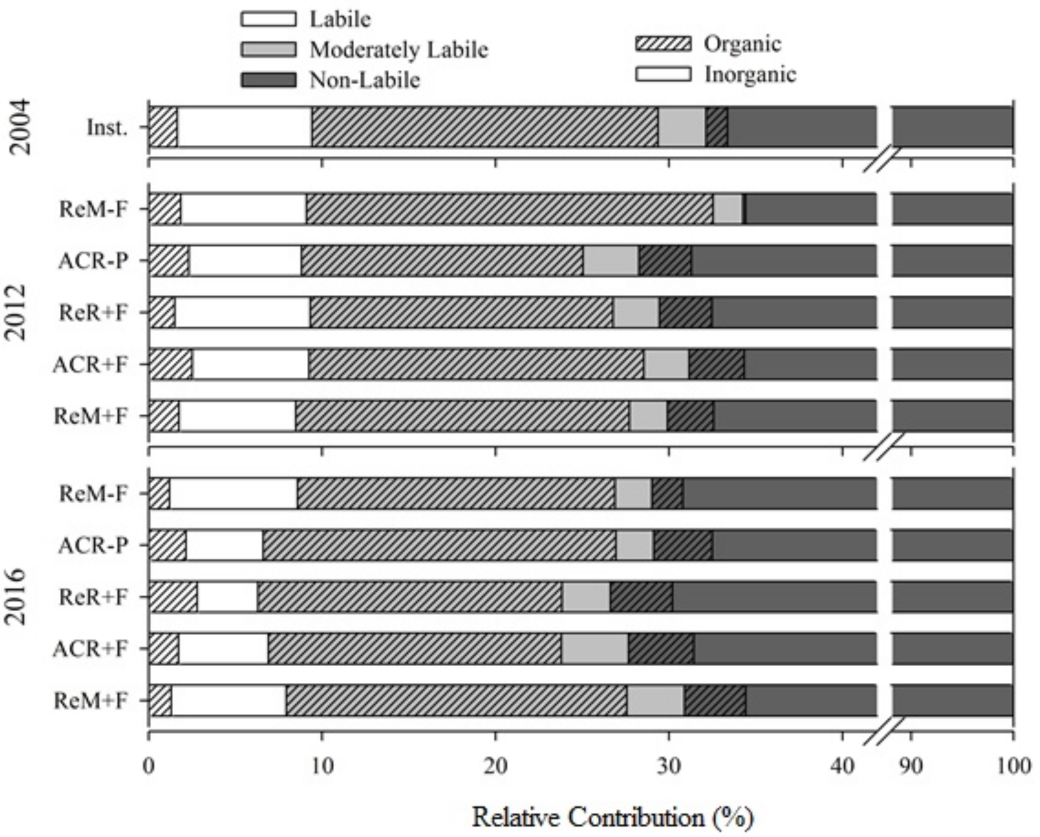

Figure 4.5. Relative contribution of the different forms of phosphorus in the soil by different timber harvest intensity management(*) in two rotations of the experiment (from 2004 to 2016).

(*) ReM-F = harvest residues maintained on the soil without fertilization, ACR-P = harvest residues partially removed from the area plus fertilization except P fertilizer, ReR+F = harvest residues removed from the area plus fertilization, ACR+F = harvest residues partially removed from the area plus fertilization, ReM+F = harvest residues maintained on the soil plus fertilization.

4.2.3. Aspects of soil management and P sustainability

Our studies at this long-term site productivity experiment using the Hedley fractionation procedure show that organic and inorganic P forms in the soil can act as source or sink of the nutrient to the soil solution, depending on the mineralogical composition of the soil, fertilization and harvest management applied (Rheinheinmer and Anghinoni, 2001; Rocha et al., 2016).

Hu et al. (2016), in a study investigating different harvest managements in a reforested spruce forest in China, concluded that harvest management techniques are responsible for improvements in soil moisture, organic matter, and microbial and phosphatase activity, and thus, to the increase in labile P stocks. Costa et al. (2016) evaluated the distribution of labile and non-labile fractions of P in soil in 10 sites of *Eucalyptus* in the Southeast of Brazil and concluded that, based on the soil and climate conditions of the experimental sites, the available P pool was highly dependent on inorganic and occluded pools. The authors also observed that the organic pool acted predominantly as a sink of P on available and inorganic pools.

These studies highlight the importance of studying the sequestration of P throughout the soil profile. The fractionation procedure of P establishes the potential for labile fractions to supply *Eucalyptus* trees, which is not possible in routine methods for analysis of soil fertility (Rodrigues et al., 2016; Maranguit et al., 2017). The reserves of P in the soil that are not readily available for plant uptake are a potentially valuable resource for plant nutrition, decreasing global pressure on finite reserves of phosphate rock (Nave et al., 2010; Ranger et al., 2011).

Most of the P fractions in the soil are mobile and susceptible to transformation depending on environmental conditions (Lukkari et al., 2007). Studies suggest that the variation in the different forms of the nutrient in the soil can only be accounted for based on OM content (Huang et al., 2015). Specifically, the accumulation and decomposition of OM drives the increasing availability of soluble forms of P in the soil. Therefore, the accumulation of harvest residues on the soil surface is a viable option for increasing SOM on a long-term basis and can help explain the variations of the different forms of P through the profile.

The complex dynamics of this nutrient in tropical conditions validates other studies about the relationships between fractions of P and other nutrients and substances present in the soil. Tropical soils may conserve more C under increased OM inputs because P is limiting decomposition. Such P limitations will also effect other microbial processes such as N transformations (Binkley et al., 2003). While most temperate ecosystems, which are limited by N supply, may store added mineral or atmospheric N for long periods, a P limited tropical forest could rapidly loose substantial amounts of N oxides upon N fertilization (Hall and Matson, 2003).

The noticeable limitations of tropical soils not only affect plant productivity, but also decomposition and immobilization processes, and therefore the storage and cycling of other essential nutrients for plant growth, such as C and N. With that, more research is essential to better understand the role of the different fractions of P in within the soil profile and how it could be related to different harvest management techniques.

CHAPTER 5. EFFECT OF TIMBER HARVEST INTENSITIES AND FERTILIZER APPLICATIONS ON STOCKS OF C, N, P AND S

In this chapter we discuss the results relative to the stocks of available P and S, total N and oxidizable C at depth in an Oxisol cultivated with *Eucalyptus* in Brazil under different timber harvest intensities and fertilizer applications over 12 years. In addition, we calculate the balance of the relationship between N, P and S outputs by harvest and their stocks in the soil during each plant cycle. A reduction in all nutrient stocks is observed in the 0-20 cm layer for all treatments. For N, this reduction is 20% higher when harvest residues were removed from the soil, and 40% higher when no N fertilizer was applied. The maintenance of harvest residues on the soil reduces the loss of N, P, and S due to harvest by 120%, 50%, and 40%, respectively.

5.1. Nutrient balance after two crop rotations

Before the installation of the experiment in 2004, the site contained 370, 11 and 9.2 kg ha^{-1} of N, P and S, respectively, in the 0-20 cm layer (Figure 5.1). On that occasion, there was an accumulation of approximately 660, 90 and 100 kg ha^{-1} of N, P and S, respectively, in the live biomass and litter layer (250 t ha^{-1} in total). The nutrient losses from harvest within the ReR+F treatment were higher than that of the ReM+F treatment by 120, 50 and 40% of N, P and S respectively. For the ACR treatments, the losses from harvest were 65, 30 and 20% higher, respectively, than the ReM+F treatment (Table 5.1).

In 2012 (the end of first rotation of the experiment), the ReM+F treatment contained the largest stocks of N (206 kg ha^{-1}) and P (13.8 kg ha^{-1}) in the 0-20 cm layer, while the highest stocks of S (13.2 kg ha^{-1}) for this layer were observed in the ACR-P treatment. The lowest stocks of N (151 kg ha^{-1}) and P (9 kg ha^{-1}) were observed in the ACR-N and ACR-P treatments, respectively (Figure 5.1).

At the end of the first rotation of the experiment (8 years) there were no observed differences in the volume of wood produced in the treatments ReM+F, ACR+F, ReR+F and ACR-N, which were about 420 m^3 ha^{-1} on average. On the other hand, the absence of fertilization with P (ACR-P treatment) resulted in approximately a 10% reduction in the final wood volume (Hubner, 2015). Even with small differences in tree growth, the renewal of the experiment for the next crop rotation resulted in great differences in the quantity of nutrient losses among the treatments, due to the different parts of the plants removed at harvest (Table 5.1).

At 41 months into the second rotation, the ReM+F treatment contained 100 Mg ha^{-1} of live biomass (wood, bark, canopy and coarse roots). Compared to the ReM+F treatment, the biomass accumulated in the ACR+F, ReR+F, ACR-N and ACR-P treatments was lower by 14, 15, 16 and 25%, respectively. The accumulated N, P, and S in the live biomass of the ACR-P treatment was 33, 40 and 20% lower, respectively, compared to the ReM+F treatment.

Between 2004 and 2012, N contents in the soil from 0-20 cm depth was reduced by around 50% with larger losses in the ACR-N treatment and smaller losses in the ReM+F treatment. From 2012 to 2016, a smaller reduction in soil N was observed, with the exception of the ACR-P treatment. Only small losses of available P were observed between 2004 and 2012, but greater losses occurred after reinstallation of treatments, especially for repeated removal of harvest residues (ReR+F). In the ACR+F treatment the concentration of available P in surface soil was constant through the years. A larger reduction was observed for the ReR+F and ACR-P treatments with losses of approximately 30% from 2004 to 2016. In regards to available S, a reduction in the ReR+F treatment from 2004 to 2012 was observed, and an increase was observed in the ACR-P treatment. For the other treatments, S stocks were constant. From 2012 to 2016, available S stocks improved for all treatments (Figure 5.1).

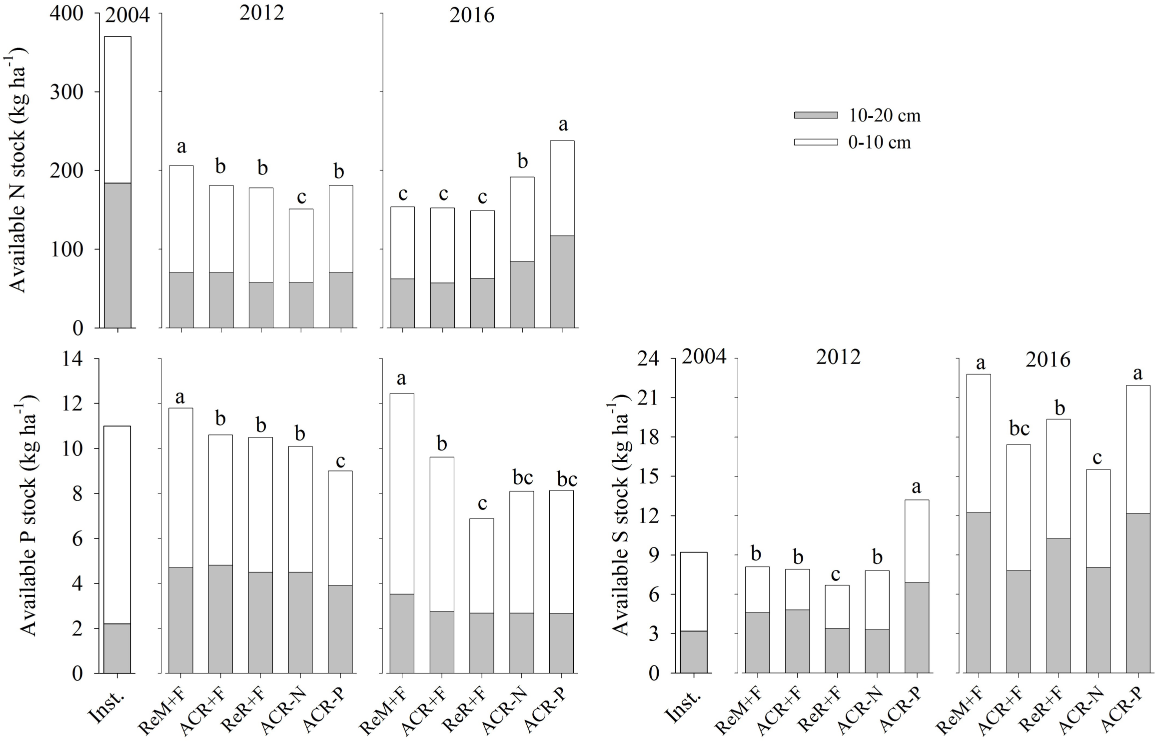

Figure 5.1. Available N, P and S stocks in the 0-20 cm soil layer before the setup of treatments (2004), before the harvest of the first rotation (2012) and at 4 years into the second rotation (2016) in a *Eucalyptus* plantation under different harvest residue management and fertilizer application strategies(*). P was determined by resin extraction, S was determined by extraction with $Ca(H_2PO_4)_2$ 0.001 mol L^{-1} and available N was calculated assuming that 10 % of total N is mineralizable (Pulito et al., 2015). Columns with the same letter are not significantly different from each other.

(*) ReM-F = harvest residues maintained on the soil without fertilization, ACR+F = harvest residues partially removed from the area plus fertilization, ReR+F = harvest residues removed from the area plus fertilization, ACR-N = harvest residues partially removed from the area plus fertilization except N fertilizer, ACR-P = harvest residues partially removed from the area plus fertilization except P fertilizer.

5.2. Soil nutrient stocks in 2016

At four years into the second study rotation (2016), large differences between treatments were found in the stocks of Total N, Available P and S and oxidizable C in the soil to 200 cm depth (Figure 5.2). The largest stock of oxidizable C in the soil was observed in the ReM+F treatment ($p>0.001$), and this treatment contained the most oxidizable C at every individual sampling layer except between 60-100 cm. The ReR+F treatment contained the lowest stocks of oxidizable C ($p>0.002$) up to 40cm depth, and did not differ from the ACR treatments in the deeper layers. The supply of oxidizable carbon in the entire 0-200 cm profile of the ReM+F treatment was approximately 130 Mg ha^{-1}, which is 10 % greater than the ACR+F treatment and 20% higher than the ReR+F treatment.

The ACR-P treatment contained the highest stock of N ($p>0.001$) in all the layers considered. ReM+F, ACR+F and ReR+F treatments presented the lowest stocks of N, not differing statistically from each other. ReR+F and ACR-N treatments presented the lowest stocks of P, and were not significantly different. The stocks of N in the 0-200 cm layer of the soil were approximately 10 Mg ha^{-1} in the ReM+F, ACR+F and ReR+F treatments. Stocks of available P in the soil were approximately 40 kg ha^{-1} in the ReM+F and ACR+F treatments, which is 60% higher than the stock of P observed in the ReR+F treatment.

ACR+F and ReR+F treatments contained the largest stocks of S in the soil ($p>0.001$). The ACR-N and ACR-P treatments contained the lowest stocks of S ($p>0.001$). The stock of S available in the 0-200cm layer was 280 kg ha^{-1} in the ReM+F, ACR+F and ReR+F treatments. ACR-N treatment had a stock 30% lower than the previous treatments.

Table 5.1. Nutrient stocked in the forest biomass, inputs and outputs of nutrients in two crop rotations a *Eucalyptus* plantation under different harvest residue management and fertilizer application strategies(*).

Component	Biomass	N	P	S
	t ha^{-1}		kg ha^{-1}	
Nutrients stocks before 2004 harvest				
Litter layer	24 (1)[1]	141 (17)	12 (1)	12 (1)
Stem wood	167 (15)	250 (20)	52 (3)	67 (5)
Stem bark	18 (1)	62 (3)	8 (1)	7 (1)
Canopy	9 (1)	100 (9)	7 (1)	6 (1)
Root	33 (5)	109 (18)	11 (2)	14 (2)
Total	251 (12)	662 (48)	90 (10)	106 (15)
Harvest outputs in 2004				
Treatment ReM+F	167 (15)	250 (20)	52 (3)	67 (5)
Treatments ACR+F, ACR-N and ACR-P	194 (18)	412 (46)	67 (4)	80 (7)
Treatment ReR+F	218 (18)	553 (49)	79 (15)	92 (8)
Inputs from 2004 to 2012				
Atmospheric deposition[2]		32	-	-
Fertilizer application[3]:				
Treatments ReM+F, ACR+F and ReR+F		130	44	143
Treatment ACR-N		10	44	11
Treatment ACR-P		130	0	143
Harvest outputs in 2012				
Treatment ReM+F	191 (10)	313 (17)	60 (3)	71 (4)
Treatment ACR+F	196 (20)	412 (37)	67 (7)	71 (7)
Treatment ReR+F	220 (16)	542 (39)	79 (6)	80 (6)
Treatment ACR-N	200 (32)	426 (60)	69 (11)	70 (11)
Treatment ACR-P	180 (23)	386 (48)	61 (8)	65 (8)
Inputs from 2012 to 2016				
Atmospheric deposition		16	-	-
Fertilizer application:				
Treatments ReM+F, ACR+F and ReR+F		130	44	143
Treatment ACR-N		10	44	11
Treatment ACR-P		130	0	143
Accumulated in the live biomass in 2016				
Treatment ReM+F	100 (3)	255 (20)	25 (4)	40 (2)
Treatment ACR+F	86 (3)	207 (26)	21 (1)	37 (2)
Treatment ReR+F	85 (7)	202 (17)	18 (2)	37 (4)
Treatment ACR-N	84 (5)	195 (12)	18 (1)	36 (1)
Treatment ACR-P	75 (2)	172 (3)	15 (2)	32 (1)

[1] Standard deviation; [2] By Laclau et al. (2010); [3] It is assumed that 100% of fertilizer application was available;

(*) ReM-F = harvest residues maintained on the soil without fertilization, ACR+F = harvest residues partially removed from the area plus fertilization, ReR+F = harvest residues removed from the area plus fertilization, ACR-N = harvest residues partially removed from the area plus fertilization except N fertilizer, ACR-P = harvest residues partially removed from the area plus fertilization except P fertilizer.

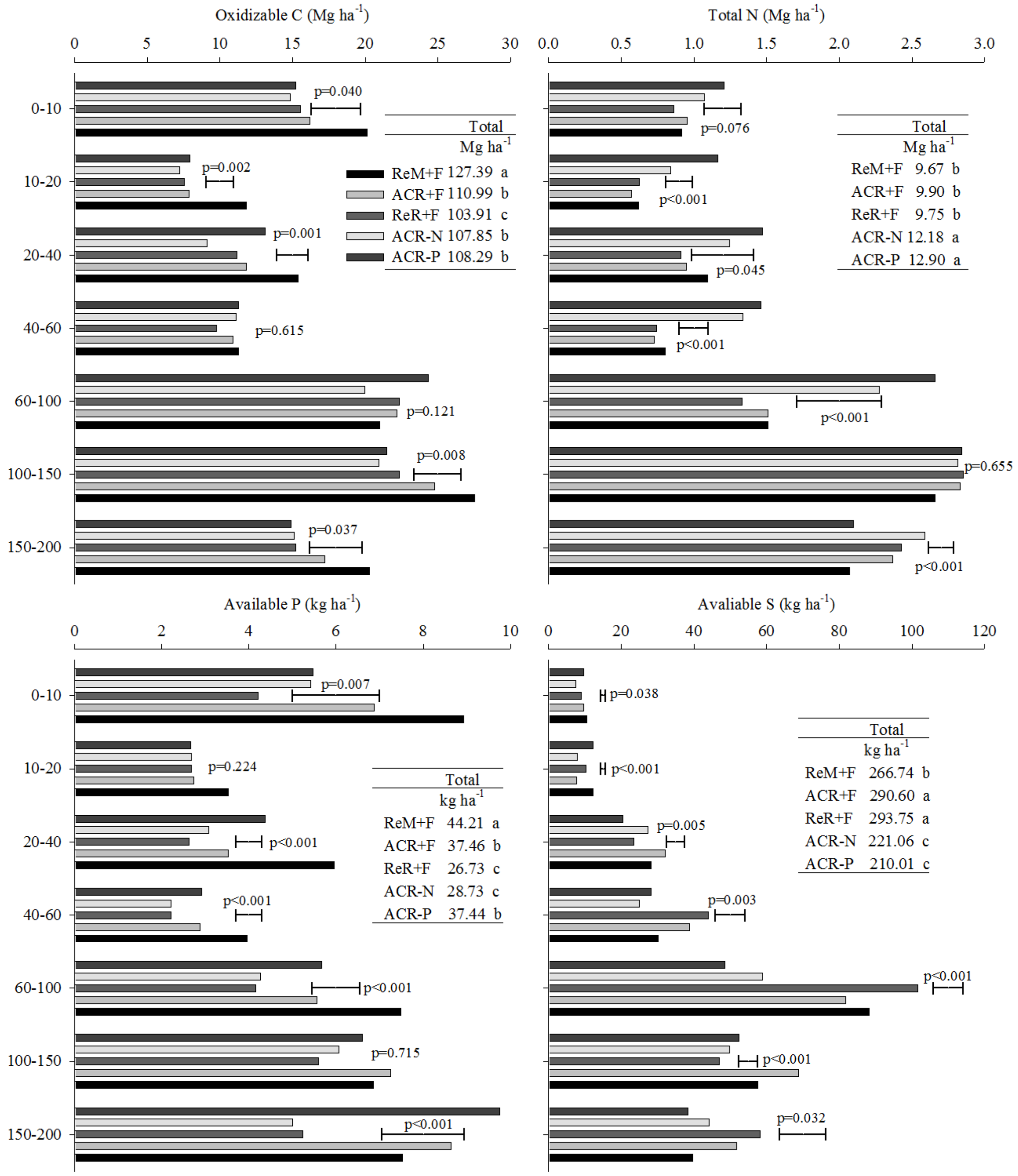

Figure 5.2. Stocks of oxidizable C, total N, available P and S to 2 m depth in a *Eucalyptus* plantation under different harvest residue management and fertilizer application strategies$^{(*)}$. The bars indicate the least significant difference based on the LSD test at 5% probability, and the values next to the bars show the significance of the F test.

$^{(*)}$ ReM-F = harvest residues maintained on the soil without fertilization, ACR+F = harvest residues partially removed from the area plus fertilization, ReR+F = harvest residues removed from the area plus fertilization, ACR-N = harvest residues partially removed from the area plus fertilization except N fertilizer, ACR-P = harvest residues partially removed from the area plus fertilization except P fertilizer.

5.3. Effects of harvest residue strategies on available nutrient stocks

The effect of forest residue removal on soil nutrients stocks is inconsistent between different studies in the literature. In some cases, no losses in soil C or nutrient stocks have been reported, even with increases in nutrients exported during harvest (Du Toit, 2008; Nambiar and Harwood, 2014; Laclau et al., 2010; Mendham et al., 2014; Mendham et al., 2003; Huang et al., 2013; Kumaraswamy et al., 2014). This observation has been attributed to several mechanisms: high buffer capacity of the soil, slow decomposition of forest residues, a long harvesting return interval (more than 7 years), and fast growth and litter deposition from new *Eucalyptus* plantations. However, reduction in soil C and nutrient stocks has been observed in wet tropical sites with sandy soils, high productivity forests, and successive harvest treatments (Rocha et al., 2016; Peng et al., 2008; Laclau et al., 2010; Mendham et al., 2003; Kumaraswamy et al., 2014). This study agrees with this latter set of studies and shows substantial changes in soil C and nutrients following removal of harvest residues.

In this study, the highest stocks of oxidizable C in the soil were observed in the 0-20 cm soil layer of the ReM+F treatment, followed by ACR+F and ReR+F treatments ($p>0.001$). Similar results were observed by Moreno-Fernandez et al. (2015) in Mediterranean mountain Scots pine forests, where the highest stocks of C were in the 0-20 cm layer of the soil under moderate intensity management.

Stocks of soil C decreased as soil depth increased, with the lowest stocks being observed in the ACR-P treatment ($p>0.006$) at 150-200cm depth. In a previous study developed in the same experimental site, Rocha (2014) found that the removal of forest harvest residues from the soil reduces the oxidizable organic C from the surface layer of the soil by 50%, and 75% of this reduction happens in its labile fractions. Vanguelova et al. (2010), on the other hand, concluded that there was no evidence that whole-tree harvest decreased soil organic C in a 28-year old second rotation stand of Sitka spruce in the UK. Slash removal has larger effects on soil C and nutrients when rotations are short, slash is removed repeatedly, clay content is low, temperatures are high, the site is relatively wet, and forest productivity is high (Du Toit, 2008; Gonçalves et al., 2002). It is well known that retaining harvest residues on the soil is extremely important to maintain high productivity of tropical forests (Nambiar and Harwood, 2014; Achat et al., 2015; Rocha et al., 2016). However, more research is needed in order to reach a plausible conclusion about the subject due to the divergence in the data available, as observed by Nambiar and Harwood (2014).

The forest residue removal for two rotations resulted in the cumulative loss of 23.5 t ha^{-1} of C from 0-200 cm soil depth (ReM+F and ReR+F treatments). This reduction was more concentrated in the top 40 cm (Figure 2). This effect is a result of the additional removal of 80 t ha^{-1} of biomass (slash and litter; 51 and 29 t ha^{-1} in R1 and R2 respectively) in the ReR+F treatment. Furthermore, the reduced initial growth in the ReR+F treatment during both rotations resulted in less litter deposition (Hubner, 2015). In the second rotation, a 40% reduction in fine roots was observed in the 0-30 cm layer of the ReR+F treatment (data not shown), which also contributed to low soil C content.

Forest residue removal resulted in a small reduction in the N stocks in the 0-20 cm layer in 2012 (Figure 5.1). However, no differences were observed between ReR+F and ReM+F treatments in 2016 (Figure 5.2) despite the higher harvest output in the ReR+F treatment. This can be attributed to the larger accumulation of N in the live biomass in the ReM+F treatment, which was 33% higher when compared to the ReR+F treatment. This aligns with other long-term productivity studies that find that subsequent tree growth following stem only harvest tends generally to be higher than whole-tree harvest due to the maintenance of harvest residues on the soil (Smolander et al., 2010).

The available P stock was found to be larger in the ReM+F treatment when compared with ReR+F treatment. This result could be caused by to two main factors: i) less ReM+F nutrient outputs by harvest and ii) larger quantities of organic matter in ReM+F which reduces the fixation of P on the soil colloids, thus improving its availability. No differences were observed between treatments after 100 cm in depth. In a long-term productivity experiment conducted in North Wales, UK, by Walmsley et al. (2009), the alterations in soil characteristics and their effects were tested using different harvest treatments (whole-tree harvest and bole-only harvest) on 23-year-old second rotation stands of Sitka spruce (*Picea sitchensis*). The authors found that the whole-tree harvest treatment is responsible for the depletion of three to four times greater quantities of N, P and K than the conventional bole-only harvest in the first rotation.

ACR+F and ReR+F treatments contained the largest stocks of S in the 40-100cm layer in the soil, being statistically different when compared to others. This can be attributed to the lower concentration of SOM of the previously mentioned treatments. Low OM in highly weathered soils results in a higher quantity of positive charges, especially in the B horizon (Brady and Weil, 2013), with consequent retention of SO_4^{2-} in the soil.

Repeated removal of forest harvest residues is likely to reduce oxidizable C, available N and total P stocks in the soil under *Eucalyptus* plantations. The use of harvest residues as soil coverage can protect the soil against erosion, improve or maintain the SOM, and improve both the quantity and availability of nutrients stored in the soil (Gonçalves et al., 2002; Rocha et al., 2016b; Achat et al., 2015).

5.4. Soil contribution to nutrient absorbed by trees

The N and P outputs in two rotations of cultivation of this experiment (2004 and 2012) were higher than the quantity applied via fertilization and atmospheric deposition. In this period, the net N balance for ReM+F, ACR+F, ReR+F and ACR-N treatments was -255, -516, -787, and -770 kg ha^{-1}, respectively. With regards to P, there was also a negative net balance of -24, -48, -70 and -128 kg ha^{-1} in the ReM+F, ACR+F, ReR+F e ACR-P treatments, respectively. The net balance was positive only for S, with approximately +120 kg ha^{-1} for all treatments with the exception of the ACR-N treatment, which contained a net balance of -128 kg S ha^{-1} (Table 5.2). N inputs through biological fixation are unlikely due to the absence of N-fixing weeds. In addition, one should not expect inputs of N from rock weathering as the soil was highly weathered (Ferralsol). The difference between nutrient losses due to harvesting and nutrients supplied via fertilization and atmospheric deposition must be made up for with nutrients absorbed from soil pools.

From 2004 to 2012, a reduction was observed in the N stocks in the soil in the 0-20 cm layer for all treatments (Figure 5.1). The ReR+F treatment was the most affected among the treatments due to the elevated nutrient outputs by harvest. For the ACR-N treatment, this was due to the small quantity of N applied via fertilization. In the ReM+F treatment, the 0-20 cm layer was responsible for providing 65% of the difference in the balance of soil N. The 0-20 cm layer was responsible for providing 35% of the difference in net soil N balance for the ACR treatments and 24% for the ReR+F treatment (Figure 5.1 and Table 5.2). The maintenance of harvest residues on the soil increases soil C and N pools – especially in labile fractions – increasing the capacity of upper layers to provide N to plants (Nambiar and Harwood, 2014; Achat et al., 2015; Rocha et al., 2016; Mendham et al., 2014). The remaining difference possibly came from deeper layers of the soil profile and factors such as biological fixation by symbiotic associations which were not examined in this study (Silva et al., 2014). Despite the large difference in the nutrient outputs between ReM+F and ReR+F treatments (more than 500 kg in the net balance), no differences in the N stocks between these treatments were found in the deeper layers of the soil (20-200cm). More research is necessary in order to fully understand the additional sources of N in such highly weathered, tropical forests.

Little reduction was observed in the stocks of available P in the 0-20 layer (Figure 5.1). The ACR-P treatment presented the highest reduction (2 kg ha^{-1}), while ReM+F treatment presented a slight increase. This happened even with the negative balance of the nutrient for all treatments evaluated. No reduction in soil P below 40 cm depth was observed during the study in 2016 (Figure 5.2). The small reduction in P content contradicts the highly negative net balance of this nutrient across treatments. P balance for all treatments was -60 kg ha^{-1} on average. ACR-P treatment had a deficit of -128 kg ha^{-1} while the reduction was only 6 kg ha^{-1} from 0-200 cm of soil depth. This discrepancy suggests that the *Eucalyptus* trees take up soil P hidden in fractions not extracted by the traditional resin method of analysis. *Eucalyptus* species can uptake organic and inorganic

P fractions by phosphatase and exudation of low molecular mass acids (Costa et al., 2016). More studies are necessary to clarify the complete origin of P absorbed by trees.

Even with a positive net balance of S stocks in the soil (Table 5.2), a small reduction was observed in the stocks of the nutrient when comparing the evaluations in 2004 and 2012. This is due to the elevated mobility of the element in the soil, especially in the upper layers (Bissani and Tedesco, 1988), along with its consequent migration to and accumulation in the 40-100cm layer (Figure 5.2) where it will adsorb to positive charges on soil surfaces exposed by losses of OM (Silva et al., 2014).

Table 5.2. Nutrient balance in two *Eucalyptus grandis* rotation under different timber harvest intensities and fertilizer application

Treatment(1)	N	P	S
		kg ha^{-1}	
		Net Nutrient Balance(2)	
ReM+F	-255	-24	148
ACR+F	-516	-48	135
ReR+F	-787	-70	111
ACR-N	-770	-48	-128
ACR-P	-490	-128	141
		Difference in soil nutrients stocks from 2004 to 2012 (0-20 cm)	
ReM+F	-164	0.8	-1.1
ACR+F	-189	-0.4	-1.3
ReR+F	-192	-0.5	-2.5
ACR-N	-219	-0.9	-1.4
ACR-P	-189	-2.0	4.0

(1) ReM-F = harvest residues maintained on the soil without fertilization, ACR+F = harvest residues partially removed from the area plus fertilization, ReR+F = harvest residues removed from the area plus fertilization, ACR-N = harvest residues partially removed from the area plus fertilization except N fertilizer, ACR-P = harvest residues partially removed from the area plus fertilization except P fertilizer;

(2) Differences between the inputs by fertilizer application and atmospheric deposition with the harvest outputs (Table 5.1 and Figure 5.1).

5.5. Management Considerations

Despite the small effect of whole-tree harvest on the soil nutrient stocks described by many authors (Du Toit, 2008; Nambiar and Harwood, 2014; Laclau et al., 2010; Mendham et al., 2014; Mendham et al., 2003; Huang et al., 2013; Kumaraswamy et al., 2014), this and other studies emphasize the importance of the maintenance of forest residues on the soil, especially in tropical sites with low buffer capacity soils, high productivity, and short cycle plantations (Nambiar and Harwood, 2014; Rocha et al., 2016; Laclau et al., 2010; Mendham et al., 2014; Mendham et al., 2003, Kumaraswamy et al., 2014). In these sites, removal of forest residues can result in loss of wood productivity of up to 40% due to the low nutrient pools remaining in the soil (Rocha et al., 2016).

In the past several years, an increasing incentive to the use of forest residues for bioenergy purposes has grown in Brazil. In many cases, models for the utilization of these residues are based on the models currently used in temperate regions, where soils often have higher buffer capacities and higher organic C contents. This study shows that in tropical conditions the use of forest residues for bioenergy purposes should be carefully considered, taking into consideration the conditions of each site. On steep sites and/or those with low buffer capacity, all forest residues should be retained on the soil in order to avoid soil erosion (Candido et al., 2014) and depletion of soil nutrient pools (Nambiar and Harwood, 2014; Rocha et al., 2016; Laclau et al., 2010; Mendham et al., 2014; Kumaraswamy et al., 2014; Costa et al., 2016). In sites with favorable conditions for residue removal, preference should be given to the coarse residues due to their high caloric power and reduced nutrient concentration. With the removal of forest residues, the application of high rates of fertilizer is necessary in order to avoid productivity losses and to ensure the sustainability of the silviculture system.

The high negative net balance, especially for N and P, and the relative low reduction in the availability of these nutrients in the soil draws attention in two main points. First, more attention should be given to production sustainability, mainly in sites harvested in the whole-tree harvest system. Second, more studies should be implemented to better understand the contributions of organic and inorganic (mainly to P) fractions of low lability on the supply of nutrients to trees. Our results and others (Laclau et al., 2010; Costa et al., 2016) suggest that *Eucalyptus* trees can access P fractions not identified by traditional soil analysis methods. Regarding N concentration, even with high harvest outputs (more than 500 kg ha^{-1} in each crop rotation) no response to N application by fertilization was found in terms of wood productivity in Brazilian conditions (Pulito et al., 2015). More research is essential to better understand the sources and the cycle of this nutrient within tropical soil-plant systems.

CHAPTER 6. CONCLUSIONS AND FUTURE WORK

This research focused on understanding the impacts of different forest management practices on the concentration of essential nutrients for plant growth and the relationship among them, such as P, C and N, in tropical areas cultivated with *Eucalyptus* species on a long-term basis. The retention of forest harvest residues can aid in the maintenance of soil physicochemical characteristics essential for maintaining the productivity of forests planted with medium and long-term species (Holub et al., 2013; Huang et al., 2015; Rocha et al., 2016).

Chapter 4 reports the concentrations of total and labile forms of P at depth in an Oxisol which was cultivated with *Eucalyptus* in Brazil over a 12-year-period. The cultivation was performed under different timber harvest intensities and different fertilizer applications.

Our results show that organic and inorganic P forms in the soil can act as source or drain to the soil solution, depending on the mineralogical composition of the soil, fertilization and harvest management applied. From 2004 to 2016, a reduction in the relative contribution of non-labile P in the ReM treatments was observed. However, this fraction of P still accounts for more than 60% of total P present in the soil, followed by labile and moderately labile forms of the nutrient. The highest stocks of total P were observed at 0-10cm. At the end of the first crop rotation of the experiment (2012), the ReM-F, ReR+F and the ACR+F treatments presented the highest values for the nutrient.

Chapter 5 documents the stocks of available P and S, total N and oxidizable C at depth in an Oxisol cultivated with *Eucalyptus* in Brazil over 12 years, under different timber harvest intensities and fertilizer application. Calculations were also performed in order to determine the balance of the relationship between N, P and S exported by harvest, and their stocks in the soil during each plant cycle.

A reduction in N, P and S stocks in the 0-20 cm layer in all treatments was also observed. For N, this reduction was 20% higher when harvest residue was removed from the soil, and 40% higher when no N fertilizer was applied. The maintenance of harvest residues on the soil surface reduces the exportation rates of N, P and S by 120%, 50%, and 40% respectively. Low OM in highly weathered soils results in a higher quantity of positive charges, especially in the B horizon (Brady and Weil, 2013), with consequent retention of SO^{4-} in the layer.

6.1. Implications for management and future research

Improving the understanding of the controls on P distribution in highly weathered soils, and their relationship to the concentrations of C and N within those soils, can strengthen models used in management and policy decisions.

Implementing cultivation methods that retain crop residue on the soil surface may benefit the soil-plant system by improving the physicochemical properties of the soil. Plants cultivated on sites that were previously treated with harvest residue may benefit from this nutrient management approach due to the higher OM content in the soil. Increased OM content increases cation exchange capacity (CEC), improves aggregation, water-holding capacity, infiltration, microbial diversity, and pH-buffering capacity of soils therefore improving site productivity. The protective layer formed by the harvest residues also reduce nutrient leaching and erosion rates which further decreases the environmental stress following timber harvest and fertilizer applications

Higher quantities of SOM resulting from the retention of harvest residues on the soil surface increase P availability to plants (Maranguit et al., 2017). The addition of organic substances to the soil serves as a C source that enhances microbial biomass and phosphatase enzyme activity, thus helping with the transformation of occluded P into a readily available form suitable for plant uptake. Activities of acid or alkaline phosphatase are known for controlling the release of biologically available nutrients from organic compounds of P.

By using techniques which allow for the determination of the different fractions of P in the soil, a broader understanding of the transformations of nutrient in the soil may be reached. This can be accomplished not only by quantifying the labile P, but also the fractions of lower lability between the labile P and total P in the soil.

Conducting this work will enable the conduct of adequate forest management based upon the soil type. It will also aid in maintaining the physicochemical properties of the soil, the adequate growth of the plant species established in the site and, most importantly, the optimization of fertilizer application (e.g. N, P). The efficient use of fertilizer is essential in order to preserve soil properties, to decrease the total energetic balance of the wood production process, and to prevent the degradation of the surrounding environment which all people rely on for maintaining biodiversity, natural resource use, and recreational pursuits.

The results obtained in this research will enable the expansion of forest research performed in Brazil and will contribute to the development and improvement of forest management techniques favorable to the sustainability of plant species cultivated in tropical ecosystems.

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