

**Impacts of Fertilization on Soil Properties in Loblolly Pine Plantations in the Southeastern United States**

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## **Abstract**

We examined the effects of periodic nitrogen (N) and phosphorus (P) fertilizer applications on the O horizon and mineral soil in loblolly pine (*Pinus taeda L.*) plantations over a 12-year period. To accomplish this, we used 9 experimental sites located across the south, which were grouped using the CRIFF Classification System. Group 1—CRIFF A, B (poorly-drained Ultisols); group 2—CRIFF C, D, G (sandy Spodosols and Entisols); and group 3—CRIFF E, F (well-drained Ultisols). Fertilization rates were 135, 202, and 269 kg N ha<sup>-1</sup> at 4 years application frequency. This resulted in a cumulative N application rate of 540, 808, and 1076 kg ha<sup>-1</sup>. P was added at 10% of the N rate. Fertilization increased the mass, N content, and P content of the O horizon in all soil groups. Fertilization did not impact mineral soil N. No significant increases in total N trends were observed to a depth of 1 m. Likewise, total inorganic N (NH<sub>4</sub><sup>+</sup> + NO<sub>3</sub><sup>-</sup>) was not affected by fertilization. These results suggest that N fertilization will have little effect on long-term soil N availability regardless of soil types. In contrast, fertilization increased extractable P in soil CRIFF groups 1, 2, and 3 by 26, 60, and 4 kg P ha<sup>-1</sup> respectively suggesting potential for long-term soil P availability and site quality improvement. However, the low extractable P in soil group 3 implies additional fertilization with P for the next rotation for sites included into this soil group.

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## Chapter 1: Introduction

The southeastern United States is one of the largest timber producing regions in the world (Prestemon and Abt, 2002; Allen et al., 2005; Fox et al., 2007b). Forest productivity has increased due to the implementation of intensive silvicultural systems that include site preparation, genetic improvements, competition control, and fertilization (Jokela, 2004; Fox et al. 2007a). However, soil nutrient deficiencies particularly nitrogen (N) and phosphorous (P) often limit productivity of loblolly pine throughout the South (Allen, 1990; Fox et al., 2007a). To ameliorate soil nutrient deficiencies, fertilization is a common silvicultural practice in the south. The use of fertilizers has increased since 1969 due to positive biological and economic responses of loblolly pine stands to fertilizer applications (Albaugh et al., 2007). Significant increases in leaf area and consequently tree growth have been achieved following fertilization with N and P (Albaugh et al., 2004; Jokela, 2004; Allen et al., 2005; Fox et al., 2007a; Albaugh et al., 2014).

Fertilization often has greater impacts on growth of loblolly pine stands compared to other silvicultural treatments (Carlson et al., 2006). The duration of the response following fertilization with N is generally short lived while the response following P fertilization is long lived (Fox et al., 2007a). The longevity of the fertilizer response may be related to the impact of fertilization on soil nutrient availability. Mudano (1986) found only temporary increases of N availability in the mineral soil following N fertilizer in sandy textured soils. Kiser and Fox (2012) found little impact on soil N in sandy soils even after repeated N fertilization that added over 1000 kg N ha<sup>-1</sup> over 20 years. Miller (1981) hypothesized that because N fertilization has little impact on N availability in the mineral soil, a long-term growth response is unlikely. In contrast, long-term increases in soil P following P fertilization have been found in both slash pine and loblolly pine systems in the South (Harding and Jokela, 1994; Miller and Fox, 2011;

Fox et al., 2011). Therefore, because P fertilization has a long-term impact on soil P availability, P fertilization will lead to a sustain growth response and an increase in long-term site quality (Pritchett and Comerford, 1982; Phelan and Allen, 2008; Fox et al., 2011, Kiser and Fox, 2012).

There are a variety of soils and site types in the southeastern United States and responses to fertilization are site specific and soil properties must be taken into consideration to develop optimal prescriptions (Fox, 2000; Allen et al., 2005; Fox et al., 2007a). To adequately make silvicultural decisions, the CRIFF soil groups were developed in the South. The CRIFF classification system divides soils into 8 major classes based on the soil drainage class, texture, and depth to the B horizon (Fisher and Garbett, 1980). The CRIFF groups are an effective tool that can be used to identify sites that will respond to forest fertilization (Fisher and Garbett, 1980; Fox, 2004b; Jokela, 2004). For example, growth responses following fertilization with 168 to 224 kg N ha<sup>-1</sup> and 28 to 56 kg P ha<sup>-1</sup> in midrotation stands lasted for approximately 6 to 10 years (Fox et al., 2007a). N and P application raised wood volume by 3.5 to 4.2 m<sup>3</sup> ha<sup>-1</sup> year<sup>-1</sup>, which was a 25% of growth increase.

Descriptions for the soil CRIFF groups are depicted in Fig. 1 and given in more detail by Fox (2004b). Sites that are included in soil CRIFF group A and B are found in the wet environments where the water table is high all year around. These soil types are described as very poorly to somewhat poorly-drained soils. The B horizon in these soils has a fine argillic material (clayey subsoil). In CRIFF A the depth of the argillic horizon occurs in the 20 inches whereas in CRIFF B the argillic horizon is deeper than 20 inches. Soil CRIFF group C and D are very poorly to moderately well-drained soils, which have a spodic horizon that is composed of organic matter with relatively high iron and aluminum contents. In this case, the argillic horizon is found beyond the spodic horizon in these sandy-textured soils. Soil CRIFF group E and F are

well-drained soils and are fine textured clays. CRIFF E has an argillic horizon in the first 20 cm depth and for the CRIFF F the argillic horizon is deeper than 20 inches. The CRIFF G is a well to excessively drained sandy soils that lacks the B horizon in the profile. Lastly, CRIFF H is a rich soil in organic matter contents that are very poorly-drained.

Because of the variety of soil types across the South responses to fertilization with N and P may be affected by soil properties that influence N and P retention or losses that may occur through processes of volatilization (Zerpa and Fox, 2011; Kissel et al., 2013; Elliot and Fox, 2014), denitrification in waterlogged soils (Lensi and Chalamet, 1982; Groffman and Tiedje, 1989; Sirivedhin and Gray, 2006), and leaching of both N and P from coarse-textured sandy soils (Lee and Shibu, 2005; Davis et al., 2012). Fine-textured soils retain more N and P following forest fertilization than coarse-textured soils because fine-textured soils tend to have higher specific anion adsorption capacity and cation exchange capacity (Amato and Ladd, 1992; Will et al., 2006) and greater amounts of nutrients are expected in these soil systems.

In this study, we examined the impact of repeated fertilization with N and P on O horizon and mineral soil N and P in a regional fertilization study in the southeastern United States. Our main objectives were to determine the accumulation of N and P in the O horizon and mineral soil following different rates of fertilizer applications. Because the CRIFF soil groups are based on texture and drainage class, two soil properties related to N and P retention, they may help identify sites where long-term responses to N and P fertilization are likely to occur. We hypothesized that fine-textured soils (CRIFF A, B and E, F groups) would retain more N and P compared to coarse textured soils CRIFF (C, D, and G groups).

## **Research Objectives**

The major objectives in this research are to:

1. Determine the accumulation of N, P and other nutrients in the O horizon and mineral soil in experimental sites with different N and P rates of fertilizer.
2. Determine if soil texture and soil drainage affect long-term soil N and P availability following repeated fertilization in the South.

## **Hypotheses Tested**

The following null hypotheses were tested:

1. HO<sub>1</sub>: Different fertilization treatments do not result in different long-term accumulations of soil nutrients.
2. HO<sub>2</sub>: The texture of soils (clay vs. sandy textured) does not have a significant impact on the accumulation of nutrients in the soil in the long-term.
3. HO<sub>3</sub>: Drainage class does not have a significant impact on accumulation of soil nutrients.

## Chapter 2: Literature Review

### Forest Fertilization

The demand for industrial wood has increased because the world's population has increased (FAO 2009). Therefore, it is essential to increase forest production on existing forest areas to meet these new demands. The southeastern United States is one of the largest timber producing regions in the world (Prestemon and Abt, 2002; Fox et al., 2007b). Loblolly pine plantations in the South increased from less than a million hectares in 1952 to around 13 million hectares by the end of the 20<sup>th</sup> century (Fox et al., 2007b). Forest productivity has increased as intensive silvicultural systems such as site preparation, genetic improvements, competition control, and fertilization have been implemented (Jokela, 2004; Fox et al. 2007a). These practices are needed to increase forest production and consequently maintain long-term soil productivity (Fox, 2000).

Fertilization with N and P is commonly used to ameliorate soil nutrient deficiencies and thus increase growth in pine plantations in the South (Fox et al., 2007a). P fertilization has been shown to have a long-term impact on soil P availability and stand growth that may last more than 25 years (Humphreys and Pritchett, 1971; Pritchett and Comerford, 1982; Fox et al., 2011; Kiser and Fox, 2012). In contrast, N fertilization does not appear to have long-term impact on soil N availability or stand growth (Miller, 1981; Phelan and Allen, 2008; Kiser and Fox, 2012). However, researchers have suggested that there is a potential for long-term soil N accumulations following fertilization (Binkley and Reid, 1985; Will et al., 2006) Long-term growth responses have been observed in some forests, such as Douglas-fir at Wind River Experiment Forest in Washington (Binkley and Reid, 1985). Therefore, additional research is needed to determine

whether N and P fertilizer applications will have long-term effect on soil nutrient availability and site quality on different soil types across the South.

Temporary increases of nutrient availability in the mineral soil are measurable after N fertilizer additions; usually lasting about 5 months in sandy textured soils (Mudano, 1986; Elliot 2006). Kiser and Fox (2012), in a study conducted with loblolly pine and sweetgum in sandy-textured soils in North Carolina and Georgia, found that N fertilization had little long-term impacts on soil N availability. However, the effect of nitrogen fertilizer may be different in clay and loamy textured soils. It has been suggested that fine-textured soils retain more C and N than coarse-textured soils (Amato and Ladd, 1992; Will et al., 2006). The increase cation exchange capacity of clay and loamy soils with high organic matter may increase N availability in the long-term (Silver et al., 2000). We suggest that further investigations need to be done in order to answer the question about whether fertilization with N has long-term impacts on nutrient availability in the soil and consequently site quality improvement.

Fertilization with P, on the other hand, has shown to have long-term impacts on mineral soil P availability. For instance, a single application at the time of planting lasted for 17 to 20 years after in slash pine plantations in Florida (Pritchett and Comerford, 1982). Kiser and Fox (2012) found P accumulation in sandy soils following fertilization in South Carolina for over 20 years. Similarly, a study by Harding and Jokela (1994) in Franklin County, Florida has demonstrated that following fertilization there is a potential for long-term P availability in the soil after 25 years increasing site organic matter on the O horizon and mineral soil. P accumulation in the mineral soil may be associated with soil properties to retain P. Clay particles, Fe and Al coating of sand particle can sorb large quantities of P (Fox et al., 2011; Arai and Levi, 2013; Yaghi and Hartikainen, 2013).

## **Nutrient Limitations**

Nutrient limitations develop when the soil is not capable of meeting the stand's requirements Allen (1990). In the southeastern United States N and P are the primary nutrient deficiencies (Fox et al., 2007a). Immediately after harvesting, N availability in the soil is high because environmental conditions promote rapid decomposition of residual slash and organic horizon (Vitousek and Matson, 1985a; Fox et al., 1986). As the organic horizon decomposes it releases large amounts of N so that small seedlings have enough nutrients in the soil to meet their needs. However, as the stand grows the demand increases while the soil N supply declines and nutrient deficiencies often develop around the time of crown closure (Fox et al 2007a). Vitousek et al (1985b) found that by age five years, N concentrations in a site-prepared Piedmont plantation were comparable with those in mature stands. Under these conditions, soil N availability is not sufficient to sustain rapid stand growth (Allen, 1990; Piatek and Allen, 2001). Large growth responses occur when these stands are fertilized (Fox et al, 2007a).

## **Growth Response in Leaf Area (LAI)**

Biomass production is largely determined by the amount of light intercepted by the tree canopy (Linders, 1987; Cannell, 1989). Nutrient deficiencies can lead to reduced leaf area and thus decrease growth (Vose et al., 1994; Albaugh et al., 1998; Vose and Allen, 1998; Albaugh et al., 2004; Fox et al., 2007a). Over 100 kg ha<sup>-1</sup> of N and 10 kg ha<sup>-1</sup> of P need to be available annually for trees in order to maintain high levels of leaf area required to maximum volume production (Ducey and Allen, 2001; Battaglia et al., 2004). Fertilization can ameliorate nutrient limitations and lead to greater leaf area and consequently increased stand volume production in loblolly pine plantations in the southeastern United States (Fox, et al., 2007a). Albaugh et al

(1998) found out that by increasing soil nutrient availability both leaf area and growth were increased.

Jokela et al (2004) and Rojas (2005) found that fertilization has the potential to increase nutrient availability in the soil, which leads to a larger production of leaf area and stand growth. The study by Rojas (2005) was conducted in Georgia and Florida in loblolly pine plantations where fertilization increased stand volume from 32 m<sup>3</sup> in untreated plots to over 84 m<sup>3</sup> ha<sup>-1</sup> in fertilized plots in a 5-year period. This suggested a strong relationship of volume growth and leaf area. Albaugh et al (2004) reported average N levels of 1.35% in foliage in loblolly pine plantations in North Carolina following an annual N application of 100 kg ha<sup>-1</sup>. This study was conducted in a 9-year period in which fertilized plots showed significant increases in LAI compared with the controls. At the age of 16, the biomass was three times greater in the fertilized plots over the controls while the basal area was 2 times greater.

### **Growth Responses in Diameter, Height, and Volume**

Increases in growth, diameter, height, and volume have been observed following fertilization in the South. On nutrient deficient sites, fertilization with 224 kg of N and 28 kg of P ha<sup>-1</sup> increase growth of loblolly pine around 3.8 m<sup>3</sup> ha<sup>-1</sup> year<sup>-1</sup> over an 8 to 10 year period (Fox et al., 2007a). Typical increases from 2.4 and 3.0 m in tree height has been observed following P fertilization on sites with P deficiencies at the end of a 25-year rotation (Gent et al., 1986; Everett and Palm-Leis 2009). Will et al (2002) reported the effect of annual fertilization on height, diameter, and basal area between control and fertilized treatments from 14.8 to 19.3 m, 14.5 to 19.3cm, and 27.6 to 42.6 m<sup>2</sup> ha<sup>-1</sup> respectively.

Albaugh et al (2009) reported significant increases in averaged volume growth for loblolly pine at four sites with sandy soils in the southeast following fertilization with 224 and 28

kg of N and P applied one or two times. The average increase in volume growth was around  $7 \text{ m}^3 \text{ ha}^{-1} \text{ year}^{-1}$ , which surpassed the average volume growth response of ( $3.8 \text{ m}^3 \text{ ha}^{-1} \text{ year}^{-1}$ ) found in the South for loblolly pine. A combination of fertilization and vegetation control was more efficient than fertilization or vegetation control alone resulting in a significant growth response at 7 out of 13 sites examined in the southeastern United States (Albaugh et al., 2012). Further, cumulative fertilizer additions in loblolly pine after 16 years of treatment, fertilization increased diameter, height, basal area, and volume by 5.2 cm (30%), 4.3 (31%),  $15 \text{ m}^2 \text{ ha}^{-1}$  (54%), and  $171 \text{ m}^3$  (92%) respectively over the control treatments (Albaugh et al., 2014). These results indicated that potential for increasing wood productivity in the South that offers landowners attractive financial returns (Yin and Sedjo 2001; Fox et al., 2007a).

## **Impacts of Fertilization on Soil Properties**

### *Long-term Effects on Mineral Soil N and P*

Urea is the most commonly used N fertilizer in forest plantations in the South (Pritchett and Fisher, 1987). Following application, urea is transformed into plant-available inorganic forms  $\text{NH}_4^+$  and  $\text{NO}_3^-$  in the soil (Gould et al., 1986). Urea is first hydrolyzed by the well-characterized enzyme urease (Estiu and Metz, 2004) to  $\text{NH}_4\text{HCO}_3$ , which dissociates the release of  $\text{NH}_4^+$  and  $\text{HCO}_3^-$ . This results in a short-term increase in available N. Following hydrolysis of the urea large amounts of N in the mineral soil can be lost via several mechanisms including volatilization (Cabrera, 2001; Elliot and Fox, 2014), denitrification (Groffman and Tiedje, 1989; Weier et al., 1993; Lowrance et al., 1995; Mohn et al., 2000), and leaching (Lee and Shibu, 2005).

Miller (1981) did not find increases in soil N in sandy-textured soils following N fertilizers. Kiser and Fox (2012) in a study conducted in North Carolina and Georgia concluded

that N fertilization had little long-term impact on mineral N in sandy soils. Miller (1981) argued that fertilization would likely increase mineral soil N only if the amount of nutrient applied is significantly larger than to the total soil N content. However, increases in N in the mineral soil have been reported where the amount of N applied to the soil was much less than the soil capital (Binkley and Reid, 1985; Will et al., 2006). Binkley and Reid (1985) found long-term increases in soil N availability that increased growth of Douglas-fir for over 15 years growing on a gravelly loam soil surface with high organic matter content. Even though the N applied as ammonium nitrate was only the 13% of the total N site capital, growth in the fertilized plots was twice of the controls. Will et al (2006) evaluated the effects of annual fertilization on loblolly pine plantations growing on somewhat poorly-drained Ultisols and Spodosols and well-drained loamy soils in the Lower Atlantic Coastal Plain of Georgia. They added 900 kg N ha<sup>-1</sup> as diammonium phosphate and ammonium nitrate, which was around 40% of the site capital. That increased extractable NH<sub>4</sub><sup>+</sup> and NO<sub>3</sub><sup>-</sup> and total N concentration from 0.55 to 0.71, 1.6 to 3.8 and 1.9 to 1.8 g kg<sup>-1</sup> respectively at 0-10 cm depth. Results from the studies by Binkley and Reid (1985) and Will et al (2006) differed from those reported by Kiser and Fox (2012) in N availability on the soil following fertilization. This may be due to the differences in soil properties such as drainage class and texture. Soils with higher clay and organic matter content like the sites studied by Binkley and Reid (1985) and Will et al (2006) may retain more added N in contrasted with well-drained sandy soils in the study done by Kiser and Fox (2012) which had low capacity to retain NO<sub>3</sub><sup>-</sup> and NH<sub>4</sub><sup>+</sup> ions.

P tends to accumulate in both O horizons and mineral soil (Kiser and Fox, 2012; Kiser et al., 2013). Fertilization with P has been found to have long-term impacts on soil P availability and consequently site quality improvement to sustain tree growth from one rotation to the next

(Pritchett and Comerford, 1982; Harding and Jokela, 1994; Fox et al., 2011). Kiser and Fox (2012) also found P increases in mineral soil following fertilization on a sandy soil in North Carolina. Fertilization almost doubled P availability in the soil (from 55 to 103 kg ha<sup>-1</sup>) due to the P fixation capacity in forest soils. P remains in the surface soil in fine-textured soils since P leaching in these soils is very low (Walker and Syers, 1976; Frossard et al., 1989).

Soil texture and drainage class may play a different role in P retention. It is often assumed that sand fractions with low surface area are less reactive than clay fractions with large surface area. However, in forest ecosystems sand fractions are coated with amorphous Fe and Al minerals and organic matter (Fuller et al., 1996; Penn et al., 2001) creating an important sink for P (Arai and Livi, 2013). Arai and Livi (2013) concluded that available P is readily retained by the sand fractions that are coated with Fe and Al that control the fate and transport of P.

#### *O horizon N and P*

The O horizon is a source of organic matter that decomposes gradually and becomes mixed with the mineral soil and subsequently forms the soil organic matter fraction (Pritchett and Fisher, 1987). Climate, soil type, species of trees, and as well as age of the forest influence O horizon development, which is critical to sustain nutrient cycles (Pritchett and Fisher, 1987). The O horizon accumulates leaf-nutrients in undisturbed loblolly pine plantations and acts as a sink for N and P (Piatek and Allen, 2000; Sanchez, 2001; Kiser and Fox, 2012). Miller (1981) affirmed that the majority of the N is retained in the O horizon in plantations of the temperate and boreal zone. However, after harvesting, the O horizon decomposes quickly and releases large amounts of nutrients into the mineral soil in a phenomenon called the Assart flush that temporarily increases N availability (Kimmins, 1997; Vitousek and Matson, 1985b).

Although fertilization often increases the O horizon in loblolly pine plantations, N and P concentrations in the O horizon may be small. This is because nutrients like N and P move from old to new foliage before litterfall (Gough et al., 2004; Albaugh et al., 2008;). Researchers have shown that 20 to 60% of N required for stand growth came from retranslocation (Nambiar and Fife, 1991; Binkley et al., 1995). However, significant increases in N and P content may be observed on the O horizon due to the high biomass as a result of fertilizer applications that compensates low nutrient concentrations. For example, Rojas (2005) reported increases in O horizon by 227% on fertilized plots over the controls in the Flatwoods of Georgia and Florida. Also, Kiser and Fox (2012) reported increases in N and P in the fertilized plots over the controls by 121 and 154% respectively in sandy soils in North Carolina. This demonstrates that the O horizon is capable of accumulating N and P.

### **Soil Processes Affecting N and P**

#### *Mineralization*

Mineralization is a process by which the soil complex organic materials are broken down by microbes into available N for plant uptake (Schimel and Bennett, 2004). Mineralization supplies  $\text{NH}_4^+$  in the soil (Müller et al., 2003). N in the soil is often in organic forms that are unavailable for plant uptake (Johnson et al., 1980; Foster et al., 1985; Gould et al., 1986). Most of the N in soils is tied up in soil organic matter that must be mineralized before it becomes available to plants. N transformations depend on the C:N ratio, which determines mineralization or immobilization. N immobilization can occur in forest soils with high C:N ratios (Gould et al., 1986). N is released by microorganisms when C:N ratio is less than 15:1 and N is immobilized when the C:N ratio is over 30:1.

Fertilization appears to increase net N mineralization in the mineral soil (Johnson et al., 1980; Maimone et al., 1991; Fox, 2004a; Gurlevik et al., 2004). Gurlevik et al (2004) assessed net N mineralization in loblolly pine plantations growing on well-drained soils in the Piedmont, North Carolina following fertilization with 224 kg N ha<sup>-1</sup>. To measure N mineralization, tests in both laboratory and fieldwork were performed from for a 3-year period. Incubations in laboratory showed N mineralization that ranged from -1 to 24 mg N kg<sup>-1</sup> while N mineralization in the field ranged from -5.0 (net immobilization) to 14 kg ha<sup>-1</sup>. Results from laboratory and field mineralization were positively correlated corroborating that fertilization increased N mineralization with higher mineralization rates in summer than winter. Similarly, Fox (2004a) reported potential N mineralization after repeated N fertilization on Douglas-fir starting in a 6-year period. However, increases in N mineralization were observed following a quadratic relationship at rates less than 450 kg N ha<sup>-1</sup>, then as fertilization rate surpassed this amount, the mineralization rate decreased. Fox (2004a) attributed this to changes in organic matter that led to reduce the efficiency of extracellular enzymes and therefore there was reduction in the rate of decomposition and mineralization.

### *Nitrification*

Nitrification is a process whereby NH<sub>4</sub><sup>+</sup> is transformed into mobile ions NO<sub>2</sub><sup>-</sup> and NO<sub>3</sub><sup>-</sup> (Müller et al, 2003; Hwang and Oleszkiewicz, 2007). Nitrification can be carried out either by autotrophic, chemoautotrophic, or heterotrophic organisms. Heterotrophic nitrifiers are capable of oxidizing NH<sub>4</sub><sup>+</sup> and organic N (Müller et al., 2003). Chemoautotrophic and Chemolithotrophic bacteria obtain their energy from oxidation of inorganic (NH<sub>4</sub><sup>+</sup>) compounds. Soil physical and chemical properties may affect nitrification. For instance, nitrification is high in soils near field capacity moisture and fine-textured sands. However, it appears to be low in soils saturated with

water because the lack of oxygen and nitrification stops in dry soils due to the lack of moisture (Sahrawat, 2008). Also, Sahrawat (2008) indicates that nitrification takes place in soil at pH ranging from 5.5 to 10.0. However, nitrification rates have been found in soils with pH levels below 3.8 (Tisdale and Nelson, 1970).

N fertilization with urea leads to nitrification in loblolly pine plantations (Johnson and Todd, 1988; Raison et al., 1990). Nitrification may increase as fertilization rates increase. In a the study by Johnson and Todd (1988) two fertilization regimes were tested: (i) 100 kg N ha<sup>-1</sup> in a single application per year and (ii) 100 kg N ha<sup>-1</sup> year<sup>-1</sup> of urea-N divided into four doses per year. The results from this experiment indicated that increasing frequency of fertilization applications increased nitrification. Similarly, Müller et al (2003) observed nitrification rates almost 20-fold in a slurry treatment, but almost no changes were observed in the control treatments. Nitrification rates are a function of temperature (Hwang and Oleszkiewicz, 2007). Nitrification was reduced by 20% as temperature was reduced from 20 to 10°C. This suggests that nitrification is a temperature-sensitive step in the biological process that occurs as temperature increases preferably during summer conversions. However, the nitrification rate may drop significantly as temperature decreases and may not occur during temperatures below freezing in the winter season. According to Müller et al (2003), transformations of NH<sub>4</sub><sup>+</sup> into mobile ions such NO<sub>3</sub><sup>-</sup> is prone to either leaching or denitrification having a high ecological significance.

### *Denitrification*

Denitrification converts N into gaseous forms and occurs under reducing conditions when NO<sub>3</sub><sup>-</sup> becomes a terminal electron acceptor and is reduced to N<sub>2</sub>. Denitrification often occurs due to the lack of oxygen in aquatic sediments with fully saturated soils (Groffman et al., 2009). This

is a poorly understood process in terrestrial N cycle in many forest ecosystems (Wexler et al., 2014). Denitrification can help reduce groundwater pollution like the ability of some wetlands to reduce nitrate-nitrogen into the atmosphere (Sirivedhim and Gray, 2006). Fertilization with urea stimulates nitrification and increases the likelihood for subsequent denitrification and N loss into the atmosphere (Johnson and Todd, 1988; Raison et al., 1990; Müller et al., 2003). This may help control nitrate movement into rivers and streams. However, N lost into the atmosphere through denitrification is not available in the soil to supply N for stand growth.

Fertilization with urea may affect denitrification differently depending on the properties of the mineral soil, particularly drainage class and texture. For example, Groffman and Tiedje (1989) used nine forest soils to estimate denitrification rates. They reported annual N loss to denitrification ranging from  $< 1 \text{ kg N ha}^{-1} \text{ year}^{-1}$  in a well-drained sand soil and over  $40 \text{ kg N ha}^{-1} \text{ year}^{-1}$  in poorly-drained clay loam soils. Lowrance et al (1995) reported average annual denitrification rates of  $68 \text{ kg N}_2\text{O-N ha}^{-1} \text{ year}^{-1}$  in a restored riparian wetland. Similarly, Weier et al (1993) observed that denitrification rates increased as soil texture became finer on the order of sand  $<$  loam  $<$  silty loam  $<$  silty clay loam. Further, increasing temperature and pH and reducing redox potential may trigger high N losses by denitrification (Mohn et al., 2000).

### *Volatilization*

Urea is the most common and preferred source of fertilizer used to supply N nutrients for its high N content (46%) and affordable cost (Gould et al., 1986). For this reason, almost all N fertilizer used in the Southeast is applied in the form of urea (Zerpa and Fox, 2011). However, as urea dissolves and converts to  $\text{NH}_4^+$  ions by the enzyme urease found in the soil and in plant residue (Pettit et al., 1976), it can be subject to volatilization losses as  $\text{NH}_3$ .

Fertilization with urea may lead to N losses through ammonia ( $\text{NH}_3$ ) volatilization on

loblolly pine plantations (Cabrera et al., 2001; Kissel et al., 2009; Cabrera et al., 2010; Zerpa and Fox, 2011; Elliot and Fox, 2014). Zerpa and Fox (2011) highlighted the importance of knowing the environmental and site conditions before fertilization with urea to minimize volatilization of  $\text{NH}_3$ . Some environmental factors affecting  $\text{NH}_3$  volatilization are temperature (Elliot and Fox, 2014), relative humidity as well as water availability (Cabrera, 2010), and wind speed (Cabrera, 2001). Following fertilization,  $\text{NH}_4^+$  ions are produced quickly increasing the likelihood for  $\text{NH}_3$  volatilization if the soil cation exchange sites do not absorb the  $\text{NH}_4^+$  produced (Zerpa and Fox, 2011). Under poor buffered soils, hydrolysis of urea leads to increased pH and then  $\text{NH}_3$  volatilization will occur (He et al., 1999; Kissel et al., 2009). Further, surface-applied urea on bare mineral soil can lead to larger N losses by  $\text{NH}_3$  volatilization compared with applications of urea to the organic horizon due to the high pH buffering capacity of the humus layer to resist changes in the pH (Kissel et al., 2009, Zerpa and Fox, 2011).

Elliot and Fox (2014) reported N losses to volatilization were 18 and 51% in winter and summer respectively. This was in agreement with Miller et al (1986) and Kissel et al (2009) who indicated that  $\text{NH}_3$  volatilization is reduced during cold seasons. N losses by volatilization can be reduced when high rainfall occurs immediately following fertilization (Flint et al., 2008; Zerpa and Fox, 2011). It has also have been demonstrated that slow released N fertilizers can significantly reduce N losses through  $\text{NH}_3$  volatilization. Zerpa and Fox (2011) found that coated-urea fertilizer (CUF) and urea treated with the urease inhibitor N-(n-butyl) thiophosphoric triamide (NBPT) helped reduced N losses through volatilization in the Atlantic Coastal Plain by 25 and 35% compared with untreated urea.

## *Leaching*

Leaching is a process by which fertilizers with N and P applied into the soil are carried beyond the reach of plants and may be considered as a threat to environmental quality (Silver et al., 2000; Flint et al., 2008; Kadyampakeni et al., 2014). For N, the conversion of  $\text{NH}_4^+$  to the mobile ions  $\text{NO}_2^-$  and  $\text{NO}_3^-$  may increase N losses through leaching (Müller et al., 2003).  $\text{NO}_3^-$  moves freely in the soil and can be easily lost from the rooting zone (Flint et al., 2008).

Following fertilization N leaching occurs primarily in sandy-textured soils (Lee and Shibu, 2005; Davis et al., 2012), but also leaching may occur in fine clays affecting N pools (Silver, et al., 2000). Lee and Shibu (2005) assessed N losses through  $\text{NO}_3^-$  leaching after seven years of fertilization in cottonwood and loblolly pine growing on well-drained Redbay sandy loamy soils. They found that N fertilizer applications above  $56 \text{ kg ha}^{-1} \text{ year}^{-1}$  increased leaching and it continued to increase as fertilization rate increased. Davis et al (2012) conducted an experiment at 10 experimental sites in New Zealand with a range of soil types from well-drained sands to imperfectly drained silt loams and clay loams. After 2 years, results showed that leaching increased at 8 of the 10 sites studied. To decrease leaching losses, some researches recommend multiple small doses of fertilizers (Paramasivam et al., 2000, Davis et al. 2012). However, by applying repetitive doses of fertilization with N, also increases nitrification leading to nitrate leaching (Johnson and Todd, 1988; Johnson, 1992) so that further investigation may be necessary to address this problem.

Soil properties, mainly texture, may play an important role in the retention or leaching of P from the soil. Leaching of P is generally thought to be low because P binds tightly in the soil with clay, Fe and Al. Following fertilization P may leach through the soil profile in sandy soils (Will et al., 2006). Low concentrations of Al may lead to low retention of P in the mineral soil in

sandy-textured soils and consequently downward movement of P may occur (Humphreys and Pritchett, 1971). However, it has been found that large amounts of P could be retained in coarse-textured soils (Arai and Livi, 2013). This was attributed to the phosphate binding capacities of amorphous Al/Si/Fe coating the sand particles suggesting that P concentrations in coarse-textured soils may be as high as the fine clays. Kadyampakeni et al (2014) in an experiment with Spodosols and Entisols in the Atlantic Coastal Plain reported large P and K retention within the rooting zone in the first 0-30 cm depth. The P retained lessened the potential for leaching and consequently reduced risk of surface and groundwater contamination. This is comparable with the study by Pritchett and Comerford (1982) and Vaananen et al (2008), in which P leached from the E horizon and was retained in the B horizon, which reduced the risks for leaching.

#### *P fixation*

P fixation refers to the conversion of available forms of P into unavailable or insoluble forms attached to the soil (Dean, 1949). This is because relative high portion of the P in the soil is fixed to clays, Fe, and Al or is in unavailable organic forms (Turner and Lambert, 1985; Condron et al., 2005; Achat et al., 2009). Because of this fixation, P often limits tree growth even though the amounts of P in forest soils appear to be sufficient to meet vigorous tree growth. This is because the pools of P in solution are small and therefore limits growth (Fox et al., 2011).

The amount of available P in the soil following fertilization is influenced by soil physical and chemical properties (Shigaki and Sharpley, 2011). In this study, they found that P fixed in soils from nine different sites ranged from 8 to 270 mg kg<sup>-1</sup>. The samples were collected from Florida, Pennsylvania, and New York and encompassed a variety of soil types. P fixation was a function of pH, clay content, organic C, Fe, and Al in the soil. Shigaki and Sharpley, (2011) also considered P availability can be influenced by other factors such type of P fertilizer applied,

time, and application rate. Little Al and consequently little P sorption was found in sandy sites compared with loamy to clayey soils.

Kadyampakeni et al (2014) highlighted the importance of adjusting P fertilization to reduce potential P losses to fixation or potential leaching in poorly and excessively drained soils in Florida. They concluded that that application with P at rates of 50 to 70 kg ha<sup>-1</sup> is suitable for optimal growth in young stands in these soils. In the study by Kadyampakeni et al (2014) higher concentrations of P showed the need to reduce the doses of P application rates in order to avoid the excess of P fixation and leaching.

Further, fertilization may lead to reductions in the total inorganic P and to an increase in the organic P pools in the long-term (Dodd et al., 2013). This is likely because inorganic P applied through fertilization is quickly converted to organic forms of P (Fox et al., 2011). Large and quick sorption capacities of P make forest soils sinks for P (Beauchemin et al., 1996; Kelly and Kelly, 2001) indicating that once in the soil, P is sorbed to Al and Fe oxides and consequently become occluded reducing soil P availability (Parfitt et al., 1975). Early publications by Dean (1949) indicated that fixation decreased as soil pH approached neutral. Therefore, liming is an alternative solution to increase P availability in acids soils (Delgado and Torrent, 2000). Neutrality also plays an important role in the predominance of Fe and Al compounds. Yaghi and Hartikainen, 2013 examined P sorption on light-expanded clay aggregates (LECA) coated with Al or Fe at pH under and over 6. At pH less than 6 the Fe-LECA was more efficient sorbent while at pH equal or greater than 6 the Al-LECA were the dominant sorbents. It appears that soil properties such as texture and Fe and Al contents will influence in the leaching or retention of P in the soil.

## Chapter 3: Materials and Methods

### Site Descriptions

A regional fertilization study was established between 1998 and 2001 by the Forest Productivity Cooperative (FPC) to examine the growth response of juvenile loblolly pine (*Pinus taeda* L.) plantations to applications of nitrogen (N) and phosphorous (P) across the southeastern United States. Nine sites were selected for this research in order to include a range of soil types that occur in the South (Tables 1 and 2). Stands ranged from 2 to 6 years old with stocking from 1247 to 2245 trees per hectare. All plots were thinned to 395 trees ha<sup>-1</sup> 12 years since treatment started. This was conducted as a row thinning that removed every 5<sup>th</sup> row followed by low thinning between the cut rows. Fertilization with N and P started between 1998 and 2001 and applied different rates of N and P at various application frequencies. Four treatments including one control and three fertilized plots with fertilizer applied every four years were selected for this study (Tables 3 and 4). These fertilizer treatments ceased after 12 years. The repeated fertilizer applications of 0, 135, 202, and 269 kg N ha<sup>-1</sup> created a range in the cumulative amount of N added ranging from 540, 808, and 1076 kg N ha<sup>-1</sup> over 12 years. P was applied at a rate of 10% of the N in each treatment. N and P were added to plots as urea (46-0-0) and di-ammonium phosphate (18-46-0) in the spring as a broadcast surface application. The experimental design was a randomized complete block design (RCBD) with 2 or 4 replications of the treatments depending on the site. Treatment plots measured 35 x 35 m with a 20 x 20 m internal measurement plot.

The nine sites selected for this study were grouped into three classes based on the CRIFF groups present. The CRIFF system groups soils together into classes based on soil drainage class, soil texture, and depth at the B-horizon (Fisher and Garbett, 1980; Jokela and Long, 2000;

Jokela, 2004). The CRIFF soil groups have been used to help guide forest management decisions in the South including fertilization and species selection (Fox, 2004b; Fox et al., 2007b). We grouped the sites into three classes: Soil group 1 combined CRIFF A and B soils that includes the poorly drained Ultisols; soil group 2 combined the CRIFF C, D, and G that includes the poorly to somewhat well-drained Spodosols and Entisols; and soil group 3 combined the CRIFF E and F which are well-drained Ultisols. This approach allowed us to compare the effect of soil drainage class and texture on N and P retention following fertilization.

### **Sampling**

Sampling of the O horizon and mineral soil was done between May and June 2013 after 13 to 16 years since treatments started during which time four applications of fertilizers at the designated rates were made at 4 year intervals. Following site preparation, tillage, treatment plots were hand-planted with 0-1 genetically improved *Pinus Taeda* L. seedlings (Carlson et al., 2006). The O horizon including twigs and small branches less than 2.5 cm diameter was sampled at four random locations in each plot with a 0.25 m<sup>2</sup> frame. The four subsamples were composited for each plot. The three sub-horizons (O<sub>i</sub>, O<sub>e</sub>, and O<sub>a</sub>) of the O horizon were composited and analyzed together. The sites included in the poorly-drained Ultisols (CRIFF A and B) and poorly to somewhat well-drained Sandy Spodosols (C, D, and G) were bedded except for the well-drained Ultisols. The sampling collection on the bedded plots was sampled on and between the beds. The surface mineral soil was sampled at five random locations within each plot with a soil auger (5.94 cm diameter) (AMS, American Fall, ID) to a depth of 30 cm. Samples were divided into two depth increments of 0-15 cm and 15-30 cm and five subsamples in each plot from each increment where composited. At one random location within each plot, where the surface soil was collected samples were collected to a depth of 100 cm and in addition

to the 0-15 and 15-30 cm increments were divided into three depth increments: 30-50 cm, 50-75 cm, and 75-100 cm.

O horizon samples were dried at 60 °C for one week, weighed, and ground with a Thomas-Wiley Model 4 mill (Thomas Scientific, USA). All values reported are for oven-dry mass. Mineral soil samples were air-dried at room temperature for approximately one month, ground using a mortar and pestle, and passed through a 2 mm sieve prior to the chemical analysis. Coarse fragments larger than 2mm were discarded.

Two bulk density cores (4.5 x 4.5 cm) (AMS, American Fall, ID) for each plot were taken at depths of 0-15 cm and 15-30 cm with a bulk density hammer. Bulk density samples were oven-dried at 105 °C for 24 hours, weighed, and passed through a 2 mm sieve. Coarse fragments greater than 2 mm were subtracted to obtain the dry mass per volume ( $\text{g cm}^{-3}$ ) of the fine fraction. Bulk density for depths 30-50 cm, 50-75 cm, and 75-100 cm were obtained from the Natural Resources Conservation Service database (NRCS) based on the soil series mapped at each location (<http://www.nrcs.usda.gov/wps/portal/nrcs/soilsurvey/soils/survey/state/>).

### **Physical and Chemical Analysis**

Particle size analysis (% sand, silt, and clay) was determined by the hydrometer method and then used to determine soil textural class (Ashworth et al., 2001). Total-N in the O horizon and mineral soil was determined by dry combustion on a VARIO CNS Analyzer (Elementar, Hanau, Germany) (Wright and Bailey, 2001). Mineral soil 2M KCl extractable  $\text{NH}_4^+$  (ammonium) and  $\text{NO}_3^-$  (nitrate) was determined on a TRACCS 2000 Auto analyzer (SEAL Analytical, Mequon, WI) following extraction with 2M KCl by shaking five grams of soil with 50 mL 2M KCl solution and filtering through a Whatman No.42 glass fiber filter (Kuo, 1996; De Boer, 1996).

Total P in the O horizon was determined by dry-ashing 0.5 grams at 500 °C and dissolving the ash in 10 mL of 6 N HCl solution and 40 mL of deionized water. Phosphorus in solution was analyzed as described by Hansen et al (2013) on a Varian Vista MPX Inductively Coupled Plasma atomic emission Spectrophotometer (ICP-AES) (Varian, Palo Alto, CA, USA). Mineral soil extractable P, Al and Fe were determined by ICP-AES after extraction using the Mehlich 3 Method (Mehlich, 1984; Zhang et al., 2009; Jalali and Ostovarzadeh, 2009). Mineral soil pH (1:1, soil/water by volume) was determined using a combination glass electrode. Extractable K, Ca, Mg, Al, and Fe were determined by ICP-AES following Mehlich I extraction and cation exchange capacity (CEC) was estimated as the sum of extractable cations. pH and CEC were determined by the Virginia Tech Soil Testing Lab.

### **Statistical Analysis**

Treatment differences in the N and P in the O horizon were tested using the mixed model ANOVA of SAS version 9.4 (PROC MIXED, SAS Institute Inc., Cary, NC, USA) and pairwise comparisons were performed with the Tukey test ( $\alpha=0.10$ ). Treatment and soil group were fixed effects and replication within study was a random effect (Littell et al., 2006).

Proc Mixed with repeated measures was used to analyze the N and P data in the mineral soil. Treatment and soil group were fixed effects and replication was a random effect. Logarithmic transformation was performed when needed on mineral soil data to meet normal distribution assumptions as indicated in Tables 6, 7, and 8. Depth was considered as a repeated measure in the model (Littell, 1998). The ‘slice’ option was used with the last square means to test interaction effects between treatments and soil group with depth in the soil profile. The unstructured (UN) covariance matrix was selected as the most appropriate based on the Akaike (AIC) and Bayesian (BIC) information criteria.

## Chapter 4: Results

### O Horizon

There was a significant soil group by fertilization interaction effect for O horizon mass ( $p=0.0521$ ) (Table 6). Mass of the O horizon was greater in the fertilized treatments than the control in soil groups 1 (poorly-drained Ultisols) and soil group 2 (poorly to somewhat well-drained Sandy Spodosols), but not in the well-drained Ultisols in soil group 3. In the nonfertilized plots, mass of the O horizon was similar in all the three soil groups ranging from 21.3 to 25.6 Mg ha<sup>-1</sup>. Total O horizon mass in the fertilized treatment increased from 36.7 to 44.6 Mg ha<sup>-1</sup> in soil groups 1 and 2 respectively (Table A1). There was no impact of fertilization rate on O horizon mass (Fig. 2).

There was a significant main effect of fertilization on N concentration in the O horizon ( $p<0.0001$ ). There was no difference among soil groups (Table 6). Total N concentration in the O horizon was increased by fertilization only in the highest fertilization rate in soil group 1 from 4.45 in the control to 5.44 g kg<sup>-1</sup> at the highest rate (Fig. 3). In the nonfertilized plots, total N concentration in the O horizon was quite similar in soil groups 2 and 3 ranging from 4.52 to 4.78 g kg<sup>-1</sup> compared with the fertilized plots that ranged from 5.21 to 5.43 g kg<sup>-1</sup> (Table A1, Fig. 3).

Fertilization also increased N content of the O horizon ( $p<0.0001$ ). There was also a soil group by fertilization interaction effect ( $p=0.1000$ ) (Table 6). O horizon total N content was greater in the fertilized treatments than the controls in the poorly drained Ultisols and poorly to somewhat well-drained Spodosols and Entisols, but only in the highest rate in the well-drained Ultisols. In the nonfertilized plots total N content in the O horizon was 94, 114, and 118 kg ha<sup>-1</sup> for soil groups 1, 2, and 3. At the highest treatment, total N content in the fertilized plots increased to 206, 231 and 175 kg ha<sup>-1</sup> respectively (Table A1, Fig. 3).

Fertilization affected O horizon total P concentration ( $p=0.0020$ ) (Table 6). Total P concentration in the O horizon increased at the highest fertilization rate in soil group 3 from 208 to 278  $\text{mg kg}^{-1}$ . In the nonfertilized plots total P was fairly similar in soil groups 1 and 2 ranging from 225 to 289  $\text{mg kg}^{-1}$  compared with the fertilized plots that ranged from 268 to 295  $\text{mg kg}^{-1}$  at the highest fertilization rate (Table A1, Fig. 4).

There was a significant soil group by fertilization interaction for P content of the O horizon ( $p=0.0342$ ) (Table 6). Total P content was greater in the fertilized treatments than the controls in the poorly drained Ultisols in soil group 1 and poorly to somewhat well-drained Spodosols and Entisols in soil group 2, but only in the highest rate in the well-drained Ultisols in soil group 3 (Fig. 4). In the nonfertilized plots total P content in the O horizon was 4.8, 7.1, and 4.9  $\text{kg ha}^{-1}$  for soil groups 1, 2, and 3 respectively. At the highest rate, total O horizon in the fertilized plots increased to 10.4, 13.2 and 8.9  $\text{kg ha}^{-1}$  (Table A1, Fig. 4).

### **Mineral Soil**

Mineral soil total N concentration was not affected by fertilization ( $p=0.4805$ ) (Table 7). However, there was significant soil group x depth interaction ( $p<0.0001$ ). N concentration was greater in the first 0-15 cm soil surface depth (Fig. 5). In the nonfertilized plots N concentration (0.61, 0.30, and 0.43  $\text{g kg}^{-1}$ ) was similar to the fertilized plots at the highest fertilization rate (0.60, 0.38, and 0.43  $\text{g kg}^{-1}$ ) in all the three soil groups. Total N in the soil profile decreased with depth (Table A2, Fig. 5), but slightly increased in the 50-75 cm subsoil depth due to a Bh horizon in the poorly to somewhat well-drained Spodosols and Entisols in soil group 2. Total N concentration decreased in the sequence of soil group 1 > 3 > 2 (Table A2, Fig. 5).

Fertilization had no effect on mineral total N content ( $p=0.5151$ ) (Table 7). However, there was an interaction between soil group and soil depth ( $p<0.0001$ ). Even though there was no

significant difference in the treatments following fertilization, there was a small increase in total N content in all the soil groups (A.2, Fig. 5).

Mineral soil extractable  $\text{NH}_4^+$  concentration in the control was higher than the fertilized plots (Fig. 6). An overall fertilization effect was observed ( $p=0.0111$ ) (Table 7, Fig. 10). There was an interaction soil group by depth for extractable  $\text{NH}_4^+$  ( $p<0.0001$ ) (Table 7). Most  $\text{NH}_4^+$  was found in the first 0-15 cm soil and lower levels of  $\text{NH}_4^+$  were found deeper in the profile in all the three soil groups (Table A3, Fig. 6). In the nonfertilized plots, extractable  $\text{NH}_4^+$  in the soil surface (0-15 cm) in the poorly and well-drained Ultisols in soil groups 1 and 3 ranged from 12.54 and 10.27  $\text{mg kg}^{-1}$  respectively while lower extractable  $\text{NH}_4^+$  (8.95 and 5.99  $\text{mg kg}^{-1}$ ) were found in the fertilized plots at the highest fertilization rate. In the poorly to somewhat well-drained Spodosols and Entisols in soil group 2, fertilization increased extractable  $\text{NH}_4^+$  in the soil surface from 3.88 to 5.73  $\text{mg kg}^{-1}$  at the highest fertilization rate, but from 15-100 cm the nonfertilized were higher than the fertilized plots. Extractable  $\text{NH}_4^+$  concentration decreased in the sequence of soil group 1 > 3 > 2 (Table A3, Fig. 6).

There was an overall treatment effect on mineral soil extractable  $\text{NH}_4^+$  content ( $p=0.0132$ ) (Table 7, Fig. 10). There was also a significant soil group by depth interaction ( $p<0.0001$ ) (Table 7). Extractable  $\text{NH}_4^+$  content in the profile from 0 to 100 cm was greater in the nonfertilized than the fertilized plots. In soil groups 1, 2, and 3 the mineral soil  $\text{NH}_4^+$  content in the control plots (49.1, 26.7, and 59.7  $\text{kg ha}^{-1}$ ) was greater than the fertilized plots (42.7, 25.6, and 53.9  $\text{kg ha}^{-1}$ ) as mean differences of fertilized and nonfertilized plots. Extractable  $\text{NH}_4^+$  content decreased in the sequence of soil group 3 > 1 > 2 (Table A3, Fig. 6).

Fertilization had an overall effect on mineral extractable  $\text{NO}_3^-$  concentration ( $p=0.0656$ ) (Table 7, Fig. 10). There was also significant soil group by depth interaction effect ( $p<0.0001$ )

(Table 7). The highest  $\text{NO}_3^-$  concentrations were found in the first 0-15 cm soil surface and decreased with depth in all the three soil groups (Table A4, Fig. 7). In the soil surface of the poorly-drained Ultisols in soil group 1,  $\text{NO}_3^-$  in nonfertilized and fertilized plots was similar ranging from 11.22 and 12.59  $\text{mg kg}^{-1}$  at the highest fertilization rate. In the well-drained Ultisols in soil group 3,  $\text{NO}_3^-$  in fertilized plots (8.5  $\text{mg kg}^{-1}$ ) was higher than the controls (3.78  $\text{mg kg}^{-1}$ ).  $\text{NO}_3^-$  in the fertilized plots (0.96  $\text{mg kg}^{-1}$ ) was higher than the controls (0.46  $\text{mg kg}^{-1}$ ) in soil group 2, but  $\text{NO}_3^-$  was very low compared with soil groups 1 and 3. Extractable  $\text{NO}_3^-$  concentration decreased in the sequence of soil group 1 > 3 > 2 (Table A4, Fig. 7).

Fertilization affected on mineral soil extractable  $\text{NO}_3^-$  content ( $p=0.0508$ ) (Table 7, Fig. 10). There was also a significant soil group by depth interaction ( $p<0.0001$ ) (Table 7). In the poorly-drained Ultisols in soil groups 1 the mineral soil  $\text{NO}_3^-$  content in the control plots (31.2  $\text{kg ha}^{-1}$ ) was similar to the fertilized plots (31.5  $\text{kg ha}^{-1}$ ) as mean differences between fertilized and nonfertilized plots. In the well-drained Ultisols in soil group 3,  $\text{NO}_3^-$  content in the fertilized plots (22.7  $\text{kg ha}^{-1}$ ) was higher than the controls (10.0  $\text{kg ha}^{-1}$ ). Finally, in the poorly to somewhat well-drained Spodosols and Entisols in soil group 2  $\text{NO}_3^-$  in fertilized plots (7.1  $\text{kg ha}^{-1}$ ) was greater than the controls (4.2  $\text{kg ha}^{-1}$ ). Extractable  $\text{NO}_3^-$  content decreased in the sequence of soil group 1 > 3 > 2) (Table A4, Fig. 7).

Fertilization had no effect on the total extractable inorganic N concentration ( $\text{NH}_4^+ + \text{NO}_3^-$ ) ( $p=0.1746$ ) and content ( $p=0.2058$ ) in any of the three soil groups (Table 8). There was an interaction between soil group and soil depth ( $p<0.0001$ ). The majority of the inorganic N was concentrated in the first 0-15 cm and decreased with depth (Fig. 8). In the surface 0-15 cm of the three soil groups, concentration and content of the total inorganic N were similar between the fertilized and control plots (Table A5, Fig. 8).

Fertilization increased mineral soil extractable P concentration ( $p=0.0013$ ) (Table 8, Fig. 10). There was an interaction between soil group and soil depth ( $p<0.0001$ ) (Table 8). In the poorly and well-drained Ultisols in soil groups 1 and 3, most P was found in the 0-15 cm soil surface depth and decreased with depth while in the poorly to somewhat well-drained Spodosols and Entisols in soil group 2; P was similar throughout the soil profile (Table A6, Fig. 9). Extractable P in the fertilized plots (20.43, 16.13, and 3.37 mg kg<sup>-1</sup>) was greater at the highest fertilization rate than the controls (9.52, 7.77, and 1.60 mg kg<sup>-1</sup>) in all the three soil groups. Results for the well-drained Ultisols in soil group, 3 showed quite low extractable P concentration throughout the soil profile. P concentration decreased in the sequence of soil group 2 > 1 > 3 (Table A.6, Fig. 9).

An overall increase was observed in P content following fertilization ( $p=0.0008$ ) (Table 8, Fig. 10). There was a significant soil group by fertilization interaction ( $p=0.0796$ ) and soil group by depth interaction effect ( $p<0.0001$ ). P content in the profile from 0 to 100 cm in the fertilized plots was higher than the controls in all the three soil groups. The average in the fertilized plots were 138, 68, and 7.5 kg ha<sup>-1</sup> while the results from the control plots were 78, 42, and 3.5 kg ha<sup>-1</sup> (Table A.6, Fig. 9). The well-drained Ultisols reported fairly low P pools compared to the poorly-drained Ultisols and poorly to well-drained Spodosols and Entisols.

## Chapter 5: Discussion

The effect of repeated N and P fertilizer applications were quantified in the O horizon and mineral soil at 9 experimental sites at a regional fertilization study in the southeastern United States that were subsequently grouped into three soil groups based on the CRIFC Classification System in order to test the hypothesis (i) Different fertilization treatments result in different long-term accumulations of soil nutrients, (ii) The texture of soils (clay vs. sandy textured) has a significant impact on the accumulation of nutrients in the soil in the long-term, and (iii) Drainage class will have a significant impact on accumulation of soil nutrients.

A potential loss mechanism that likely affected to all the soil groups was ammonia ( $\text{NH}_3$ ) volatilization. Elliot and Fox (2014) reported N loss by  $\text{NH}_3$  volatilization following urea applications by 18 and 51% in summer and winter respectively.  $\text{NH}_3$  volatilization can be associated to other factor such as wind speed (Cabrera et al., 2001) along with high relative humidity that plays a critical role in the  $\text{NH}_3$  volatilization (Cabrera, 2010), and high soil pH (He et al., 1999). Likewise, N loss through denitrification and volatilization may be associated with low N accumulations found in all soil groups in this study.

Fertilization increased mass in the O horizon in two of the three soil groups: 1 and 2 (poorly-drained Ultisols and poorly to somewhat well-drained Spodosols and Entisols) by 15.4 and 19.0  $\text{Mg ha}^{-1}$  at the highest fertilization rate (1076  $\text{kg N ha}^{-1}$ ) (Fig. 2). Other researches have found that fertilization has a strong effect on the production of mass in the O horizon by increasing leaf area (Albaugh et al., 1998; Gough et al., 2004), which likely led to higher litterfall and consequently greater biomass accumulation in our two soil systems. Balster and Marshall (2000) and Vose and Allen (1998) also found out increases in leaf area as a result of fertilization applications. Our results (mean differences between the fertilized and control plots)

from the poorly to somewhat well-drained sandy Spodosols and Entisols in soil group 2 (19.0 Mg ha<sup>-1</sup>) (Table A1) were comparable with what Kiser and Fox (2012) reported on a sandy site in North Carolina where repeated fertilization increased mass in the in the O horizon by 16.7 Mg ha<sup>-1</sup>. Will et al (2006) reported a 90% of increase in O horizon mass after applying 900 kg N ha<sup>-1</sup>. Soil properties from the study by Will et al (2006) were similar to our soil groups 1 and 2, which increased by 72 and 74 % after applying 1076 kg N ha<sup>-1</sup>. In contrast, the O horizon mass did not increase in the well-drained Ultisols in soil group 3 even at the highest fertilization rate (Table A1, Fig. 2). We hypothesize that the decomposition rate was greater in soil group 3 that were well-drained Ultisols compared to soil groups 1 and 2, which were poorly-drained Ultisols to poorly to somewhat well-drained Spodosols and Entisols. The excess of soil moisture due to high water table in the poorly-drained soils creates anaerobic conditions likely decreased microbial activity leading to a low decomposition rates and consequently greater O horizon mass accumulations. This is consistent with findings by Yoon et al (2014) who conducted their research on litter decomposition. They concluded that litter decomposition rate was higher under low soil moisture conditions. However, in higher soil moisture conditions the litter decomposition was lower leading to greater biomass accumulation.

N and P concentrations in the O horizon were similar between the fertilized and control plots in all the soil groups (Fig. 3 and 4). Fertilization had little effect on the O horizon concentration likely because nutrients were removed back to the plant by retranslocation prior to litterfall (Albaugh et al., 2008). Prescott (1995) found that more than 50% of the N and P requirements for tree growth were met from retranslocation. Further, retranslocation efficiency depends on the soil fertility (Nambiar and Fife, 1991) so that at high foliar concentrations after fertilization, there will be more N and P retranslocated. Binkley and Reid (1985) reported an

average of 20% of the N applied was retranslocated in fertilized plots at Wind River fertilization experiment. Will et al (2006) reported increases in N concentration for the O horizon, but not in P in southern Georgia with similar soil properties as our sites in soil group 1 (poorly-drained Ultisols). Kiser and Fox (2012) reported increases on the O horizon N and P concentrations after fertilization on sandy soils in North Carolina. In the study reported by Kiser and Fox (2012) more N and P were applied to the soil compared to ours (30% and 56% respectively). These higher fertilization rates likely led to greater N and P concentrations than we found in our regional fertilization study.

Fertilization increased O horizon N and P content. N and P content were the product of O horizon mass and nutrient concentration. The increase of mass in the O horizon led to greater O horizon N and P content. Our results (Table A.1, Fig. 3 and 4) are in agreement with Kiser and Fox (2012) and Will et al (2006), who also found that N and P content of the O horizon were largely driven in biomass accumulation. The implications are that we will have large N and P contents in the O horizon regardless of its low concentrations because of the increase of the mass in the O horizon caused by fertilization.

In this study, fertilization did not affect mineral soil total N concentration and content. This is consistent with early studies by Miller (1981), who claimed that fertilization benefits the trees and not the site because it has little impact mineral soil N concentration on sandy soils. He reported no differences in soil N between fertilized and unfertilized plots. Kiser and Fox (2012), on the other hand, found increases in the mineral soil N. However, they concluded that following harvesting the O horizon left that provides N to the mineral soil is going to be lost in a short-term by fast decomposition so that long-term accumulations of N in the soil to improve site quality are unlikely.

Given that we did not observe significant increases in the mineral soil N in any soil group (Table 7) we failed to reject the null hypothesis and there appeared to be no differences in mineral soil N retention due to soil texture and drainage class. This suggests N fertilization will have little impact on N availability throughout subsequent rotations and thus has little impact on long-term site quality and potential productivity. Because there was a significant increase in N content in the O horizon, if the O horizon remaining after harvesting can be managed in some way so that the Assart effect is extended, it may be possible to delay the onsite of N deficiency. This could lead to extend the availability of nutrients in the soil for longer periods of time decreasing the need for fertilization and thus leading to cost reductions and increase in profits. Perhaps incorporating organic matter into the soil during site preparation may lead to slow litter decomposition rates as pointed out by Papa et al (2014). However, further research needs to be done to address this topic. For now, low N found in soils suggests that fertilization with N in the southeastern United States will be necessary in each rotation as suggested by Kiser and Fox (2012) despite the fact that we added over 1000 kg N ha<sup>-1</sup>.

We also examined levels of extractable NH<sub>4</sub><sup>+</sup> and NO<sub>3</sub><sup>-</sup> in the soil systems to find out if they were more sensitive than total N. Extractable NH<sub>4</sub><sup>+</sup> content were higher in the nonfertilized than the fertilized plots in all the three soil groups (Fig. 6). A reasonable hypothesis is that fertilization stimulated nitrification so that NH<sub>4</sub><sup>+</sup> was quickly converted to NO<sub>3</sub><sup>-</sup> following fertilization and then the NO<sub>3</sub><sup>-</sup> produced was rapidly lost through denitrification in the poorly-drained Ultisols or leaching in the sandy Spodosols and Entisols (Fig. 6 and 7). Martikainen (1984) found out that urea fertilization increased nitrification rates in soils with pH similar to ours (Table 5) supporting this hypothesis. This suggests that N fertilization with urea will not increase extractable NH<sub>4</sub><sup>+</sup> pools regardless the soil type given that fertilization will stimulate

nitrification.

Groffman and Tiedje (1989) reported annual N losses by denitrification of over 40 kg N ha<sup>-1</sup> year<sup>-1</sup> in the spring and summer in poorly-drained clay loam soils similar to soil group 1. Mohn et al (2000) detected maximum N loss by denitrification as soil temperature and pH increased and redox potential decreased. Denitrification rates in spring and summer reached up to 6.49 mg N m<sup>-2</sup> (23.7 kg N ha<sup>-1</sup> year<sup>-1</sup>). However, when they added high N inputs followed by rainfall events, N was lost up to 13.58 mg N m<sup>-2</sup> (49.6 kg N ha<sup>-1</sup> year<sup>-1</sup>). Weier et al (1993) observed that N loss by denitrification increased as soil texture became finer and soil moisture content was increased. All the results described above support our assertion that low NO<sub>3</sub><sup>-</sup> levels observed our in poorly-drained Ultisols in soil group 1 were due to denitrification.

In the poorly to somewhat well-drained sandy Spodosols and Entisols (soil group 2), concentrations of NO<sub>3</sub><sup>-</sup> were low throughout the whole soil profile (Fig. 7). We hypothesize that any NO<sub>3</sub><sup>-</sup> produced following fertilization in these sandy soils were partly taken up by the trees in the rooting zone and considerable amounts of N was lost through leaching. Lee and Shibu (2005) assessed N losses through leaching from 7-year-old cottonwood and loblolly pine stands treated with repetitive N fertilization in Santa Rosa County, Florida on soils with similar characteristics to this soil group. The N application was 276 kg N ha<sup>-1</sup> after a period of 2-year study from June 2001 through May 2002. NO<sub>3</sub>-N concentrations for both species at 120 cm depth were 37.7 and 50.7 mg L<sup>-1</sup> respectively. Fluxes of N that leached beyond the rooting zone demonstrate that sandy soils do not retain NO<sub>3</sub><sup>-</sup> as we observed in this sandy Spodosols and Entisols in soil group 2. The high NO<sub>3</sub><sup>-</sup> concentrations found deep in the horizon may produce small impacts on water contamination of water as pointed out by Flint et al (2008) and Wexler et al (2014).

Fertilization increased extractable  $\text{NO}_3^-$  in the well-drained Ultisols in soil group 3. The highest extractable  $\text{NO}_3^-$  concentration was found in the soil surface with low levels in the subsoil (Fig. 7). This may be attributed to the high clay contents of these soils that prevented  $\text{NO}_3^-$  leaching out of the system (Table 5). The fine-textured soils likely played an important role in the retention of  $\text{NO}_3^-$  due to its higher anion adsorption capacity (Amato and Ladd, 1992; Will et al., 2006). In these well-drained Ultisols N losses by denitrification were probably very low. Losses of less than  $1 \text{ kg N ha}^{-1} \text{ year}^{-1}$  have been reported by denitrification in well-drained soils (Groffman and Tiedje, 1989). Mohn et al (2000) also reported low denitrifying activities throughout the year in well-aerated raw humus so that negligible denitrification rates can be considered in well-aerated soils.

When combined extractable  $\text{NH}_4^+$  and  $\text{NO}_3^-$  to find out if fertilization had a treatment effect in the total inorganic N, the combination of  $\text{NH}_4^+$  and  $\text{NO}_3^-$  was canceled out with no significant overall effect (Table 8). In all the soil groups, the total inorganic N in fertilized plots were fairly similar to the controls (Fig. 8) indicating that when  $\text{NH}_4^+$  was increased,  $\text{NO}_3^-$  was decreased particularly in soil group 2 and 3. This suggests that fertilization will not increase either total mineral soil N or the extractable inorganic N ( $\text{NH}_4^+$  and  $\text{NO}_3^-$ ) in any of the three soil groups.

Fertilization increased mineral soil P in all soil groups. Mean differences of fertilized and nonfertilized plots in soil groups 1, 2, and 3 indicated that Mehlich 3 extractable P increased after fertilization by 62, 76 and 114% respectively (Table A6). The elevated P levels in all soil types after fertilization (Fig. 9) will likely affect long-term P availability and suggests a long-term increase in site quality (Pritchett and Comerford, 1982); Fox et al, 2011; Kissler and Fox, 2012). Similarly, Harding and Jokela (1994) reported sustained growth in slash pine for 25 years

following P fertilization. However, Everett and Palm-Leis (2009) conducted a study with similar soil properties as soil group 1 (poorly-drained Ultisols) and recommended for loblolly pine stands to fertilized with 45 kg P ha<sup>-1</sup> in the first rotation may need additional P fertilization by age 3 in the second rotation to keep desirable foliar N concentrations in trees. Our study at the lower treatment (412 = 54 kg P ha<sup>-1</sup>) reported P content of 70.6 kg ha<sup>-1</sup> (Table A6) recovered after fertilization, which was the 68% increase. Although in our study we added around 20% more than the in study by Everett and Palm-Leis (2009) at the lowest fertilization rate, the results were in agreement. Our data suggests that additional P fertilization in subsequent rotations may not be needed on these soils.

P accumulation in the mineral soil was probably related to the Al, Fe and clay in the profile (Table 5). Yuan et al (1960) reported over 80% of P retention by Al and Fe phosphates in soils with similar characteristics to soil group 1 (poorly-drained Ultisols) and 2 (poorly to somewhat well-drained Spodosols and Entisols). In soil group 1 higher extractable P concentrations occurred in the soil surface and decreased gradually in the subsoil (Fig. 9). The P distribution in the soil profile of soil group 1 was in agreement with the description of the soil properties for CRIFF soils A and B made by Fox et al (2004b). This was likely due to the relative increase of clays as depth increased, suggesting P retention as it reached the clays (Table 5).

The poorly to somewhat well-drained Spodosols and Entisols in soil group 2 showed the largest accumulation of P. This was likely because the Fe and Al coated the P to the sand fractions (Arai and Livi, 2013). Yuan and Lavkulich (1994) found good correlations between oxalate-extractable Al and Fe and P sorption in Spodic horizons. The phosphate binding capabilities of the Fe and Al in soil group 2 suggests that P concentrations in coarse-textured soils may be as high as the fine clays. The high contents of P found in this sandy Spodosols and

Entisols in soil group 2 are in agreement with what Kiser and Fox (2012) reported in sandy sites in North Carolina.

Extractable P concentrations in the well-drained Ultisols in soil group 3 were much lower than soil groups 1 and 2 despite the fact that we added the same amounts of P (Fig. 9). This may be related to the high clay content in this soil group (Table 5), which strongly sorbed the added P. Early studies have shown that clay minerals such as kaolinite, halloysite, and montmorillonite can fix large quantities of P, decreasing extractable P (Dean, 1949). Kadyampakeni et al (2014) suggested decreasing or adjusting fertilization rates of P to minimize its movement beyond the rooting zone in sandy soils and reduce excess of retention of P on clay, Fe, Al. Humphreys and Pritchett (1971) found that soils with high P sorption capacity the majority of the P applied was retained in the soil surface in very poor available forms, which is in agreements with our finding in this soil group 3 that were well-drained Ultisols.

These results suggest that P fertilization will affect the long-term soil P availability and consequently improve site quality. The P retained in the poorly-drained Ultisols and poorly to somewhat well-drained Spodosols and Entisols (Fig. 9) appears to be enough to sustain plant growth for much of the next rotation without any further P additions. However, the lower extractable P in the well-drained Ultisols suggests that P fertilization may again be needed in the next rotation.

## Chapter 6: Conclusions

Fertilization with N did not have significant impacts on mineral soil N content in any of the three soil groups. N fertilization did increase N contents in the O horizon by 112, 113, and 61 kg N ha<sup>-1</sup> for soil groups 1, 2, and 3 respectively. However, previous work has shown that the N accumulated in the O horizon will be rapidly mineralized as part of the Assart flush following harvesting. This is a point in the rotations when soil N supply already exceeds plant demand, so that added N releases from the O horizon in fertilized stands may not improve long-term N availability in fertilized stands. Therefore, we failed to reject the null hypothesis that N fertilization will have no effect on mineral soil N regardless soil texture, soil drainage class, and fertilization rate. However, we reject the null hypothesis for O horizon N and conclude that N fertilization had an effect on N in the O horizon. Our results suggest that N fertilization will not have significant effect on mineral soil N supply and therefore long-term increases in site quality on most forest soils in the southeastern United States following fertilization is unlikely. Likewise, fertilization did not increased extractable inorganic N (NH<sub>4</sub><sup>+</sup> + NO<sub>3</sub><sup>-</sup>) in any of the soil groups. The inorganic N found in different levels among the soil groups suggests that soil type will influence in the magnitude of loss through denitrification or leaching and retention of N. Denitrification will occur in poorly-drained Ultisols mostly during spring and summer. Sandy-textured soils in soil group 2 will be prone to leaching because their low capacity to retain NO<sub>3</sub><sup>-</sup>. Well-drained Ultisols (soil group 3) appear to accumulate NO<sub>3</sub><sup>-</sup> due to is high content of clays. However, when considering NH<sub>4</sub><sup>+</sup> + NO<sub>3</sub><sup>-</sup> fertilization will not impact soil group 3 and consequently long-term site quality is unlikely. Perhaps a potential solution to fix N losses by denitrification, leaching, as well as volatilization is to consider the application of slow release fertilizers.

In contrast, fertilization increased P in both O horizon and mineral soil. Fertilization increased the O horizon by 5.6, 6.1 and 6.0 kg ha<sup>-1</sup> in soil group 1, 2, and 3 respectively. The relatively large increases in Mehlich 3 extractable P in the mineral soil occurred in the poorly-drained Ultisols and poorly to somewhat well-drained Spodosols and Entisols by 29 and 60 kg P ha<sup>-1</sup>. Therefore, we reject the null hypothesis for O horizon and mineral soil and conclude that P fertilization did have a significant effect on soil P. However, very low P detected in soil group 3 indicates that sites in this group need to be fertilized in the early next rotation. This suggests that P fertilization has a long-term effect on soil P availability and will increase long-term site quality.

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## Tables and Figures

Table 1. Location and stand properties of installation of the regional fertilization study included in the study of fertilization effects on soil nutrients.

Site code	County and state	Latitude	Longitude	Specie	Study Establishment Year	Replications	Base Site Index*	Years since treatment (YST) in 2013	Stand age at establishment	Age at sampling
0101	Kershaw, South Carolina	34.45°N	80.5014°W	Loblolly pine	2000	2	60	14	4	18
0601	Brunswick, Virginia	36.67833°N	77.988911°W	Loblolly pine	1999	4	62	15	6	21
1101	Berkeley, South Carolina	33.1879°N	80.1932°W	Loblolly pine	1999	4	68	15	5	20
1502	Floyd, Georgia	34.1492°N	85.3772°W	Loblolly pine	2001	2	52	13	3	16
2201	Wilkes, Georgia	33.81°N	82.96°W	Loblolly pine	2000	2	63	14	3	17
2401	Nassau, Florida	30.6661°N	81.8361°W	Loblolly pine	1999	2	45	15	5	20
3901	Marengo, Alabama	32.3731°N	87.8386°W	Loblolly pine	2001	2	67	13	3	16
4201	Brantley, Georgia	31.3353°N	81.8217°W	Loblolly pine	1998	2	67	16	3	19
4202	Brantley, Georgia	31.3415°N	81.8338°W	Loblolly pine	1998	2	67	16	2	18

\*Site index in feet, base age 25 years, on unfertilized control plot.

Table 2. Site code, physiographic province, parent material, series names, soil drainage class, taxonomic description of dominant soil, CRIFF soil group, and study soil group at each site included in the study.

Site code	Physiographic Province	Parent material	Serie names	Soil drainage class	Taxonomy description	CRIFF Soil Group	Study Soil Group
0101	Sandhills	Sand	Blanton	Well	Loamy, siliceous, semiactive, thermic Grossarenic Paleudults	G	2
0601	Piedmont	Gneiss	Cecil	Well	Fine, kaolinitic, thermic Typic Kanhapludults	E	3
1101	Atlantic Coastal Plain	Marine sediments	Lynchburg	Somewhat poorly	Fine-loamy, siliceous, semiactive, thermic Aeric Paleaquults	B	2
1502	Ridge and Valley	Shale	Townley	Moderate well	Fine, mixed, semiactive, thermic Typic Hapludults	E	3
2201	Piedmont	Metadacite	Appling	Well	Fine, kaolinitic, thermic Typic Kanhapludults	E	3
2401	Flatwoods	Marine sediments	Meggett	Poorly	Fine, mixed, active, thermic Typic Albaqualfs	A	1
3901	Upper Gulf Coastal Plain	River terrace	Lenoir	Somewhat poorly	Fine, mixed, semiactive, thermic Aeric Paleaquults	A	1
4201	Flatwoods	Marine sediments	Leon	Somewhat poorly	Sandy, siliceous, thermic Grossarenic Alaquods	D	2
4202	Flatwoods	Marine sediments	Pottsburg	Sandy aeric	Sandy, siliceous, thermic Aeric Alaquods	D	2

Source: Official Soil Series Description – USDA NRCS Soil Survey Division and University of Florida IFAS Extension.

Table 3. Treatment code, N rate (kg ha<sup>-1</sup>), application frequency (years), cumulative N (kg ha<sup>-1</sup>), and cumulative P (kg ha<sup>-1</sup>) at the regional fertilization study over a 12-year period. Each site included a control (0) and three fertilized treatments.

Treatment code	N rate (kg ha <sup>-1</sup> )	Application frequency (years)	Cumulative N (kg ha <sup>-1</sup> )	Cumulative P (kg ha <sup>-1</sup> )*
0	0	0	0	0
412	135	4	540	54
418	202	4	807	81
424	269	4	1076	108

\*P fertilization was 10% of N.

Table 4. Detailed cumulative amounts of applied nitrogen (kg ha<sup>-1</sup>) for the regional fertilization study for a 12-year period at four years frequency of application.

Treatment	N rate (kg ha <sup>-1</sup> )	Application frequency (years)	Soil sampled at year 13																
			Years																
			0	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16
0	0	4	0	--	--	--	0	--	--	--	0	--	--	--	0	--	--	--	--
412	135	4	135	--	--	--	270	--	--	--	405	--	--	--	540	--	--	--	--
418	202	4	202	--	--	--	404	--	--	--	605	--	--	--	807	--	--	--	--
424	269	4	269	--	--	--	538	--	--	--	807	--	--	--	1076	--	--	--	--

Table 5. Selected mineral soil physical and chemical properties by depth from 0-100 cm at the regional fertilization study on a control plot basis for soil group 1: CRIFF A and B (Top); Soil group 2: CRIFF C, D, and G (Middle); and soil group 3: CRIFF E and F (Bottom). Percentages of sand and silt and clay, texture, bulk density (BD), pH (1:1 soil / water by volume), CEC (Cation Exchange Capacity), C (Carbon), N (Nitrogen), P (Phosphorus), C:N, K (Potassium), Ca (Calcium), Mg (Magnesium), Fe (Iron), Al (Aluminum), and Cu (Copper).

Depth (cm)	Sand (%)	Silt (%)	Clay (%)	Texture	BD (g cm <sup>-3</sup> )	pH 1:1 soil/water by volume	CEC (cmol <sub>c</sub> kg <sup>-1</sup> )	C (mg kg <sup>-1</sup> )	N (mg kg <sup>-1</sup> )	P (mg kg <sup>-1</sup> )	C:N	K (mg kg <sup>-1</sup> )	Ca (mg kg <sup>-1</sup> )	Mg (mg kg <sup>-1</sup> )	Fe (mg kg <sup>-1</sup> )	Al (mg kg <sup>-1</sup> )	Cu (mg kg <sup>-1</sup> )
Soil Group 1: A and B																	
0-15	81.5	11.3	7.2	Loamy sand	1.32	4.5	4.9	12337	608.5	9.5	20.3	13.6	121.9	13.9	143.2	626.3	0.1
15-30	78.4	11.8	9.8	Sandy loam	1.54	5.0	4.3	5607	258.3	4.4	21.4	8.7	117.5	16.4	106.9	698.1	0.1
30-50	74.1	13.1	12.8	Sandy loam	1.34	5.0	5.1	2824	186.2	2.3	15.7	10.3	198.3	37.3	60.7	580.7	0.1
50-75	71.5	9.7	18.9	Sandy loam	1.40	5.1	3.1	1871	151.6	1.2	14.1	17.2	251.9	69.5	48.5	502.1	0.1
75-100	67.4	8.9	23.8	Sandy clay loam	1.40	5.0	4.0	1559	143.9	0.7	10.6	22.1	303.9	98.1	32.1	530.6	0.1
Soil Group 2: C, D, and G																	
0-15	92.6	4.4	3	Sand	1.27	4.5	3.5	8688	303.5	7.8	28.6	4.6	21.9	4.8	48.1	305.9	0.1
15-30	91.6	5	3.4	Sand	1.59	4.6	2.1	5545	190.3	8.7	27.6	2.6	10.9	2.0	39.5	433.7	0.1
30-50	91.1	5.5	3.4	Sand	1.55	4.6	2.1	5326	174.5	2.9	27.7	1.9	15.9	3.9	31.6	384.0	0.1
50-75	89.5	6	4.5	Sand	1.45	4.7	1.9	5957	202.3	4.6	26.1	3.7	9.1	2.1	34.7	601.3	0.6
75-100	90.2	5	4.8	Sand	1.45	4.9	1.7	5019	167.9	4.6	26.4	2.2	5.1	0.8	29.0	631.6	0.1
Soil Group 3: E and F																	
0-15	54.5	26.2	19.3	Sandy loam	1.17	5.0	3.7	9205	425.9	1.6	21.6	26.9	115.6	27.8	66.4	547.1	0.4
15-30	39.3	22.7	38	Clay loam	1.43	5.1	5.1	4311	244.5	0.2	18.6	35.8	142.0	56.6	29.4	708.2	0.3
30-50	37.3	17.7	45	Clay	1.40	5.1	5.9	4058	263.9	0.1	16.6	46.9	120.6	60.0	22.6	770.9	0.2
50-75	28.2	20.4	51.3	Clay	1.40	5.2	6.0	2328	190.9	0.0	14.0	51.3	130.8	97.9	17.1	738.8	0.1
75-100	32	15.9	52	Clay	1.40	5.3	6.2	1771	184.5	0.0	11.5	49.9	117.1	104.3	14.3	726.4	0.2

Table 6. Mean and interaction effect p-values at the regional fertilization study O horizon total N and P concentration and content by CRIFF soil groups. Statistically significant p-values are shown in bold ( $\alpha=0.10$ ). † Indicates statistical analysis performed after logarithmic transformation.

Effect	p-value	
	O Horizon Mass	
	Mg ha <sup>-1</sup>	
Fixed Effect		
Soil Group	0.2634	
Fert	< <b>0.0001</b>	
Soil Group x Fert	<b>0.0521</b>	
	O Horizon N	
	g kg <sup>-1</sup>	kg ha <sup>-1</sup>
Fixed Effect		
Soil Group	0.7164	† 0.4171
Fert	< <b>0.0001</b>	† < <b>0.0001</b>
Soil Group x Fert	0.7071	† <b>0.1000</b>
	O Horizon P	
	mg kg <sup>-1</sup>	kg ha <sup>-1</sup>
Fixed Effect		
Soil Group	0.2419	0.1339
Fert	<b>0.0020</b>	< <b>0.0001</b>
Soil Group x Fert	0.1843	<b>0.0342</b>

Table 7. Main and interaction effect p-values at the regional fertilization study mineral soil total N concentration and content and KCl extractable  $\text{NH}_4^+$  and  $\text{NO}_3^-$  concentration and content by CRIFF soil groups. Statistical significant p-values are shown in bold ( $\alpha=0.10$ ). † Indicates statistical analysis performed after logarithmic transformation.

Effect	p-value	
	total-N	
	$\text{g kg}^{-1}$	$\text{kg ha}^{-1}$
Fixed Effect		
Soil Group	0.6062	0.8201
Fert	0.4805	0.5151
Depth	< <b>0.0001</b>	< <b>0.0001</b>
Soil Group x Fert	0.7917	0.7985
Soil Group x Depth	< <b>0.0001</b>	< <b>0.0001</b>
Fert x Depth	0.3203	0.5895
Soil Group x Fert x Depth	0.6881	0.6477
	KCl Extractable $\text{NH}_4^+$	
	$\text{mg kg}^{-1}$	$\text{kg ha}^{-1}$
Fixed Effect		
Soil Group	† <b>0.0894</b>	† 0.1322
Fert	† <b>0.0111</b>	† <b>0.0132</b>
Depth	† < <b>0.0001</b>	† < <b>0.0001</b>
Soil Group x Fert	† 0.8882	† 0.9423
Soil Group x Depth	† < <b>0.0001</b>	† < <b>0.0001</b>
Fert x Depth	† 0.9598	† 0.9703
Soil Group x Fert x Depth	† 0.3938	† 0.3406
	KCl Extractable $\text{NO}_3^-$	
	$\text{mg kg}^{-1}$	$\text{kg ha}^{-1}$
Fixed Effect		
Soil Group	† <b>0.0111</b>	† 0.1068
Fert	† <b>0.0656</b>	† <b>0.0508</b>
Depth	† < <b>0.0001</b>	† < <b>0.0001</b>
Soil Group x Fert	† 0.2790	† <b>0.0818</b>
Soil Group x Depth	† < <b>0.0001</b>	† < <b>0.0001</b>
Fert x Depth	† 0.2495	† 0.2213
Soil Group x Fert x Depth	† 0.7553	† 0.7530

Table 8. Main and interaction effect p-values at the regional fertilization study mineral soil inorganic N ( $\text{NH}_4^+ + \text{NO}_3^-$ ) and Mehlich 3-P concentration and content by CRIFF soil groups. Statistical significant p-values are shown in bold ( $\alpha=0.10$ ). † Indicates statistical analysis performed after logarithmic transformation.

Effect	p-value	
	Extractable $\text{NH}_4^+ + \text{NO}_3^-$	
	mg $\text{kg}^{-1}$	kg $\text{ha}^{-1}$
Fixed Effect		
Soil Group	† 0.1087	† 0.1429
Fert	† <0.1746	† <0.2058
Depth	† < <b>0.0001</b>	† < <b>0.0001</b>
Soil Group x Fert	† <0.9574	† <0.9203
Soil Group x Depth	† < <b>0.0001</b>	† < <b>0.0001</b>
Fert x Depth	† 0.8399	† 0.8323
Soil Group x Fert x Depth	† 0.9511	† 0.9234

Effect	p-value	
	Mehlich 3-P	
	mg $\text{kg}^{-1}$	kg $\text{ha}^{-1}$
Fixed Effect		
Soil Group	† <b>0.0013</b>	† <b>0.0008</b>
Fert	† <b>0.0013</b>	† <b>0.0008</b>
Depth	† < <b>0.0001</b>	† < <b>0.0001</b>
Soil Group x Fert	† 0.1933	† <b>0.0796</b>
Soil Group x Depth	† < <b>0.0001</b>	† < <b>0.0001</b>
Fert x Depth	† 0.2283	† 0.6985
Soil Group x Fert x Depth	† 0.5990	† 0.7338

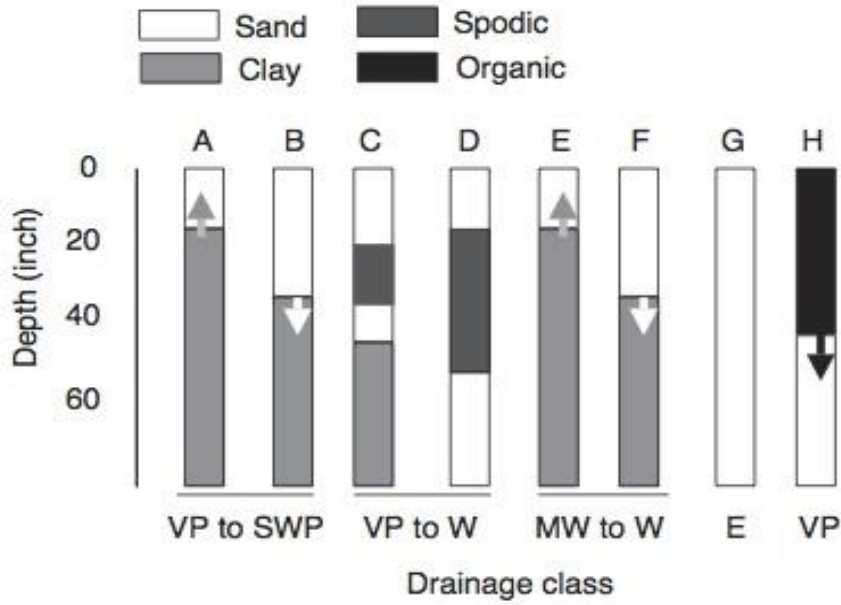


Fig. 1. Properties of the eight CRIFF soil groups: Soil texture and drainage class. VP = very poorly drained, SWP = somewhat poorly drained, W = well drained, MW = moderately well drained, E = excessively well drained. Figure adapted from Fox (2004b).

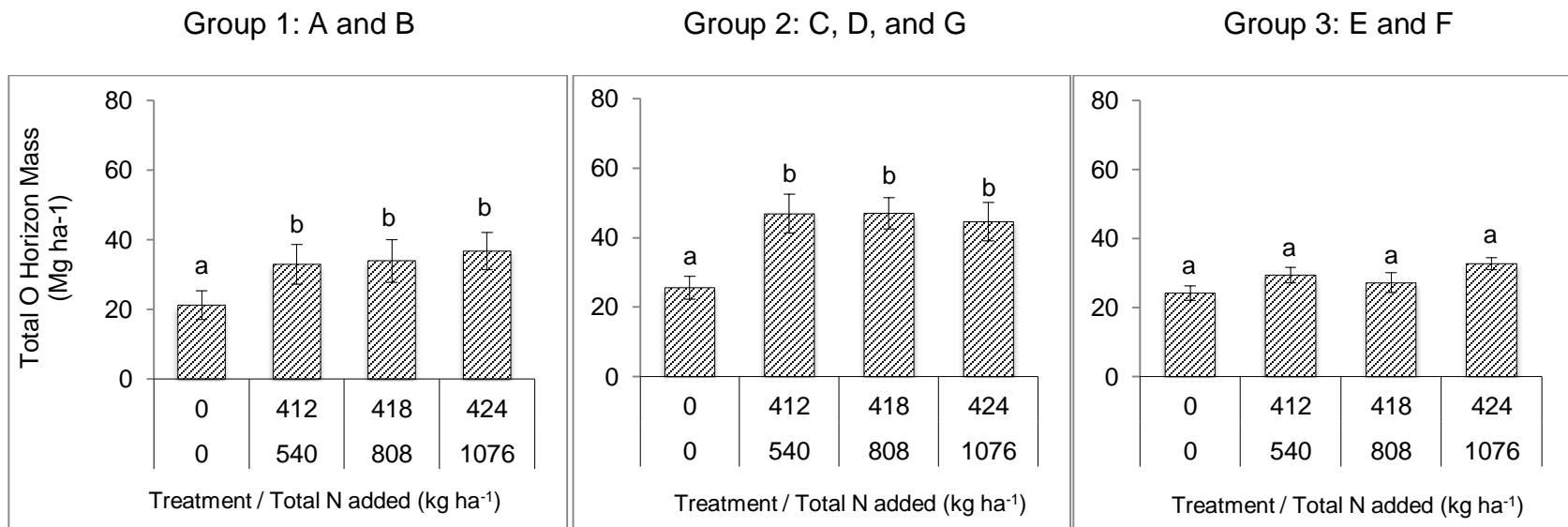


Fig. 2. Mean O horizon mass (Mg ha<sup>-1</sup>) by soil group 1: CRIFF A and B; soil group 2: CRIFF C, D and G; and soil group 3: CRIFF E and F. Different letters within a soil group indicate statistical significance at  $\alpha = 0.10$ . Bars represent standard error of the means.

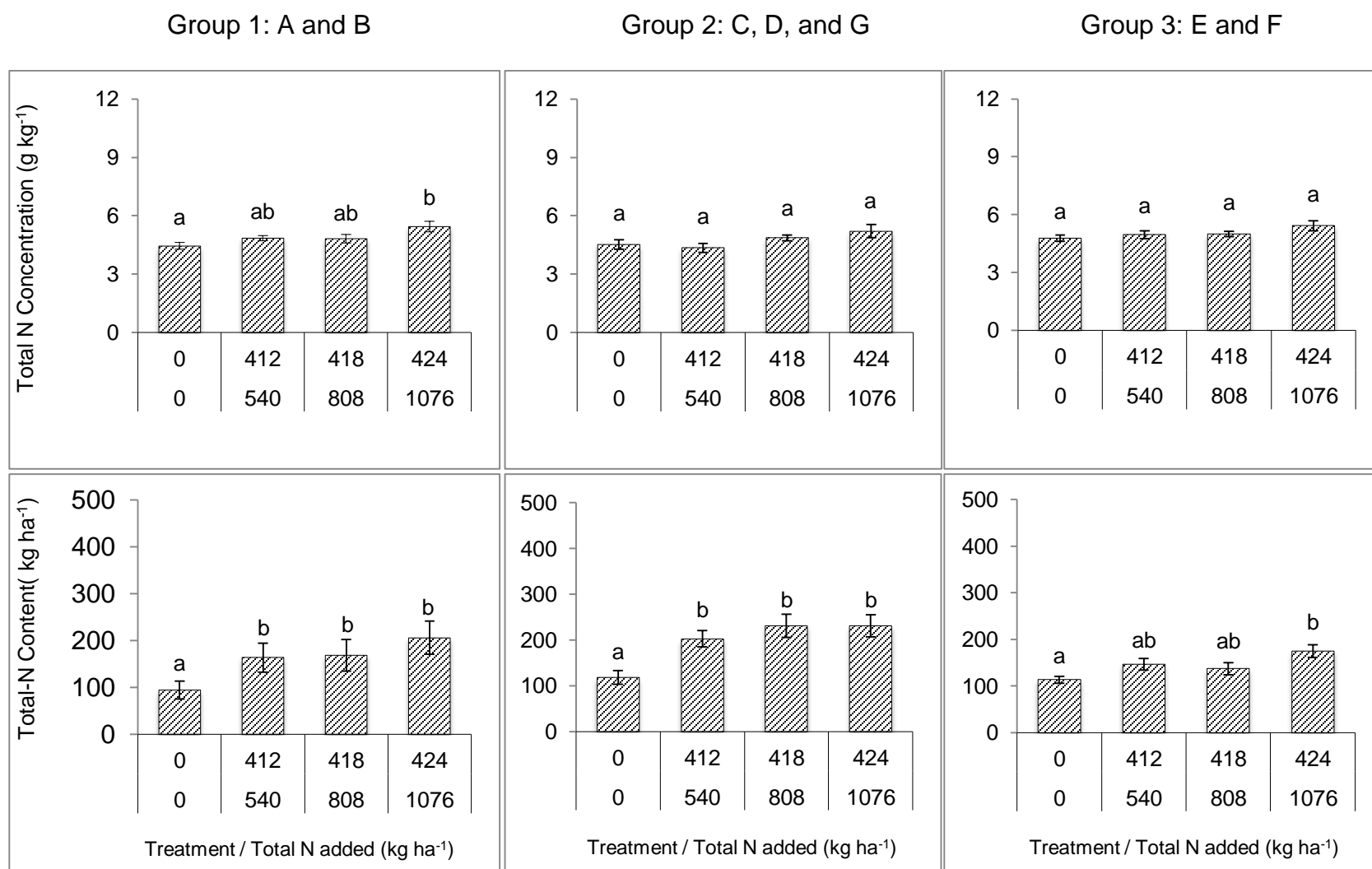


Fig. 3. Mean O horizon total N concentration (g kg<sup>-1</sup>) (Top) and content (kg ha<sup>-1</sup>) (Bottom) by soil group 1: CRIFF A and B; soil group 2: CRIFF C, D and G; and soil group 3: CRIFF E and F. Different letters within a soil group indicate statistical significance at  $\alpha = 0.10$ . Bars represent standard error of the means.

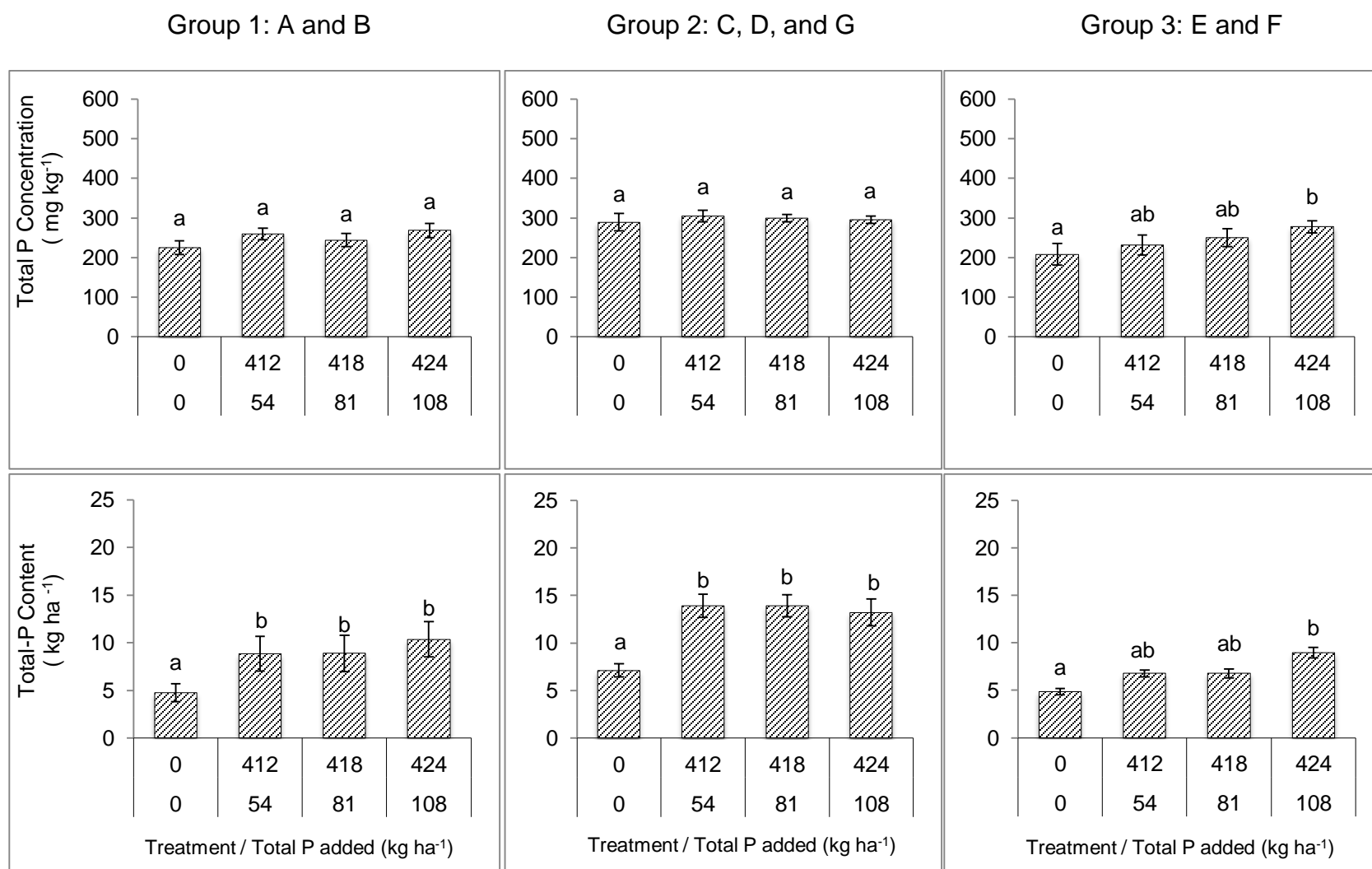


Fig. 4. Mean O horizon total P concentration (mg kg<sup>-1</sup>) (Top) and content (kg ha<sup>-1</sup>) (Bottom) for soil group 1: CRIFF A and B; soil group 2: CRIFF C, D and G; and soil group 3: CRIFF E and F. Different letters within a soil group indicate statistical significance  $\alpha = 0.10$ . Bars represent standard error of the means.

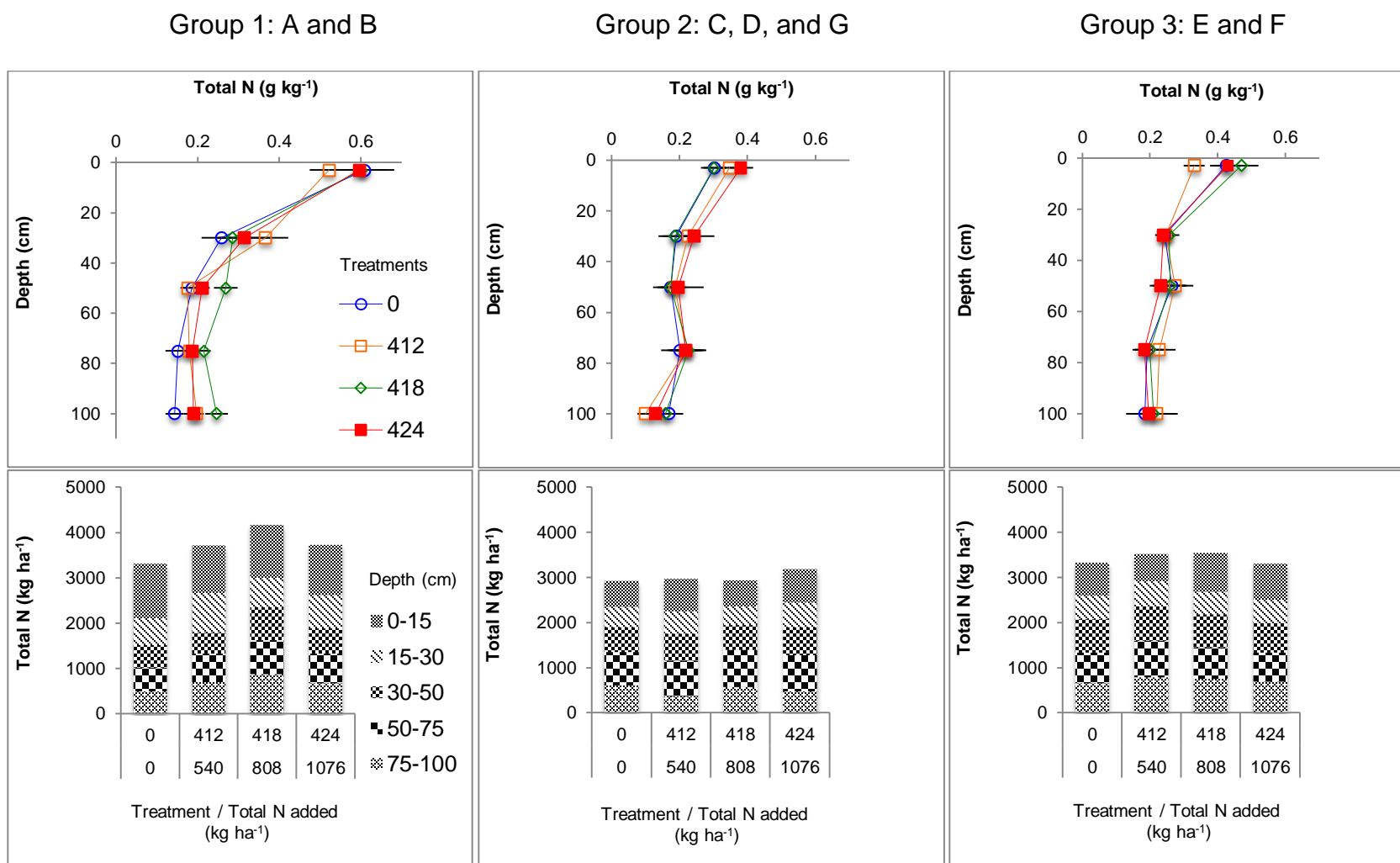


Fig. 5. Mean mineral soil total N concentration (mg kg<sup>-1</sup>) by depth (Top) and cumulative total N content (kg ha<sup>-1</sup>) to 1 meter (Bottom) for soil group 1: CRIFF A and B; soil group 2: CRIFF C, D, and G; and soil group 3: CRIFF E and F. Bars represent standard error of the means.

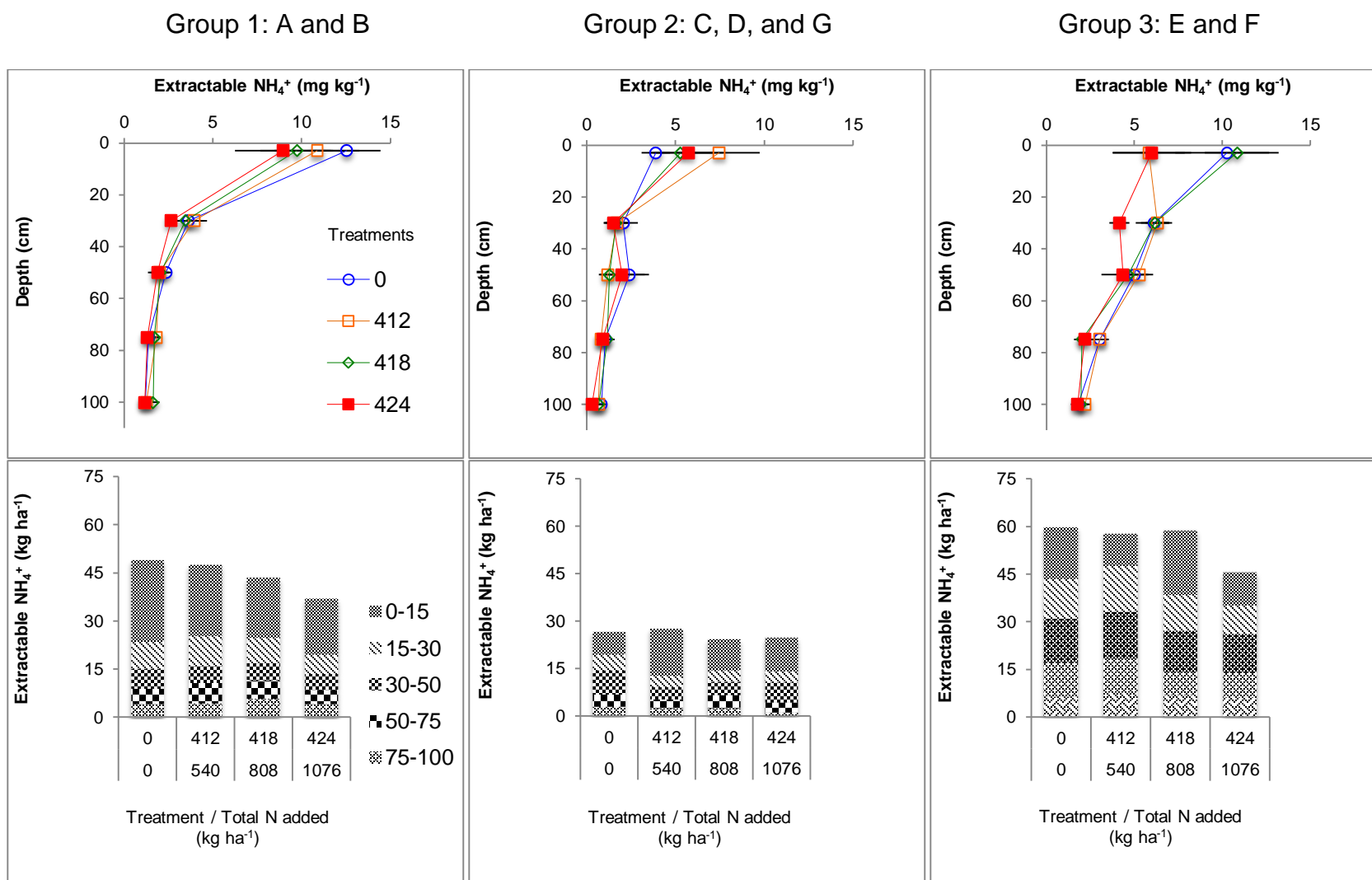


Fig. 6. Mean mineral soil extractable  $\text{NH}_4^+$  concentration ( $\text{mg kg}^{-1}$ ) by depth (Top) and cumulative extractable  $\text{NH}_4^+$  content ( $\text{kg ha}^{-1}$ ) to 1 meter (Bottom) for soil group 1: CRIFF A and B; soil group 2: CRIFF C, D, and G; and soil group 3: CRIFF E and F. Bars represent standard error of the means.

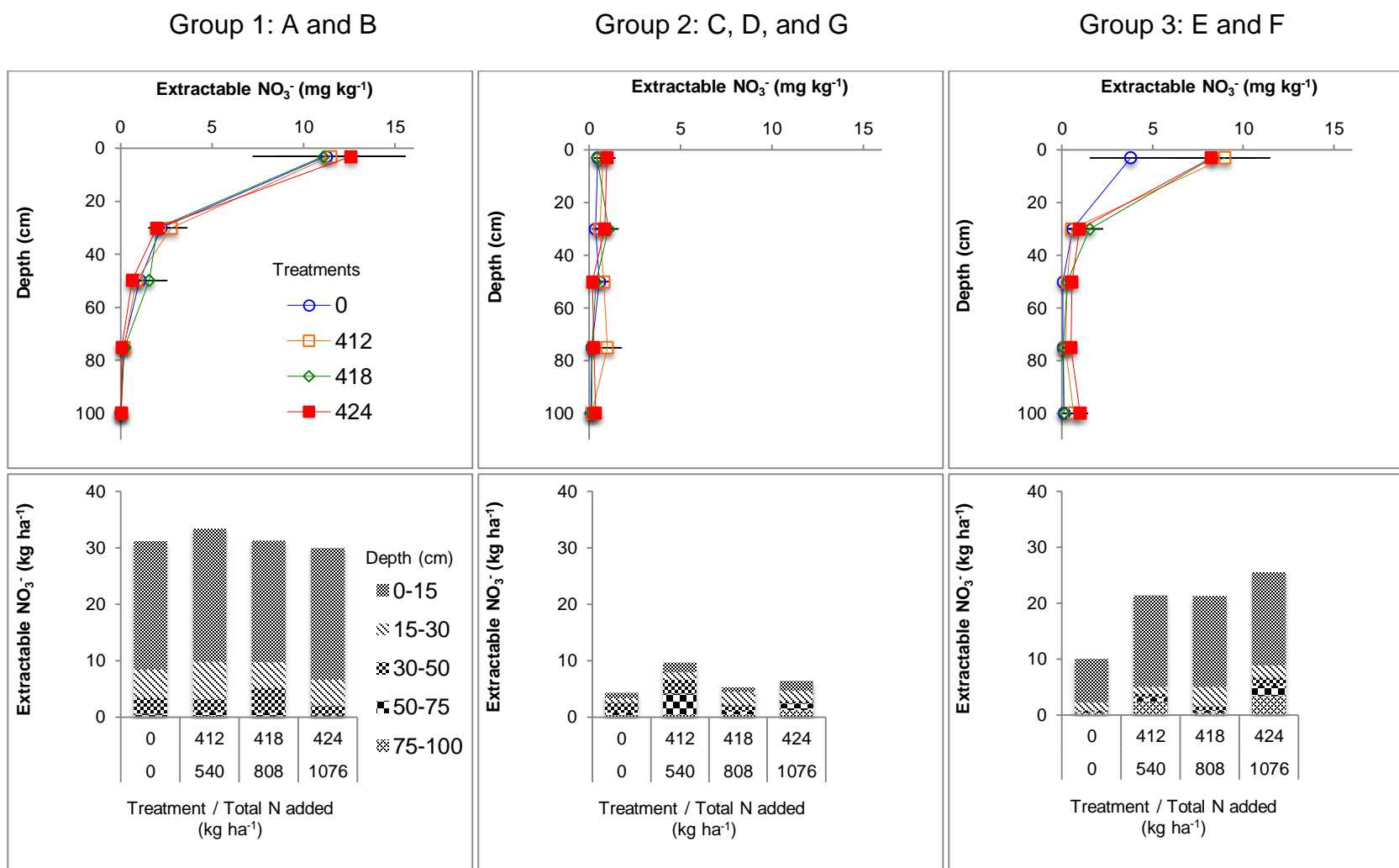


Fig. 7. Mean mineral soil extractable NO<sub>3</sub><sup>-</sup> concentration (mg kg<sup>-1</sup>) by depth (Top) and cumulative extractable NO<sub>3</sub><sup>-</sup> content (kg ha<sup>-1</sup>) to 1 meter (Bottom) for soil group 1: CRIFF A and B; soil group 2: CRIFF C, D, and G; and soil group 3: CRIFF E and F. Bars represent standard error of the means.

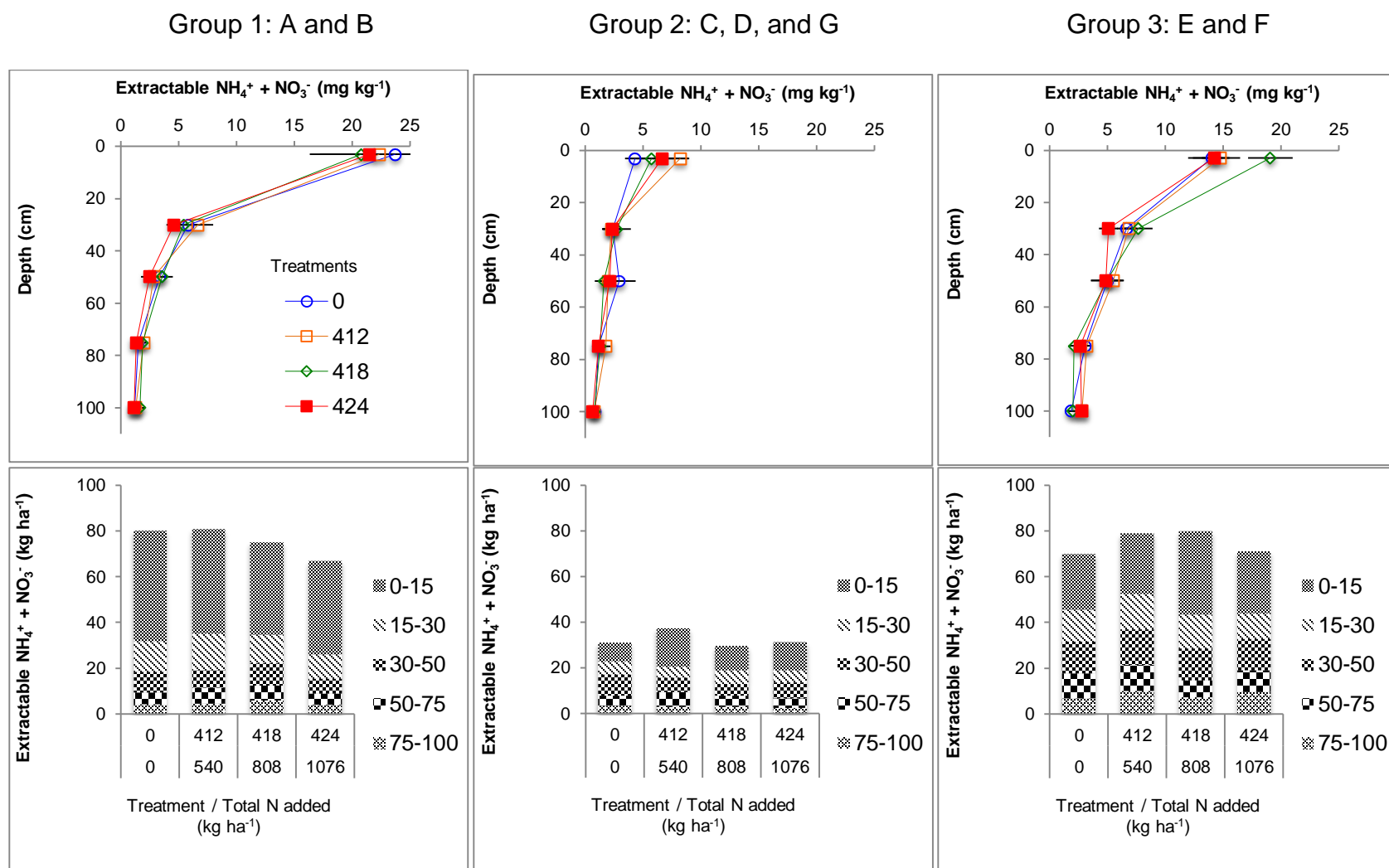


Fig. 8. Mean mineral soil extractable inorganic N ( $\sum \text{NH}_4^+ + \text{NO}_3^-$ ) concentration ( $\text{mg kg}^{-1}$ ) by depth (Top) and cumulative extractable inorganic N ( $\sum \text{NH}_4^+ + \text{NO}_3^-$ ) content ( $\text{kg ha}^{-1}$ ) to 1 meter (Bottom) for soil group 1: CRIFF A and B; soil group 2: CRIFF C, D, and G; and soil group 3: CRIFF E and F. Bars represent standard error of the means.

Group 1: A and B

Group 2: C, D, and G

Group 3: E and F

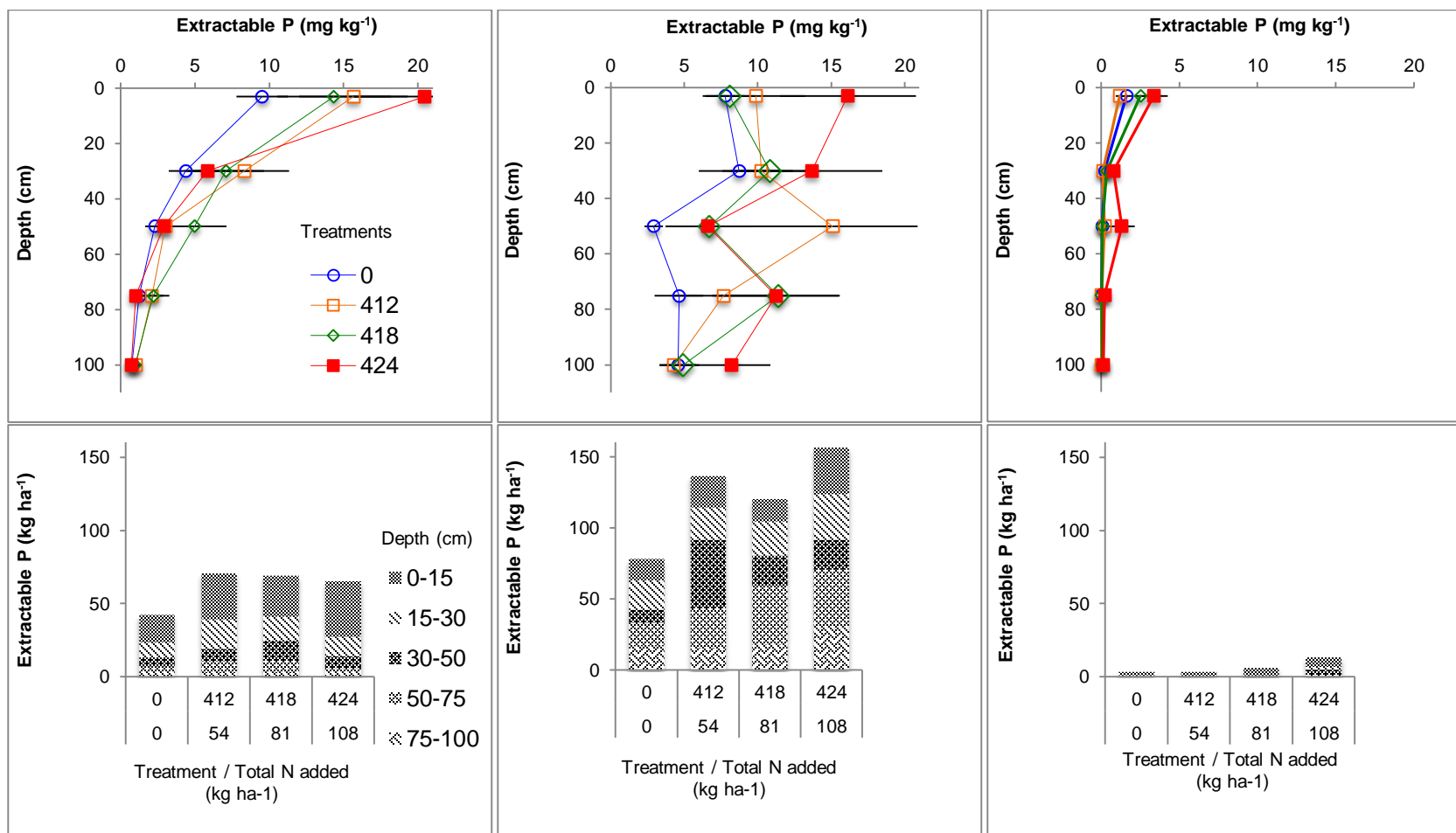


Fig. 9. Mean mineral soil Mehlich 3 extractable P concentration (mg kg<sup>-1</sup>) by depth (Top) and cumulative Mehlich 3 extractable P content (kg ha<sup>-1</sup>) to 1 meter (Bottom) for soil group 1: CRIFF A and B; soil group 2: CRIFF C, D, and G; and soil group 3: CRIFF E and F. Bars represent standard error of the means.

Group 1: A and B

Group 2: C, D, and G

Group 3: E and F

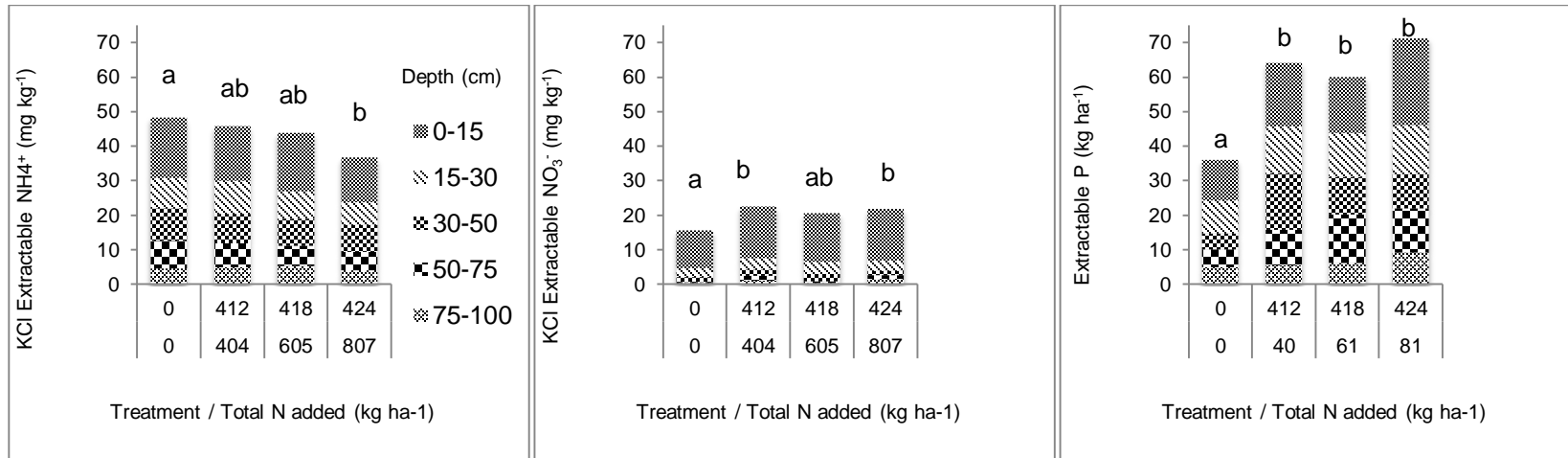


Fig. 2.10. Mean cumulative mineral soil KCl Extractable  $\text{NH}_4^+$  content ( $\text{kg ha}^{-1}$ ) (Left) and  $\text{NO}_3^-$  content ( $\text{kg ha}^{-1}$ ) (Middle) and Mehlich 3 extractable P content ( $\text{kg ha}^{-1}$ ) (Right) to 1 meter across all soil types. Different letters on the top the bars indicate statistical significance at  $\alpha = 0.10$ .

## Appendix A: O Horizon and Mineral Soil

Table A1. Mean O horizon mass, total N and P concentration and content at the regional fertilization study by soil groups: CRIFF A and B (n=8); CRIFF C, D and G (n=6); and CRIFF E and F (n=8). Standard deviations are indicated within parenthesis. Means followed by different lowercase letters are statistically significant ( $\alpha=0.10$ ). Results showed represent the total O horizons (Oi, Oe, and Oa).

CRIFF group	Fertilization treatment			
	0	412	418	424
O horizon Mass Mg ha <sup>-1</sup>				
A & B	21.3 (11.7) a	32.9 (16.1) b	34.0 (17.2) b	36.7 (15.1) b
C, D, & G	25.6 (8.0) a	46.9 (13.8) b	47.0 (11.1) b	44.6 (13.5) b
E & F	24.2 (6.0)	29.5 (6.3)	27.2 (8.2)	32.8 (4.9)
Total-N g kg <sup>-1</sup>				
A & B	4.45 (0.5) a	4.84 (0.4) ab	4.81 (0.6) ab	5.44 (0.8) b
C, D, & G	4.52 (0.7)	4.35 (0.7)	4.86 (0.5)	5.21 (1.0)
E & F	4.78 (0.6)	4.97 (0.8)	5.00 (0.5)	5.43 (1.0)
Total-N kg ha <sup>-1</sup>				
A & B	94.0 (54.9) a	164 (87.5) b	169 (96.3) b	206 (100) b
C, D, & G	118 (47.4) a	203 (58.5) b	231 (69.6) b	231 (77.9) b
E & F	114 (23.3) a	147 (45.1) ab	137 (46.0) ab	175 (17.0) b
Total-P mg kg <sup>-1</sup>				
A & B	225 (48.6)	260 (42.9)	244 (47.9)	268 (49.2)
C, D, & G	289 (63.0)	305 (42.1)	299 (21.5)	295 (24.0)
E & F	208 (68.9) a	232 (41.3) ab	250 (35.4) ab	278 (51.0) b
Total-P kg ha <sup>-1</sup>				
A & B	4.8 (2.7) a	8.9 (5.1) b	8.9 (5.4) b	10.4 (5.3) b
C, D, & G	7.1 (1.6) a	13.9 (2.8) b	13.9 (2.6) b	13.2 (4.2) b
E & F	4.9 (1.4) a	6.8 (1.6) ab	6.8 (2.0) ab	8.9 (1.1) b

Tables A2. Mean mineral soil total-N concentration (Top) and content (Bottom) at the regional fertilization study by soil groups: CRIFF A and B (n=8); CRIFF C, D, and G (n=6); and CRIFF E and F (n=8). Standard deviations are indicated in parenthesis.

CRIFF group	Depth (cm)	Fertilization treatment			
		0	412	418	424
total-N g kg <sup>-1</sup>					
A & B	0-15	0.61 (0.21)	0.52 (0.13)	0.60 (0.14)	0.60 (0.19)
	15-30	0.26 (0.14)	0.37 (0.16)	0.29 (0.10)	0.31 (0.19)
	30-50	0.19 (0.07)	0.18 (0.05)	0.27 (0.08)	0.21 (0.05)
	50-75	0.15 (0.08)	0.18 (0.05)	0.21 (0.05)	0.19 (0.06)
	75-100	0.14 (0.06)	0.20 (0.05)	0.25 (0.08)	0.19 (0.07)
C, D, & G	0-15	0.30 (0.08)	0.35 (0.09)	0.30 (0.09)	0.38 (0.09)
	15-30	0.19 (0.10)	0.23 (0.09)	0.19 (0.12)	0.24 (0.14)
	30-50	0.17 (0.09)	0.19 (0.08)	0.17 (0.07)	0.20 (0.18)
	50-75	0.20 (0.14)	0.22 (0.13)	0.23 (0.13)	0.22 (0.11)
	75-100	0.17 (0.11)	0.10 (0.06)	0.16 (0.08)	0.13 (0.02)
E & F	0-15	0.43 (0.13)	0.33 (0.09)	0.47 (0.15)	0.43 (0.12)
	15-30	0.24 (0.08)	0.25 (0.06)	0.26 (0.08)	0.24 (0.05)
	30-50	0.26 (0.11)	0.27 (0.16)	0.26 (0.14)	0.23 (0.09)
	50-75	0.19 (0.12)	0.23 (0.13)	0.20 (0.12)	0.18 (0.08)
	75-100	0.18 (0.16)	0.22 (0.17)	0.21 (0.19)	0.20 (0.10)
total-N kg ha <sup>-1</sup>					
A & B	0-15	1200 (400)	1041 (372)	1172 (322)	1108 (293)
	15-30	598 (325)	885 (401)	654 (237)	732 (441)
	30-50	500 (175)	468 (118)	728 (241)	569 (148)
	50-75	519 (263)	628 (167)	746 (148)	650 (186)
	75-100	494 (190)	693 (172)	870 (301)	669 (269)
	Total	3311 (384)	3715 (328)	4171 (306)	3728 (331)
C, D, & G	0-15	578 (175)	724 (262)	578 (198)	742 (170)
	15-30	454 (226)	498 (184)	414 (242)	556 (293)
	30-50	549 (305)	589 (265)	549 (238)	623 (586)
	50-75	733 (500)	796 (485)	823 (457)	796 (416)
	75-100	609 (383)	365 (210)	575 (298)	474 (87)
	Total	2923 (327)	2972 (320)	2938 (310)	3191 (353)
E & F	0-15	734 (227)	601 (105)	863 (315)	802 (241)
	15-30	523 (190)	556 (146)	492 (172)	513 (128)
	30-50	745 (336)	774 (461)	733 (420)	650 (261)
	50-75	676 (450)	811 (491)	706 (435)	647 (293)
	75-100	656 (572)	781 (641)	749 (693)	692 (371)
	Total	3334 (371)	3523 (415)	3543 (436)	3304 (273)

Table A3. Mean mineral soil KCl Extractable  $\text{NH}_4^+$  concentration (Top) and content (Bottom) at the regional fertilization study by soil groups: CRIFF A and B (n=8); CRIFF C, D, and G (n=6); and CRIFF E and F (n=8). Standard deviations are indicated in parenthesis.

CRIFF group	Depth (cm)	Fertilization treatment			
		0	412	418	424
Extractable $\text{NH}_4^+$ mg $\text{kg}^{-1}$					
A & B	0-15	12.54 (5.4)	10.86 (6.8)	9.72 (5.8)	8.95 (7.7)
	15-30	3.63 (2.4)	3.92 (2.1)	3.44 (1.4)	2.64 (0.9)
	30-50	2.35 (1.0)	2.00 (0.7)	2.04 (1.5)	1.87 (1.5)
	50-75	1.38 (0.8)	1.81 (0.7)	1.68 (1.0)	1.30 (0.9)
	75-100	1.19 (0.7)	1.24 (0.6)	1.62 (1.1)	1.16 (0.8)
C, D, & G	0-15	3.88 (2.0)	7.45 (5.6)	5.29 (4.4)	5.73 (4.8)
	15-30	2.04 (2.0)	1.68 (1.5)	1.67 (1.7)	1.49 (1.3)
	30-50	2.41 (2.7)	1.20 (0.8)	1.27 (1.2)	1.98 (3.1)
	50-75	1.04 (0.7)	0.79 (0.6)	1.16 (1.0)	0.90 (0.8)
	75-100	0.83 (0.7)	0.66 (0.8)	0.68 (0.8)	0.30 (0.3)
E & F	0-15	10.27 (8.3)	5.83 (5.8)	10.83 (5.2)	5.99 (6.3)
	15-30	6.11 (3.0)	6.31 (1.9)	6.16 (2.3)	4.15 (1.6)
	30-50	4.98 (2.3)	5.26 (2.3)	4.66 (1.6)	4.36 (3.5)
	50-75	3.04 (1.3)	3.04 (1.4)	2.03 (0.7)	2.16 (1.7)
	75-100	1.79 (0.7)	2.19 (0.9)	1.95 (1.2)	1.77 (1.1)
Extractable- $\text{NH}_4^+$ kg $\text{ha}^{-1}$					
A & B	0-15	25.6 (12.1)	22.1 (15.2)	18.8 (11.1)	17.3 (15.7)
	15-30	8.4 (5.5)	9.4 (4.8)	7.9 (3.6)	6.2 (2.4)
	30-50	6.3 (2.5)	5.3 (1.7)	5.4 (3.8)	4.9 (3.9)
	50-75	4.7 (2.4)	6.3 (2.2)	5.8 (3.3)	4.5 (2.9)
	75-100	4.1 (2.1)	4.3 (1.8)	5.7 (3.9)	4.0 (2.9)
	Total	49.1 (9.6)	47.4 (9.5)	43.6 (7.6)	37.0 (8.8)
C, D, & G	0-15	7.3 (3.8)	14.9 (10.8)	9.9 (8.4)	10.7 (8.7)
	15-30	5.0 (5.2)	3.7 (3.2)	3.7 (3.8)	3.4 (3.1)
	30-50	7.6 (8.6)	3.8 (2.7)	4.0 (3.9)	6.3 (10.1)
	50-75	3.8 (2.7)	2.9 (2.2)	4.2 (3.5)	3.3 (2.9)
	75-100	3.0 (2.5)	2.4 (3.1)	2.5 (3.1)	1.1 (1.1)
	Total	26.7 (5.1)	27.6 (7.0)	24.3 (5.3)	24.8 (6.7)
E & F	0-15	16.3 (10.8)	10.1 (9.0)	20.2 (10.6)	10.4 (10.7)
	15-30	12.5 (5.4)	14.4 (5.2)	11.3 (2.9)	9.0 (4.1)
	30-50	14.0 (6.5)	14.8 (6.7)	13.1 (4.5)	12.3 (9.9)
	50-75	10.7 (4.5)	10.7 (5.0)	7.1 (2.6)	7.6 (6.0)
	75-100	6.3 (2.6)	7.7 (3.3)	6.9 (4.3)	6.2 (3.9)
	Total	59.7 (7.1)	57.7 (6.4)	58.6 (7.4)	45.5 (7.4)

Table A4. Mean mineral soil KCl Extractable NO<sub>3</sub><sup>-</sup> concentration (Top) and content (Bottom) at the regional fertilization study by soil groups: CRIFF A and B (n=8); CRIFF C, D, and G (n=6); and CRIFF E and F (n=8). Standard deviations are indicated in parenthesis.

CRIFF group	Depth (cm)	Fertilization treatment			
		0	412	418	424
KCl Extractable NO <sub>3</sub> <sup>-</sup> mg kg <sup>-1</sup>					
A & B	0-15	11.22 (11.4)	11.48 (7.9)	11.07 (6.9)	12.59 (8.5)
	15-30	2.21 (1.6)	2.76 (2.5)	2.00 (1.2)	1.98 (1.4)
	30-50	1.03 (1.1)	0.86 (0.7)	1.56 (2.9)	0.63 (0.9)
	50-75	0.16 (0.3)	0.20 (0.3)	0.21 (0.3)	0.07 (0.1)
	75-100	0.02 (0.0)	0.04 (0.1)	0.03 (0.1)	0.03 (0.0)
C, D, & G	0-15	0.46 (0.3)	0.77 (1.0)	0.44 (0.5)	0.96 (1.1)
	15-30	0.35 (0.5)	0.59 (0.7)	1.06 (1.3)	0.86 (0.9)
	30-50	0.54 (0.9)	0.78 (0.9)	0.36 (0.6)	0.16 (0.3)
	50-75	0.14 (0.2)	1.00 (1.9)	0.13 (0.1)	0.27 (0.6)
	75-100	0.08 (0.2)	0.14 (0.2)	0.14 (0.3)	0.36 (0.6)
E & F	0-15	3.78 (6.4)	8.97 (7.2)	8.28 (6.6)	8.25 (6.3)
	15-30	0.57 (1.0)	0.54 (0.7)	1.54 (2.1)	0.97 (0.8)
	30-50	0.04 (0.1)	0.28 (0.4)	0.30 (0.3)	0.54 (0.4)
	50-75	0.05 (0.1)	0.19 (0.3)	0.09 (0.2)	0.50 (0.6)
	75-100	0.12 (0.2)	0.65 (1.7)	0.11 (0.2)	0.98 (1.3)
KCl Extractable NO <sub>3</sub> <sup>-</sup> kg ha <sup>-1</sup>					
A & B	0-15	22.7 (22.2)	23.6 (16.9)	21.6 (13.4)	23.4 (15.6)
	15-30	5.1 (3.6)	6.7 (6.3)	4.6 (2.7)	4.5 (3.0)
	30-50	2.8 (2.9)	2.3 (1.8)	4.3 (8.0)	1.7 (2.4)
	50-75	0.6 (1.0)	0.7 (0.9)	0.7 (1.0)	0.2 (0.4)
	75-100	0.1 (0.2)	0.2 (0.4)	0.1 (0.2)	0.1 (0.2)
	Total	31.2 (12.2)	33.4 (11.7)	31.3 (10.4)	29.9 (11.3)
C, D, & G	0-15	0.9 (0.7)	1.7 (2.4)	0.8 (0.9)	1.7 (2.0)
	15-30	0.8 (1.3)	1.3 (1.4)	2.4 (3.0)	1.9 (2.0)
	30-50	1.7 (2.8)	2.5 (2.9)	1.2 (2.0)	0.5 (0.9)
	50-75	0.5 (0.6)	3.6 (7.1)	0.5 (0.5)	1.0 (2.3)
	75-100	0.3 (0.7)	0.5 (0.8)	0.5 (1.2)	1.3 (2.2)
	Total	4.2 (1.5)	9.6 (3.6)	5.4 (1.8)	6.4 (1.9)
E & F	0-15	7.9 (13.8)	16.3 (12.9)	16.2 (14.4)	16.6 (14.8)
	15-30	1.4 (2.7)	1.3 (1.6)	3.5 (5.2)	2.2 (2.1)
	30-50	0.1 (0.3)	0.8 (1.1)	0.8 (0.9)	1.5 (1.2)
	50-75	0.2 (0.5)	0.7 (1.1)	0.3 (0.7)	1.7 (2.2)
	75-100	0.4 (0.6)	2.3 (5.8)	0.4 (0.8)	3.4 (4.4)
	Total	10.0 (6.7)	21.4 (8.6)	21.2 (9.0)	25.4 (8.9)

Table A5. Mean mineral soil KCl Extractable inorganic N ( $\text{NH}_4^+ + \text{NO}_3^-$ ) concentration (Top) and content (Bottom) at the regional fertilization study by soil groups: CRIFF A and B (n=8); CRIFF C, D, and G (n=6); and CRIFF E and F (n=8). Standard deviations are indicated in parenthesis.

CRIFF group	Depth (cm)	Fertilization treatment			
		0	412	418	424
KCl Extractable $\text{NH}_4^+ + \text{NO}_3^-$ mg $\text{kg}^{-1}$					
A & B	0-15	23.76 (15.5)	22.34 (13.8)	20.79 (10.1)	21.54 (14.6)
	15-30	5.84 (2.8)	6.69 (3.7)	5.44 (2.3)	4.61 (1.9)
	30-50	3.38 (1.5)	2.86 (1.0)	3.60 (2.6)	2.50 (2.0)
	50-75	1.54 (0.7)	2.01 (0.7)	1.89 (1.0)	1.37 (1.0)
	75-100	1.21 (0.7)	1.29 (0.6)	1.65 (1.1)	1.19 (0.8)
C, D, & G	0-15	4.34 (2.1)	8.22 (5.8)	5.74 (4.7)	6.69 (5.5)
	15-30	2.39 (2.0)	2.27 (1.9)	2.73 (3.0)	2.35 (2.0)
	30-50	2.95 (3.5)	1.98 (1.7)	1.63 (1.4)	2.14 (3.1)
	50-75	1.19 (0.8)	1.79 (2.1)	1.29 (1.1)	1.16 (0.9)
	75-100	0.92 (0.8)	0.79 (1.1)	0.81 (0.8)	0.66 (0.8)
E & F	0-15	14.05 (7.7)	14.79 (8.0)	19.12 (10.1)	14.24 (10.0)
	15-30	6.68 (2.3)	6.85 (2.4)	7.70 (1.7)	5.13 (2.0)
	30-50	5.02 (2.3)	5.54 (2.3)	4.96 (1.7)	4.90 (3.6)
	50-75	3.09 (1.2)	3.23 (1.4)	2.12 (0.8)	2.67 (1.6)
	75-100	1.91 (0.7)	2.84 (1.7)	2.06 (1.3)	2.75 (1.7)
KCl Extractable $\text{NH}_4^+ + \text{NO}_3^-$ kg $\text{ha}^{-1}$					
A & B	0-15	48.3 (31.6)	45.7 (30.7)	40.3 (19.5)	40.7 (28.9)
	15-30	13.4 (6.4)	16.0 (9.0)	12.5 (5.7)	10.8 (4.4)
	30-50	9.0 (4.1)	7.6 (2.5)	9.7 (7.2)	6.6 (5.1)
	50-75	5.3 (2.3)	7.0 (2.2)	6.5 (3.2)	4.7 (3.2)
	75-100	4.2 (2.2)	4.5 (1.9)	5.8 (3.9)	4.1 (2.8)
	Total	80.2 (20.7)	80.8 (20.6)	74.9 (16.1)	66.9 (19.0)
C, D, & G	0-15	8.2 (4.1)	16.6 (11.4)	10.7 (9.0)	12.4 (9.9)
	15-30	5.8 (5.1)	5.0 (4.0)	6.1 (6.7)	5.3 (4.4)
	30-50	9.3 (11.3)	6.3 (5.6)	5.2 (4.5)	6.8 (10.0)
	50-75	4.3 (2.8)	6.5 (7.6)	4.7 (3.9)	4.2 (3.3)
	75-100	3.3 (2.9)	2.9 (3.8)	2.9 (2.9)	2.4 (2.9)
	Total	30.9 (6.2)	37.2 (8.1)	29.6 (6.0)	31.2 (7.3)
E & F	0-15	24.1 (13.3)	26.5 (12.1)	36.4 (22.7)	27.0 (21.0)
	15-30	14.0 (4.2)	15.6 (6.3)	14.8 (4.4)	11.2 (5.6)
	30-50	14.1 (6.4)	15.6 (6.7)	13.9 (4.9)	13.8 (10.2)
	50-75	10.9 (4.4)	11.4 (5.1)	7.4 (2.8)	9.4 (5.7)
	75-100	6.7 (2.7)	10.0 (6.1)	7.3 (4.8)	9.7 (6.0)
	Total	69.8 (9.0)	79.0 (9.4)	79.9 (14.9)	71.0 (12.6)

Table A6. Mean mineral soil Mehlich 3-P concentration (Top) and content (Bottom) at the regional fertilization study by soil groups: CRIFF A and B (n=8); CRIFF C, D, and G (n=6); and CRIFF E and F (n=8). Standard deviations are indicated in parenthesis. Means in a row followed by different uppercase letters are significantly different.

CRIFF Group	Depth (cm)	Fertilization treatment			
		0	412	418	424
Mehlich 3-P mg kg <sup>-1</sup>					
A & B	0-15	9.52 (4.9)	15.66 (10.3)	14.35 (10.8)	20.43 (13.5)
	15-30	4.37 (3.2)	8.31 (8.5)	7.09 (7.3)	5.85 (4.4)
	30-50	2.30 (1.8)	3.01 (2.4)	4.99 (6.0)	2.89 (1.8)
	50-75	1.24 (0.9)	2.12 (2.1)	2.18 (3.1)	1.02 (0.6)
	75-100	0.75 (0.6)	1.02 (0.8)	0.96 (1.5)	0.70 (0.6)
C, D, & G	0-15	7.77 (3.3)	9.86 (8.2)	8.09 (4.5)	16.13 (11.4)
	15-30	8.74 (6.7)	10.26 (5.2)	10.81 (7.9)	13.67 (11.8)
	30-50	2.92 (1.5)	15.10 (14.1)	6.71 (7.3)	6.60 (7.0)
	50-75	4.64 (4.1)	7.67 (8.6)	11.38 (10.1)	11.26 (10.6)
	75-100	4.57 (3.0)	4.33 (2.5)	4.91 (2.8)	8.22 (6.5)
E & F	0-15	1.60 (1.6)	1.19 (0.7)	2.52 (1.2)	3.37 (2.5)
	15-30	0.23 (0.2)	0.13 (0.2)	0.36 (0.5)	0.80 (0.6)
	30-50	0.06 (0.1)	0.23 (0.4)	0.06 (0.1)	1.33 (2.3)
	50-75	0.04 (0.1)	0.05 (0.1)	0.02 (0.0)	0.22 (0.2)
	75-100	0.04 (0.1)	0.01 (0.0)	0.06 (0.1)	0.16 (0.2)
Mehlich 3-P kg ha <sup>-1</sup>					
A & B	0-15	18.7 (9.2)	31.4 (21.6)	27.9 (20.6)	37.7 (25.3)
	15-30	10.1 (7.3)	20.3 (20.5)	16.1 (17.3)	13.4 (10.3)
	30-50	6.4 (5.1)	8.2 (6.8)	13.8 (16.9)	7.9 (5.0)
	50-75	4.3 (3.2)	7.3 (7.2)	7.7 (10.9)	3.6 (2.2)
	75-100	2.6 (2.1)	3.4 (2.5)	3.4 (5.2)	2.5 (2.2)
	Total	42.1 (8.1)	70.6 (16.9)	68.9 (16.7)	65.1 (17.6)
C, D, & G	0-15	14.9 (6.8)	22.1 (20.9)	15.8 (10.0)	32.9 (24.8)
	15-30	20.8 (15.5)	22.9 (11.9)	24.1 (16.8)	32.2 (28.8)
	30-50	9.0 (5.0)	48.0 (45.4)	21.3 (23.5)	20.9 (22.5)
	50-75	16.8 (14.8)	27.8 (31.1)	41.2 (36.7)	40.8 (38.5)
	75-100	16.6 (11.0)	15.7 (9.0)	17.8 (10.1)	29.8 (23.6)
	Total	78.1 (11.1) A	136.6 (27.6) B	120.2 (22.2) B	156.5 (27.0) B
E & F	0-15	2.5 (2.0)	2.1 (1.2)	4.8 (3.2)	6.4 (5.0)
	15-30	0.5 (0.5)	0.3 (0.4)	0.7 (1.0)	1.6 (1.2)
	30-50	0.2 (0.3)	0.7 (1.2)	0.2 (0.3)	3.7 (6.6)
	50-75	0.1 (0.3)	0.2 (0.4)	0.1 (0.1)	0.8 (0.9)
	75-100	0.1 (0.2)	0.0 (0.1)	0.2 (0.3)	0.6 (0.6)
	Total	3.5 (1.3)	3.3 (1.1)	6.0 (2.3)	13.1 (4.2)