

STRATEGIC SOIL MANAGEMENT FOR RAINFED AGRICULTURE IN WEST
AFRICA

A Thesis

by

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ABSTRACT

Soil quality enhancements will be a vital link to food security as world populations are projected to swell by 2 billion people over the next 30 years. Efficient and productive agronomic practices will be essential for the subsistence farmers of Sub-Saharan Africa (SSA), where more than half of the estimated population growth is projected to occur. Conservation agriculture and fertilizers offer substantial soil fertility benefits if adopted appropriately for the local physical conditions, climates, crops, and farmer household requirements. The objective of this study is to investigate the effects of tillage method, cropping system, mineral fertilizer, and compost on soil quality in 4 major agricultural regions of Ghana. An ancillary objective is to determine if there was a significant difference in nutrient extraction concentrations from 0.1M HCl extract compared to a cold ultrapure water extract.

Two field experiments were established in 2011 each featuring a split-plot design with three replications per agro-ecosystem. In 2013, soils were collected and analyzed for concentrations of $\text{NO}_3\text{-N}$, $\text{NH}_4\text{-N}$, extractable organic nitrogen (EON), extractable organic carbon (EOC), and $\text{PO}_4\text{-P}$. Findings indicate that agro-ecosystem had a significant effect on soil nutrient concentrations, where the Guinea and Coastal Savannahs had the greatest residual inorganic N, the Forest had the greatest organic C and N, and the Coastal Savannah and Transition had the greatest P concentrations. Within the Coastal Savannah region maize only crop sequence was linked with the greatest amounts of inorganic N, application of urea fertilizer without a P source nor

compost correlated with the higher residual inorganic N, and the combination of triple superphosphate (TSP) with an N source was associated with greater inorganic P concentrations. In the Transition zone no-till was associated with high concentrations of inorganic N and P, while the maize-mucuna rotation had high residual inorganic N, and TSP fertilizer was closely linked to greater amounts of inorganic P. Finally, in the Guinea Savannah urea and compost additions are beginning to influence inorganic N soil concentrations and TSP was associated with inorganic P concentrations.

For my parents.

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NOMENCLATURE

SSA	Sub-Saharan Africa
NT	No tillage
ZT	Zonal or minimum tillage
TT	Traditional tillage
CA	Conservation agriculture
M	Maize
MC	Maize-cowpea rotation
MM	Maize-mucuna rotation
MCM	Maize-cowpea-mucuna relay
CM	Cowpea-maize rotation
MCI	Maize-cowpea intercrop
SOM	Soil organic matter
SOC	Soil organic carbon
EOC	Extractable organic carbon
TEN	Total extractable nitrogen
EON	Extractable organic nitrogen
DOC	Dissolved organic carbon
DON	Dissolved organic nitrogen

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1. INTRODUCTION

Over the next 30 years, world population is projected to grow by 2 billion people, with more than half of this growth expected to occur in Africa (United Nations Information Center for India and Bhutan [UN], 2013). One of the most pressing global challenges will be meeting the food requirements of 9.6 billion people by 2050. For the most food insecure regions of the world, this challenge will be particularly daunting. Agricultural productivity in Sub-Saharan Africa (SSA) already faces numerous socio-economic and natural constraints. Access to capital, land scarcity, and irrigation are among the primary socio-economic constraints to food production. Some of the critical physical limitations to African food security include climatic conditions and inherently poor soil fertility.

1.1 SOCIO-ECONOMIC CONSTRAINTS

Access to capital is a significant limitation to smallholder farms. Studies have found that capital has a direct influence on the ability of farmers to improve their soil quality and crop yield. Inputs of equipment, such as no-till drills, improved seed varieties and fertilizers are prohibitively expensive for the resource poor (Sanchez et al., 1997; Lal and Stewart, 2012). Indeed it is such a significant cost that even those that have the means to invest often hesitate due to uncertainty about the effectiveness of costly inputs. This observation is especially common when farmers were faced with other stressors such as erratic rains, frequent droughts, and a high incidence of weeds

and pests (Lal and Stewart, 2012). This suggests that farmers faced with multiple strains to crop production are cautious of spending cash resources.

While financial capital can directly influence soil quality by enabling farmers to purchase farm inputs, capital also indirectly influences soil quality by reducing cultivation pressures. For example, Nkonya et al. (2005) reported a positive correlation between Ugandan households with an off-farm income and soil nutrient balances. Nkonya et al. (2005) found that heads of household with off-farm income purchased food to supplement their family's dietary requirements, which decreased cropping intensity and soil nutrient mining on the farm. Therefore limited finances constrain agriculture by hindering investments in the farm and intensifying cultivation pressures.

The scarcity of land or inequity of land distribution is another key socio-economic limitation to African agriculture. Because most farmers in SSA are smallholder farmers, most farmers have limited farm yield and therefore little cash to re-invest in their operations. In Ghana, the average farm size is 1.6 ha (Oppong-Anane, 2001), which is similar to smallholder farms in neighboring Benin which averages 3.3 ha (Kherallah et al., 2001), and its northern neighbor, Nigeria, with a range of 0.7-2.2 ha (Apata et al., 2011). For these small scale farmers, purchasing improved seed varieties, mineral fertilizers, and other farm inputs is difficult.

Although development literature focuses almost exclusively on the plight and needs of the small scale farmer, it is important to note that large scale, commercial farming does exist in Sub-Saharan Africa, especially in Central and East Africa. In fact,

some nations are encouraging large scale mechanized farming as a path to modernization (Byerlee and Deininger, 2010), which can be evidenced by the recent influx of East Asian agricultural investors. For these large scale growers, finances and capital may not exert as great a constraint on crop production as it does for their subsistence farmer counterparts. Purchasing fertilizer and large mechanized equipment would not be difficult for these large operators. Examples of nations with rising numbers of large commercial farms are Ethiopia where the medium size of new land acquisitions is 700 ha, Sudan where the average area of new land acquisition mean size is 8,000 ha, and Liberia where the average area of new land acquisition is a staggering 60,000 ha (Byerlee and Deininger, 2010). Although large scale farming is on the rise, farm size remains highly inequitable. Only a small percent of farmers own large swathes of land, meanwhile the majority of farmers are small scale operators, cultivating less than 3 hectares. Therefore, although large scale farming is important to consider in terms of food security and nutrient mining, focus is still needed on the small scale farmer because she/he represents the vast majority of African growers.

It is also important to note that land scarcity in SSA is not a natural physical limitation, but rather a social constraint due to land tenure arrangements and distribution of land (Salami et al., 2010). Many argue that land scarcity, by way of both small farm size and the land tenure structure, is a constraint on African agriculture (Nkonya et al., 2005; Salami et al., 2010). Meanwhile, others present empirical evidence that land tenure is not limiting (Migot-Adholla et al., 1991) and others still reference the food

production achievements of the millions of small-scale farmers in China and Japan as a contradiction to this long standing view (Carr, 2013). This counter argument suggests that smallholder farmers in China and Japan overcame the constraint of small farm size by increasing per hectare production with the aid of national fertilizer subsidies, thereby allowing them to achieve high food production on small land holdings (Carr, 2013).

What is not mentioned in Carr's (2013) article is the influence that inherent conditions, such as climate and soil properties, have on crop yield when comparing Asian and SSA agriculture. Carr (2013), and those like him, raise a concern worth further exploration, however, the predominant view remains that limited land holding size of subsistence farmers in SSA significantly limits potential agricultural productivity and profit.

1.2 NATURAL CONSTRAINTS

Understanding limitations to SSA agriculture also requires understanding natural and physical properties. Current natural constraints include challenging climatic conditions and soils with poor nutrients. Furthermore, projections of natural constraints associated with various climate change scenarios predict a worsening of already difficult climatic conditions.

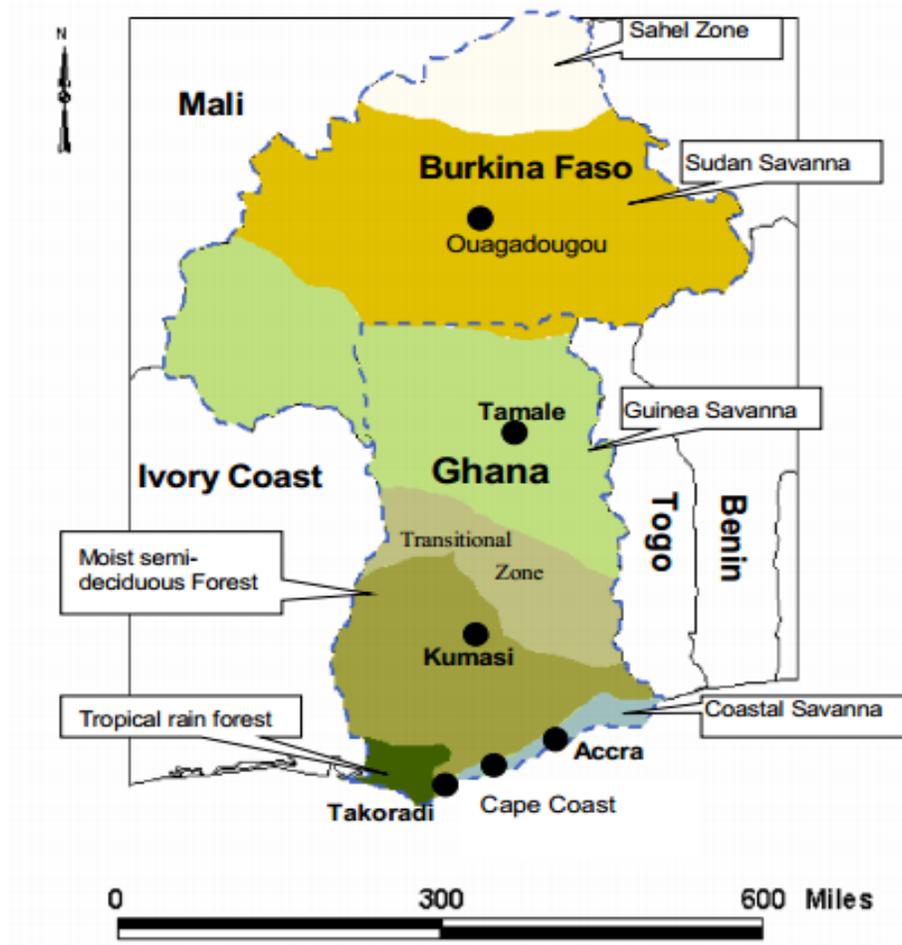


Fig. 1. Climatic zones of Ghana. Reprinted with permission from Obuobie et al., 2006.

Temperature and rainfall are known to exert great influence on plant growth. Warm temperatures, or Growing Degree Days (GDD), must be coupled with adequate moisture to ensure plant growth. There are six major climatic zones in Ghana: Rainforest, Deciduous Forest, Forest-Savannah Transition, Coastal Savannah, Guinea Savannah and the Sudan Savannah (Fig. 1; Oppong-Anane, 2001). Much of Ghana's

land area lies in the harsher climatic zones of the Transition zone and Guinea Savannah zone (Table 1). These regions typically have a distinct wet and dry season. During the rainy season, precipitation may be intense and erratic, which is not ideal for plant growth. The dry season is plagued by frequent and persistent droughts, sometimes for consecutive years (Food and Agriculture Organization [FAO], 2008). Lack of irrigation and its infrastructure mean that the vast majority of SSA farmers are dependent on this unreliable rainfall. According to Wani et al. (2012) more than 95 percent of SSA agriculture is dryland.

Currently in developing countries, rain-fed grain yields average 1.5 Mg ha^{-1} compared to irrigated yields of 3.1 Mg ha^{-1} (Rosegrant et al., 2002 as cited by Wani et al., 2012). Since the 1950s, water stress has increased in many regions (Wani et al., 2012) and climate change threatens to make attaining even the modest current yields more difficult in the future. According to the Intergovernmental Panel on Climate Change (IPCC) Climate Change report (Wani et al., 2012), SSA is estimated to lose 12% of its cultivation potential, especially in the Sudan-Sahelian zone. The University of Illinois and the International Food Policy Research Institute (IFPRI) combined 17 global climate change models to make predictions of temperature and precipitation changes over the next 40 years and their results indicate that although yield in Asia is likely to take a larger overall hit, SSA will be more affected because of lower present-day yields (Ringler et al., 2011). With limited effective precipitation now and decreases likely in many parts of SSA efficient use of soil water will be paramount.

Table 1. Annual precipitation in the climatic regions of Ghana.

Region	Area (‘000 ha)	Percent of total area	Mean annual rain (mm)	Growing period (days)	
				Major season	Minor season
Rain Forest	750	3	2,200	150-160	100
Deciduous Forest	740	3	1,500	150-160	90
Transition	6,630	28	1,300	200-220	60
Guinea Savannah	14,790	63	1,100	180-200	-
Sudan Savannah	190	1	1,000	150-160	-
Coastal Savannah	580	2	800	100-110	60

Source: Amended from Oppong-Anane, 2001.

The soils of SSA are another critical natural factor. Alfisols, Ultisols and Entisols are the most abundant soils in SSA, particularly in West Africa. These soils tend to have low inherent fertility due to natural properties rather than by virtue of nutrient mining or other anthropogenic degradation of fertility. Such natural properties could include age of soils, weathering, and parent material. The soils of interest in this study are considered Alfisols by U.S. Taxonomy.

Major constraints of Alfisols include low water holding capacity, poor nutrient reserves, low cation exchange capacity, low organic matter content (particularly in cultivated soils), and poor soil physical characteristics (Cogle, 1997). Examples of the latter are a tendency to surface seal, or crust, and hardset on drying, low soil strength under saturated soil conditions leading to slumping, increased bulk density, and loss of surface roughness (Cogle, 1997). The International Union of Soil Scientists' (IUSS) World Reference Base (WRB) has a more narrow classification for highly weathered

soils. The soils of interest in this study are classified as Lixisols and Plinthisols (IUSS Working Group WRB [WRB], 2006), which are both Alfisols by U.S. Taxonomy.

Lixisols are soils with a pedogenic clay differentiation where the top soil has lower clay content than the underlying argic subsoil horizon. At certain depths the Lixisols have low activity clays with a high base saturation. These soils can be developed from a wide variety of parent materials, but typically feature unconsolidated highly weathered and leached fine-textured materials. Lixisols are typically found in tropical, subtropical, or warm temperate climates with a pronounced dry season, especially on old erosional or depositional surfaces. Lixisols are thought to have been formed under a more humid climate in the past. Lixisols under natural savannah or woodland vegetation are suitable and frequently used for grazing. Degraded Lixisols have low aggregate stability and are prone to slaking or erosion when the surface is left exposed. These soils have low absolute levels of plant nutrients and low cation retention, making inputs of fertilizers and/or lime a precondition for continuous cultivation. Also, perennial crops are generally preferred to annual crops on these soils (WRB, 2006).

Plinthisols are soils characterized by the presence of plinthite, petroplinthite or pisoliths. Plinthite is an iron rich, humus-poor mixture of kaolinite clay (or other highly weathered clay) with quartz and other constituents that upon repeated wetting and drying change irreversibly to a layer with hard nodules or a hardpan. Petroplinthite is a continuous, fractured or broken sheet of connected, cemented nodules or mottles. And pisoliths are discrete strongly cemented indurated nodules. The parent material of

Plinthisols is typically a basic rock with significant iron content either from parent material or alluvium or colluvium deposits and are usually situated on level to gently sloping areas with fluctuating groundwater or stagnating surface water. Plinthisols are found extensively in the Sudano-Sahelian zone, Southern African savannah, Indian subcontinent and drier regions of Southeast Asia. They have poor natural soil fertility due to weathering and waterlogging. Many are shallow soils with highly root limiting horizons. These soils are best suited for grazing, as under cultivation crops are likely to suffer from drought in the dry season (WRB, 2006).

1.3 INVESTIGATION AIMS

Food security is a major global challenge of the next 30 years. In SSA, there are numerous constraints to food production. Comprehensive national strategies will be required of each African nation to address the varied social, economic, and natural challenges to farming within their country. Part of that national strategy ought to be agronomic innovation.

Conservation agriculture (CA) is often mentioned as a practical, low cost, and sustainable method of stabilizing soil and maximizing yields. Although CA may have many benefits leading to more sustainable agricultural production, fertilizer is still critical for provision of optimal yields. Because fertilizer is very expensive for the smallholder, application rates should be optimized for greatest efficiency. In addition, low cost, organic fertilizer sources should be utilized whenever possible.

The aim of this study is to increase understanding of the benefits of conservation agriculture, mineral fertilizers, and organic inputs, and recognizes the importance of investigating agronomic practices under local conditions. Field trials were established in four regions of Ghana utilizing the indigenous farming practices of each region to the fullest extent possible. To meet the food demands of the present and prepare for the future, one must build upon local knowledge with innovative technologies.

2. APPLYING CONSERVATION AGRICULTURE TO SMALL HOLDER FARMING IN GHANA

2.1. INTRODUCTION

Smallholder farmers in Sub-Saharan Africa (SSA) stand to gain improved crop yield, income, and health from agronomic innovations. Numerous agricultural technologies are touted as being integral to increased food security in developing countries. Conservation agriculture (CA) is one such technology. Conservation agriculture has three fundamental principles: (i) minimum soil disturbance, (ii) diversification of crops and (iii) permanent organic soil cover (FAO, 2008). Conservation agriculture is often promoted because of its potential benefits to soil quality, water holding capacity, and yield. However, nutrient-depleted agricultural systems of the Tropics have reported varied nutrient responses to CA practices. More CA research is needed in rainfed, sub-humid and tropical regions, such as Sub-Saharan Africa (SSA), to determine adoption benefits to the smallholder farmer in local regions. This chapter will focus on the first two fundamental principles of CA: tillage and crop diversification.

2.1.1. Tillage

One of the key components of CA is minimum soil disturbance. This can be achieved either through reducing tillage or practicing no-till. Some of the key benefits of reduced soil disturbance include increased soil organic carbon (SOC) (Zibilske et al.,

2002), increased soil biological activity (Weil et al., 1993), improved soil physical structure, and enhanced soil moisture (Franzluebbers et al., 1995). In addition, when soil is disturbed, aggregates are broken leading to increased evaporation and lowered soil water storage (Troeh and Thompson, 2005). On the other hand, breaking up the soils also has benefits. For example, tillage can incorporate manufactured fertilizers and organic inputs, potentially making nutrients more available for plant uptake. Tillage can also break compaction and aerate the soil promoting mineralization and nitrification. Because of the competing advantages and disadvantages associated with tillage, several studies have attempted to evaluate its effects. Previous research in tropical dry land agricultural systems have demonstrated mixed results from reducing tillage. Some have observed improvements to soils and crops from plots managed with no-till (Ikpe et al., 1999; Lal, 2007; Zibilske et al., 2002), while others have not necessarily found such clear results (Barbera et al., 2012; Dick et al., 1991; Wander et al., 1998; Yang and Wander, 1999; Alvarez et al., 1998; Carter and Rennie, 1982; Angers et al., 1995, 1997; Deen and Kataki, 2003; Yang et al., 2008).

On an Alfisol in semi-arid Niger, tilled fields had higher millet yield than plots under no-till (Ikpe et al., 1999). This result could be because of enhanced nutrient availability under tillage. As noted by Lal (2007), tillage can incorporate residues and fertilizers supporting soil N mineralization. Another reason for the higher millet yield in the Ikpe et al. (1999) study could be due to the improved soil tilth from tillage. Tillage

can reduce compaction of soils prone to surface sealing, improving porosity and structure (Lal, 2007).

Regions of the world without low inherent soil fertility have also found mixed carbon sequestration results from practicing no-till. For example, some studies have found no net change to SOC stocks in soils under no-till compared to moldboard plowed soil, such as in Sicily (Barbera et al., 2012), the Midwest USA (Dick et al., 1991; Wander et al., 1998; Yang and Wander, 1999), eastern Canada (Carter and Rennie, 1982; Angers et al., 1995, 1997; Deen and Kataki, 2003) and Argentina (Alvarez et al., 1998). These studies indicated that no-till only served to alter where SOC accumulated. SOC that typically accumulates beneath the plow layer, accumulated near the surface when no-till was practiced (Yang et al., 2008 as cited by Barbera et al., 2012).

A delayed benefit to soil is another challenge with no-till. It has been noted crops perform much better when no-till has been practiced for several years (Rhoton, 2000). Unfortunately, few SSA tillage studies are long term, so it is not clear whether there are long-term no-till benefits in SSA. Insufficient long-term SSA research is likely due to many factors including funding limitations. For the subsistence farmer, delayed crop benefits may not be realistic; an initial period of depressed crop yield may lead to abandonment of no-till in favor of a tillage method that offers more immediate crop benefits.

Minimum tillage is another option for reducing soil degradation due to tillage disturbance. Similar to the varied no-till results, results of experiments on reduced tillage

in tropical dryland agricultural systems are also mixed. Some researchers found that conventional tillage led to greater soil and crop gains (Paul et al., 2013), while others found that reducing tillage had greater benefits (Sommer et al., 2011). For example, a study on a Ferrasol in sub-humid Kenya found higher soil C and residue retention under conventional tillage when compared to reduced tillage with or without residue retention at the 15-30 cm soil depth (Paul et al., 2013). The results of Paul et al. (2013) indicate that the deep and vigorous disturbance associated with conventional tillage better incorporated organic residues, ultimately leading to greater soil C. Conversely, in the drier semi-arid climate on a Syrian Vertisol, positive results were found from reduced till (Sommer et al., 2011). In a study comparing shallow and deep tillage in combination with four residue management systems, Sommer et al. (2011) determined that shallow tillage, not deep tillage, increased organic matter across all residue management scenarios. It was also reported that shallow tillage combined with residue incorporation and compost additions every two years had the highest soil organic matter (SOM) content of all the tillage and residue scenarios (Sommer et al., 2011). These results suggest that shallow tillage leads to better soil organic matter sequestration than deep tillage and that that shallow tillage plus compost provides the highest SOM sequestration of all.

Conflicting results of studies evaluating tillage methods on crop yields and soil fertility suggests that other factors may play a role in determining which tillage practices are best. Based on the literature some of these factors are: climate, the initial fertility

status of the soil, the quality or type of organic inputs, and addition of mineral fertilizer. Based on this, the promotion of various tillage methods ought to be appropriate for the specific climate, soils, and cropping systems where it is empirically shown to be effective.

2.1.2. Crop Diversity

The diversification of crops grown in sequences or associations is the second component of CA. Some rotational sequences feature a single crop grown during one growing season followed by a different crop in the next growing season. This is referred to as successional cropping. Intercropping, alley cropping, companion cropping, and agroforestry exemplify rotations where two or more crops are grown during the same growing season within the same parcel of land. Relay cropping is a hybrid between successional and intercropping where a crop is seeded into the previous crop before the first crop is harvested.

Conservation crop rotation not only involves the manipulation of cropping sequences but also altering the selection of crops in order to achieve greater crop complementarity. Increasing crop diversity can contribute organic matter, fix atmospheric N, improve soil water storage, reduce erosion and enhance yields. Some crop rotations, however, particularly those with intercropping can result in greater competition for water and nutrient resources between crops and impede yields (Van Duivenbooden et al., 2000). Investigations into crops that are well suited to be grown together are important precursors to recommending crop rotations.

SSA subsistence farmers often practice forms of intercropping (Van Duivenbooden, 2000). In West Africa two dryland farming systems dominate: Cereal-root mixed farming and agro-pastoral millet-sorghum (*Sorghum bicolor* L Moench) farming. In the cereal-root farming, maize, sorghum and millet are grown along with yams (*Dioscorea* sp.) and cassava (*Manihot esculenta*). In the agro-pastoral system sorghum and pearl millet are the main cereal grains while sesame and legumes are common (Kofi Boa, personal communication, December 2013).

In East Africa forage legumes have not been widely adopted in semi-arid dryland farming (Rao and Mathuva, 2000). When legumes are included in crop rotations their importance is to fix atmospheric N which improves the soil nutrient balance. Research has shown, however, that grain legumes, such as soybean and cowpea, contribute less nitrogen to the soil than herbaceous legumes (Giller et al., 1997). Giller et al. (1997) suggested that this was due to the translocation of biologically fixed-N in root nodules to the grain of the plant where it is then removed by humans or livestock for consumption and not incorporated into the soil as litter. Although grain legumes may be poorer contributors to soil N, they are often preferred by farmers primarily in mixed crop-livestock systems because of their dual-purpose of food and feed (Rao and Mathuva, 2000). Herbaceous legumes such as *Mucuna pruriens*, have shown promise as a high-quality addition to farming systems. Significant increases in maize yields following mucuna have been demonstrated in the literature (Kaizzi et al., 2004; Versteeg et al., 1998; Fischler, 1999). Inclusion of legumes with a sole purpose of improving soil

fertility, will likely face adoption barriers, however, as farmers cannot afford to grow them at the expense of food crops (Rao and Mathuva, 2000).

Low cost methods of improving subsistence agriculture need empirical investigation. More than 80% of the world's population are living in developing countries (World Bank) and are depending on new technologies and improved management practices to help them increase their livelihoods. Unfortunately, because most agricultural research has been conducted in developed countries, many of the sub-humid and tropical regions lack the robust research necessary to identify the most appropriate farm management for specific geographic regions. Conservation agriculture presents an opportunity for low cost improvement to agricultural production if an effective combination of reduced tillage, crop diversity, and organic inputs are selected. The factors that will determine the effectiveness of CA for increasing crop yields and soil fertility include the climate, soils, geography, cropping system, mechanization, access to organic inputs, and fertilizers.

2.2. OBJECTIVES & HYPOTHESIS

The objective of this study was to evaluate the effect of tillage and cropping systems on extractable soil nutrient concentrations in four major agricultural agro-ecosystems of Ghana, West Africa. There were three hypotheses for this objective: Hypothesis 1. In moderate and low rainfall regimes, conservation tillage such as zonal till, will result in highest soil fertility quantified as 0.1M HCl extractable nutrients

because of its ability to reduce compaction and hard setting without complete soil disaggregation.

Hypothesis 2. For a high rainfall regime, where hard setting and compaction are less common, no tillage will enhance soil fertility, quantified by significantly higher soil extractable C, N and P, by preventing soil water storage depletion.

Hypothesis 3. Crop diversity with the addition of a legume will increase soil fertility quantified by higher extractable soil N species relative to a non-legume monoculture because of the ability of legumes to biologically fix nitrogen.

2.3. METHODOLOGY

2.3.1. Experimental Sites

The project was established in January 2011 in the Coastal Savannah (5°42'6N, 0°17'15W), Forest (6°34'11N, 1°51'26W), Forest-Guinea Savannah Transition (7°26'12.6"N, 1°29'31.6"W), and Guinea Savannah (9°31'18N, 0°55'9W) agro-ecosystems of Ghana.

2.3.1.1. Coastal Savannah

The Coastal Savannah experimental site was located in the Ga West District near the town of Pokuase. It lies on the southern coast of Ghana, north of the Capital of Accra (Fig. 2). The research plot lies adjacent to a wooded area and was at a shoulder position in the landscape. Mean annual rainfall in the Coastal Savannah is 800 mm (Oppong-Anane, 2001). At this site the primary growing season is 100-110 days long and runs

from March to June, while the minor growing season is about 60 days starting in October (Oppong-Anane, 2001).

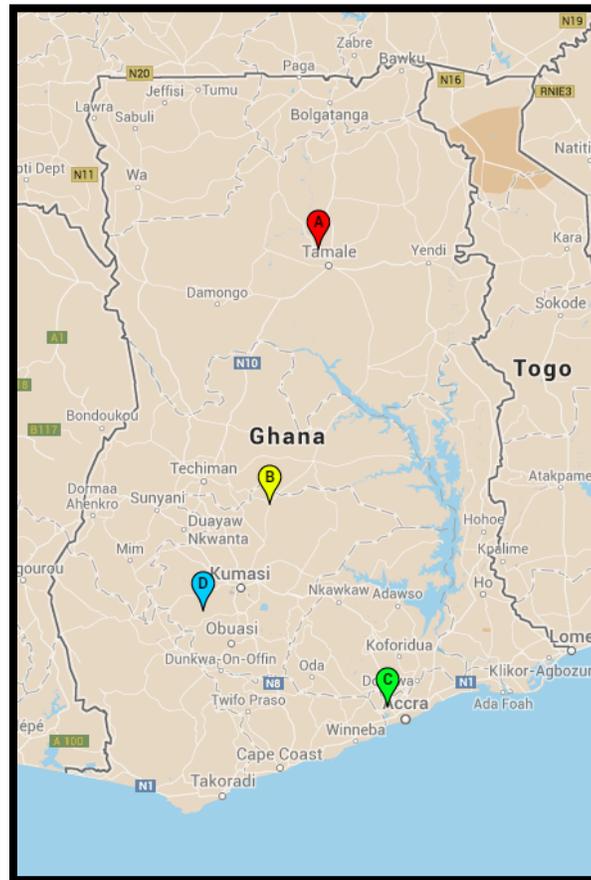


Fig. 2. Experimental plot site locations in Ghana. “A” was the Guinea Savannah zone site; “B” was the transition zone site; “D” was the forest zone site; and “C” was the coastal savannah zone site. Source: Developed by L. K. R. Pitts.

Soils at this site were classified using the World Reference Base (WRB) soil classification system as a Haplic Lixisol by the Ghanaian government's Soil Research Institute (SRI). Haplic lixisols are soils with clay-enriched subsoils that exhibit high base saturation and low-activity clays (WRB, 2006). The Haplic prefix qualifier indicates that the Lixisol has a typical expression of Lixisol features, with no other distinctive characteristics (WRB, 2006). The local soil series is Adawso developed over granite. Average soil pH at 0-10 cm at project initiation was 6.2 (Davies et al., 2014). Field observations indicated coarse-textured soils, with large size sand particles. The slope of the site was less than 4%. Apart from termites and ants there was little evidence of abundant insect communities. Also of note is that this site was previously under cultivation for many years prior to starting the trials.

2.3.1.2. Forest

The Forest experimental site was located in Amansie West District near the town of Ahwerewa. It is northwest of the Coastal Savannah site (Fig. 2). This plot is nestled in the interior of a lush cocoa plantation and natural wooded area, approximately 50 m off a main road. Mean annual rainfall in the Forest site is 1500 mm (Oppong-Anane, 2001). There are two growing seasons. The major season of 150-200 growing days lasts from March to July, and a minor season of about 90 days starts in September or October (Oppong-Anane, 2001; Obuobie et al., 2006).

The soils of this site are a Leptic Lixisols which are soils with an accumulation of clay with a high base saturation and low-activity in the subsoil layer (WRB, 2006).

Unlike the Coastal Savannah, the Forest site soils have a Leptic qualifier, indicating continuous rock starting within 100 cm of the soil surface (WRB, 2006). The local soil series is Amuni developed over phyllite. Average soil pH values prior to initiation of the trials were 6.3 at 0-10 cm depth (Davies et al., 2014).

Field observations of the Forest site include that insects were prolific, with evidence of beetles, centipedes, lady bugs, and ants which is usually a sign of good soil moisture and a healthy soil ecosystem. This site was historically tilled, but had been fallow 2-4 years prior to the start of this trial. This site had the greatest slope between 6-7%.

2.3.1.3. Transition

The Forest – Guinea Savannah Transition site, referred to as Transition, was located in the Ejura-Sekodumase District near the town of Ejura-Adiembra. It is northeast of the Forest experimental site (Fig. 2). This site occupies a summit position on the landscape. Adjacent to the plot are farmlands and a road buffered by about 20 m of grass vegetation. Mean annual rainfall is 1300 mm (Oppong-Anane, 2001). The transition site has a major growing season with 200-220 growing days and a minor season of 60 growing days (Oppong-Anane, 2001).

The transition site crops were planted into a Leptic Lixisol, the same soil as the Forest site. This site had the most acidic soils of all four agro-ecosystems with average soil pH at 0-10 cm at initiation of the study of 4.8 (Davies et al., 2014). Slope at this site was approximately 3%.

2.3.1.4. Guinea Savannah

The Guinea Savannah lies in northern Ghana, outside of the city of Tamale. This plot was located in the Tolon-Kumbungu District near town of Kumbungu-Kukuo (Fig. 2). Surrounding the Guinea Savannah experimental plots was farmland and a major road buffered by less than 5 m of exposed soil. Mean annual rainfall at this site is 1100 mm (Oppong-Anane, 2001). This was the only experimental site with one growing season which starts in April or May and ends in September or October, with 180-200 growing degree days. This difference in growing season is due to the difference in rainfall distribution throughout the year compared to the other three sites. The soils of this site were Pisoplinthic Plinthisols which are soils with an accumulation of iron under hydromorphic conditions (WRB, 2006). The Pisoplinthic prefix qualifier indicates a Pisoplinthic horizon starting within 100 cm of the soil surface (WRB, 2006). This plot was the most level with a slope of less than 2%. Average soil pH at 0-10 cm prior to initiation of the experiment was 5.3 (Davies et al., 2014).

2.3.2. Tillage and Cropping Treatments

Three methods of tillage and four types of cropping systems were laid out in a split-plot design with three replications for each treatment combination. Tillage was the main plot treatment and cropping system was the sub-plot treatment. The tillage types investigated were traditional till, zonal till, and no-till (Table 2). Traditional tillage was defined as hand hoeing to a depth of approximately 10 cm in the Coastal Savannah, Transition, and Guinea Savannah. In the Forest zone however, the traditionally practiced

form of seed bed preparation was slash and burn with no mechanical disturbance to the soil. Zonal tillage is defined as hand hoe only in the row to be planted in all agro-ecosystems whereas no till is the absence of any form of mechanical disturbance throughout the field, with the exception of the hole in which the seed is planted.

Table 2. Factor treatments in the Coastal Savannah, Forest, and Transition agro-ecosystems

Main Plot Factors	Sub-Plot Factors
No-Till (NT)	Maize (M)
Zonal Till (ZT)	Maize-cowpea (MC)
Traditional Till (TT)	Maize-mucuna (MM)
	Maize-Cowpea-mucuna (MCM)

The cropping systems were continuous maize (M), maize-cowpea rotation (MC), maize-mucuna rotation (MM), and a maize-cowpea-mucuna relay (MCM). These four cropping systems were tested in the 3 agro-ecosystems with two growing seasons: Coastal Savannah, Forest, and Transition zones. In the northern Guinea Savannah zone, due to the single growing season, the cropping systems were altered to include: M, MC, maize and cowpea intercropping (MCI), and cowpea-maize rotation (CM) (Table 3). For the Guinea Savannah, the difference between MC and CM crop rotations is that where the MC rotation starts with maize in the first trial year followed by cowpea in the next trial year, the CM crop rotation features cowpea, a nitrogen fixer, as the first crop of the 5 year trial, followed by maize the next year.

Table 3. Factor treatments in the Guinea Savannah agro-ecosystem

Main Plot Factors	Sub-Plot Factors
No-Till (NT)	Maize (M)
Zonal Till (ZT)	Maize-cowpea (MC)
Traditional Till (TT)	Maize-cowpea intercrop (MCI)
	Cowpea-Maize rotation (CM)

2.3.3. Plot Management

Field plots were initially cleared of vegetation by hand slashing and spraying with glyphosate herbicide. For tillage plots, tillage was performed on the main plots prior to fertilizer application. Traditional tillage involved hand hoeing to 10 cm depth over the entire main plot. Zonal tillage involved hoeing to a 10 cm depth only in the rows where the seeds would be placed. Fertilizer, in the form of 15-15-15 NPK (Kofi Boa, personal communication, March 14, 2014), was point placed by mixing fertilizer with soil in the planting hole. Two seeds were placed per hole and covered with soil. Seed spacing was 40 cm and row spacing was 80 cm. Fields were maintained by hand weeding or hand hoeing. Rain gauges were installed at all sites in the latter part of May 2011.

2.3.4. Soil Collection and Processing

Soil samples were collected in December 2013 from all sites. All plots were composite sampled to a 0-15 cm depth. Three cores were taken from the middle row of each plot using a 2-cm diameter auger and placed in cotton soil bags. Samples were laid out to air dry within 4-48 hours of collection. Prior to shipment from Ghana to the

NAWA lab at Texas A&M University, cotton bags were placed in sealed zip-lock plastic bags contained within sealed Tupperware containers.

On arrival at Texas A&M University the soils were logged into the NAWA laboratory soils database where they were given a unique ID number to track their analysis and chain of custody as required by BL2 protocols. Soils were further air-dried prior to gently breaking up any large soil pedes using a mortar and pestle before sieving through a 2mm sieve.

2.3.5. Soil Extractions

To quantify total extractable soil nitrogen (TEN) and extractable organic carbon (EOC), 3.5 g of soil were combined with 30 mL of 0.1 M HCl and shaken for two hours at 500 rpm on a rotary shaker (Davies et al., 2014). Samples were then centrifuged for 15 minutes at 19,974 g-force and filtered using a Whatman GF/F filter (nominal pore size 0.7 μm) to remove any floating organic material in the supernatant. Extracts were diluted with ultrapure water to ensure enough sample was available for chemical analysis. Extracts were analyzed between 1 and 24 hours after extraction.

To quantify extractable soil $\text{PO}_4\text{-P}$, 3 g of soil were combined with 21 mL Bray 1 solution and shaken for 1 minute on a rotary shaker (Bray, 1945). Samples were then centrifuged for 5 min at 2,809 g-force and filtered with Whatman GF/F filters (nominal pore size 0.7 μm) to remove any floating organic material. Extracts were analyzed between 1 and 24 hours after extraction.

2.3.6. Chemical Analyses

Extractable organic carbon and TEN were measured using high temperature Pt-catalyzed combustion with a Shimadzu TOC-VCSH and Shimadzu total measuring unit TNM-1 (Shimadzu Corp. Houston, TX, USA). EOC was measured as non-purgeable carbon, which entailed acidifying the sample (250 μ L 2 M HCl) and sparging for 4 min with C-free air. $\text{NH}_4\text{-N}$ was analyzed using the phenate hypochlorite method with Na-nitroprusside enhancement (USEPA method 350.1) and $\text{NO}_3\text{-N}$ quantified using Cd-Cu reduction (USEPA method 353.3). $\text{PO}_4\text{-P}$ was analyzed using the ascorbic acid, molybdenum blue method (APHA 1992). Colorimetric methods were performed using a Westco Scientific Smartchem Discrete Analyzer (Westco Scientific Instruments Inc. Brookfield, CT, USA). Sample replicates, blanks, NIST (National Institute of Standards and Technology) traceable and check standards were run every 12th sample to monitor instrument precision. Instrument lower detection concentrations were 0.1 mg L^{-1} for $\text{NO}_3\text{-N}$, $\text{NH}_4\text{-N}$ and $\text{PO}_4\text{-P}$, 0.5 mg L^{-1} for TEN and EOC and 0.7 mg L^{-1} for EON.

2.3.7. Statistical Analyses

Prior to performing statistical analyses, data were reviewed for outliers and samples were re-run as necessary. Data were evaluated for meeting the following key assumptions of analysis of variance (ANOVA):

1. There are k simple random samples from k populations.
2. The k samples are independent of each other; that is, the subjects in one group cannot be related in any way to subjects in a second group.

3. The populations are normally distributed.
4. The populations have the same variance; that is, each treatment group has population variance s^2 .

In order to determine if these assumptions of ANOVA were met, data were tested for normality using the Shapiro-Wilks test, and at p values > 0.05 the data was considered normally distributed. Normality tests revealed that the data was not normally distributed. Several transformations were performed, a logarithmic function was ultimately chosen because it was the most effective at achieving normal distribution for the majority of the data.

Univariate analysis of variance a general linear model (GLM) was conducted to determine if significant differences in concentrations of $\text{NO}_3\text{-N}$, $\text{NH}_4\text{-N}$, EON, TEN, EOC, and $\text{PO}_4\text{-P}$ across all agro-ecosystems of Ghana. The main factors were 1) agro-ecosystem, 2) tillage, and 3) cropping sequence. Interaction effects were 1) agro-ecosystem x tillage, 2) agro-ecosystem x cropping, 3) tillage x cropping sequence and 4) agro-ecosystem x tillage x cropping sequence.

Analysis of variance (ANOVA) was used to determine factor effects on the concentrations of $\text{NO}_3\text{-N}$, $\text{NH}_4\text{-N}$, EON, TEN, EOC, and $\text{PO}_4\text{-P}$ within each agro-ecosystem. The fixed factors were 1) tillage and 2) cropping sequence. Interaction effects were: 1) tillage x cropping sequence. Differences in individual treatment combinations were determined with Duncan's New Multiple Range Test ($\alpha < 0.05$). Significant effects of TSP or urea and compost and their interactions (univariate analysis

of variance) were determined at $p < 0.05$. Where an interaction of tillage and cropping occurred for any nutrient then a one-way analysis of variance was run with cropping as the independent variable and the nutrient as the dependent variable for each tillage treatment in turn.

2.4. RESULTS & DISCUSSION

Hypothesis 1 was not accepted. The hypothesis that low rainfall regions would have higher soil nutrients when a certain forms of tillage were practiced was not accepted. In the Guinea Savannah and Coastal Savannah, there was no significant difference in soil nutrient concentrations among plots treated with different tillage methods.

The second hypothesis was that in high rainfall regions such as the Forest agro-ecosystem, no-till would be associated with higher soil nutrients. This hypothesis was also not accepted. The Forest agro-ecosystem showed no significant differences in soil nutrient concentrations. Neither the tillage nor cropping system factor had any significant difference in extractable soil nutrient concentrations.

The final hypothesis was that the addition of a legume as part of a crop rotation will increase soil nitrogen because of the ability of legumes to biologically fix nitrogen. This hypothesis was rejected. Crop rotations that featured legumes in the rotation were not found to be significantly higher in the soil nutrients evaluated in this study. Although no hypotheses were accepted some treatment differences were found.

2.4.1. Results Across Agro-ecosystems

Data across all agro-ecosystems were aggregated and evaluated for factor effects from agro-ecosystem, tillage method, cropping system, and their interactions. Agro-ecosystem had a significant effect on the concentrations of all extractable soil nutrients evaluated, whereas, tillage, cropping system, and their interactions were only significant for some soil nutrients.

2.4.1.1. N-results: NO₃-N, NH₄-N and EON

Univariate analysis of variance revealed a significant effect of agro-ecosystem on extractable soil NO₃-N ($p < 0.0001$). There was also a significant interaction of agro-ecosystem and cropping ($p = 0.016$). The Guinea Savannah had the highest extractable soil NO₃-N concentrations followed by Coastal Savannah, Forest, and Transition agro-ecosystems, each having successively lower concentrations of extractable soil NO₃-N (Fig. 3).

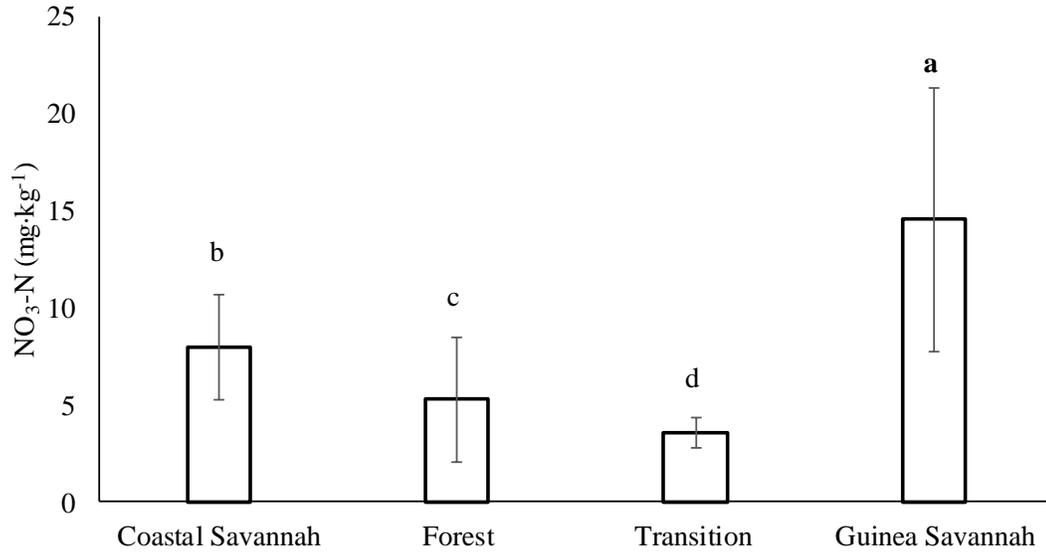


Fig. 3. Mean extractable soil NO₃-N concentrations. Error bars are standard deviation. Differences in lower case letters indicate a significant difference between agro-ecosystem types. Data shown is untransformed data but the Duncan's new multiple range test was performed on log transformed data.

Univariate analysis of variance revealed a significant effect of agro-ecosystem on extractable soil NH₄-N ($p < 0.001$). An interaction of agro-ecosystem x tillage x cropping had a significant effect on soil extractable NH₄-N ($p = 0.04$). The Coastal Savannah had the highest concentrations of extractable soil NH₄-N followed by the Forest. Soil NH₄-N concentration was not significantly different in the Transition and Guinea Savannah agro-ecosystems (Fig. 4).

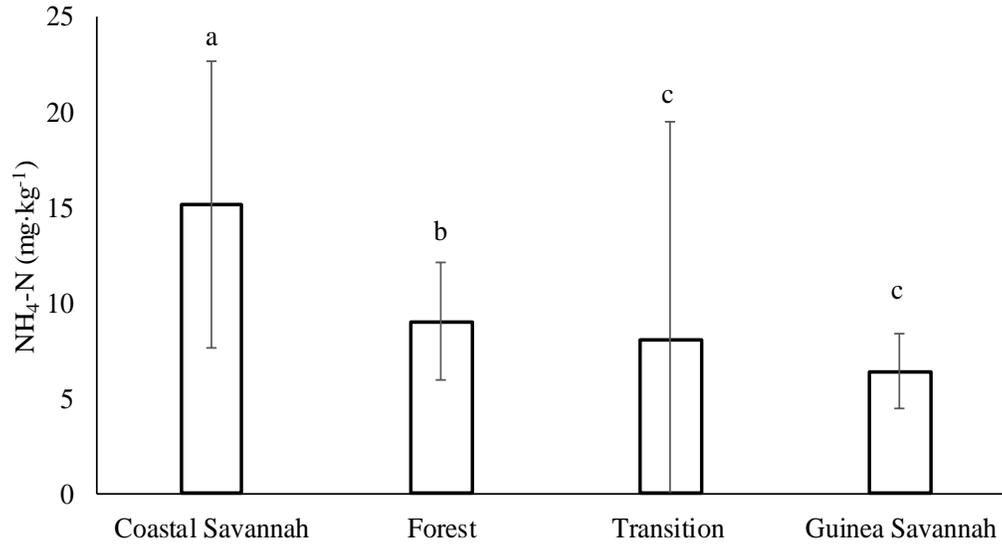


Fig. 4. Mean extractable soil NH₄-N concentrations. Error bars are standard deviation. Differences in lower case letters indicate a significant difference between agro-ecosystem types ($p < 0.05$). Data shown is untransformed data but the Duncan's new multiple range means separation test was performed on transformed data.

Univariate analysis of variance revealed that both agro-ecosystem ($p = 0.001$) and tillage ($p = 0.04$) had a significant effect on extractable organic nitrogen (EON). The Forest had significantly higher soil EON concentrations than the Coastal and Guinea Savannahs. Samples from the Transition zone had zero recoverable EON and so it was omitted from statistical analysis (Fig. 5).

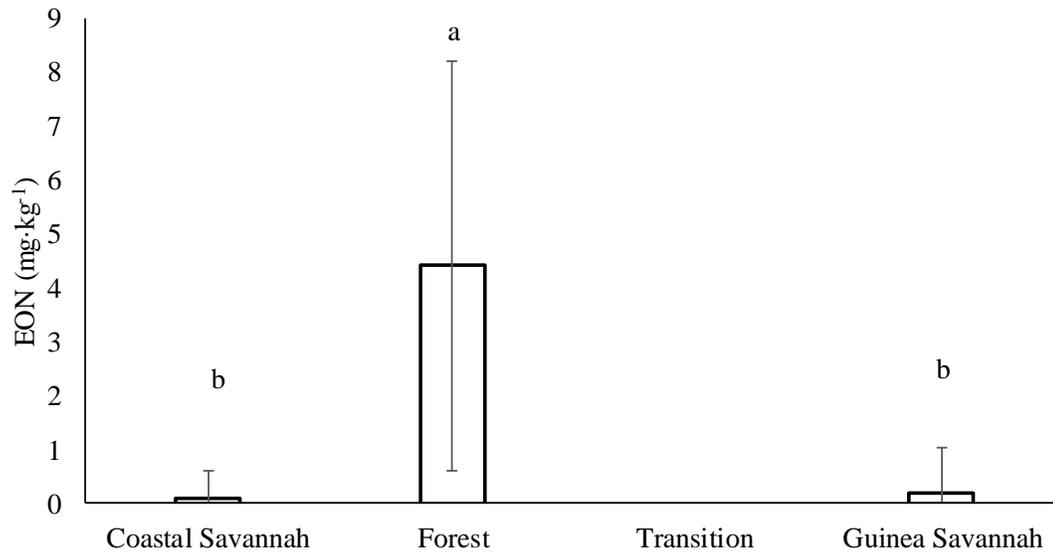


Fig. 5. Mean soil extractable organic nitrogen (EON) concentrations. Error bars are standard deviation. Differences in lower case letters indicate a significant difference between agro-ecosystem types ($p < 0.05$). Data shown is untransformed data but the Duncan's new multiple range means separation test was performed on transformed data. No measurable EON was extracted from the Transition agro-ecosystem soils.

Higher rainfall and enhanced biomass production in the Forest and Transition agro-ecosystems may have enhanced plant uptake of $\text{NO}_3\text{-N}$ resulting in lower concentrations at the two agro-ecosystems. Conversely, the Coastal and Guinea Savannahs, with lower rainfall and lower biomass production, had higher concentrations of both $\text{NO}_3\text{-N}$ and $\text{NH}_4\text{-N}$ than the other agro-ecosystems. Higher extractable N could be due to lower plant uptake of soil available-N. Also the lower biomass production in these regions may have contributed low enough amounts of OM whereby net immobilization did not occur. Also, microbial activity may have been lower in the

Coastal Savannah due to the sandy soil texture and uneven rainfall distribution that could have inadequately provided soil moisture to sustain high microbial activity. Either of these scenarios is supported by significantly higher extractable soil N in the Coastal Savannah.

Additionally, the timing of sampling in the Coastal Savannah was sampled in December after October's minor rainy season but before the start of the main rainy season in March. The Guinea Savannah was also sampled before the April start of their mono-modal rainfall period. In dry regions, crop biomass produced during the growing season accumulates until the rainy period at which point a microbial nitrate flush can quickly convert organic N to $\text{NO}_3\text{-N}$. This is likely the cause for the high $\text{NO}_3\text{-N}$ in these regions, despite the soils having the low water holding capacity, low cation exchange, and low organic matter typical of Alfisols (Cogle, 1997) which would otherwise make high concentrations of $\text{NO}_3\text{-N}$ an unexpected observation.

EON represents the organic-N pool associated with soil biomass and soil organic matter that is easily solubilized (Ros et al., 2009) and responds similarly to wetting and drying as does DOC (Xiang et al., 2008). The Forest zone likely accumulated the highest EON concentrations due to its higher biomass production, greater organic matter inputs, and loamy soil texture, which provided an organic-N source, stimulated microbial activity, and allowed for greater adsorption of EON than other agro-ecosystems.

DON meta-analysis by Ros et al. (2009) found that EON concentrations were lower when $\text{pH} > 6$, lower in sandy soils, higher during spring/summer seasons, and

higher under grassland compared to arable land. The findings for the current study are somewhat consistent. The Forest agro-ecosystem likely accumulated the highest EON concentrations because of its finer soil texture, which allowed for greater adsorption of EON to soil minerals and which protected it from degradation compared to the sandy soils of the other agro-ecosystems. High biomass production in the Forest agro-ecosystem would also provide an organic N source and stimulate microbial activity. That the Forest had significantly higher EON is generally consistent with most of Ros et al., (2009) meta-analysis findings. Because EON is closely linked to organic matter and microbes, it follows that the agro-ecosystem with the highest annual rainfall, greatest biomass production, and loamiest texture likely has the most active microbial community and provides for the greatest accumulation of organically bound EON.

2.4.1.2. Comparison of N species in 2012 vs 2013 trial years

Agro-ecosystem had a significant effect on 0.1 M HCl extractable soil nitrogen species and this was illustrated with univariate analysis of variance with means separation tests for the 2013 trial year. One surprise in this study was the extremely low residual EON in soils collected, processed and analyzed in 2013. This prompted an examination of the 2012 data (Davies, 2014) and comparisons made. In the Coastal Savannah agro-ecosystem, while there was no significant difference in soil TEN between the two years, the distribution of N species was altered significantly (Table 4). NO₃-N and EON were significantly higher in 2012, yet NH₄-N was significantly lower in 2012 compared to 2013 (Table 4). The change in percentage of NO₃-N, NH₄-N and

EON in the Coastal Savannah agro-ecosystem showed that in 2013 $\text{NO}_3\text{-N}$ was 12% lower, $\text{NH}_4\text{-N}$ was 40% higher and EON was 28% lower relative to soils collected and processed in 2012 (Fig. 6).

This distribution change may be due to the tillage and cropping treatments within each agro-ecosystem, weather during 2013 growing season, or a change in standard operating procedure for the extraction and analysis of N species. Ecological function dictates that the source of EON is generally in the form of amino acids, proteins and amines, a continuum of molecule sizes. A proportion of this EON is biodegradable (Gregorich et al., 2003) which means quite simply that the EON is mineralized and disappears from solution. The mechanism of EON biodegradation has not been fully researched but it is highly likely that biodegradation, rather than the complete consumption of the EON molecule, is due to extracellular enzymes. Extracellular enzymes most often observed in soils, include, aminases that are responsible for cleaving the amino functional group from simple single chain amino acids and protease that breaks down proteins. This latter group of extracellular enzymes is substrate induced, as protein concentrations in the soil increased the activity of protease also increases (Geisseler and Horwath, 2008).

The N distribution in the Coastal Savannah could be due to the historic land use of savannah grass and the subsequent land use of cropping that resulted in EON being in the form of amino acids that are readily mineralized to NH_4^+ and DOC. The different

NO₃-N concentrations between the two years may be due to increased plant uptake in 2012.

Table 4. Comparison of average nitrogen species concentrations for 2012 and 2013 soil collections. Data are pooled (includes tillage and cropping treatments) for ease of comparison.

Agro-ecosystem	Year	NO ₃ -N	NH ₄ -N	EON	TEN
		mg kg ⁻¹			
Coastal Savannah	2012	11.6 ^a	6.4 ^a	7.3 ^a	25.3 ^a
	2013	8.0 ^b	15.2 ^b	0.1 ^b	23.3 ^a
Forest	2012	11.3 ^a	10.3 ^a	19 ^a	40.6 ^a
	2013	5.3 ^b	9.0 ^a	4.4 ^b	18.7 ^b
Transition	2012	3.3 ^a	6.3 ^a	10.3 ^a	19.9 ^a
	2013	3.6 ^a	8 ^a	0.0 ^b	11.6 ^b
Guinea Savannah	2012	16.8 ^a	6.8 ^a	5.2 ^a	28.8 ^a
	2013	14.6 ^a	6.4 ^a	0.2 ^b	21.2 ^b

2012 Soil Analysis (Year 2 of 5 in the trial). Source: Davies (2014)

In the Forest agro-ecosystem a loss of TEN was evident between 2012 and 2013 (Table 4). Significantly higher TEN was extracted from soils processed in 2012 compared to 2013 suggesting a loss of total N from the Forest agro-ecosystem between collections of soil in 2012 and 2013. Loss of total N at the magnitude observed might suggest either enhanced plant uptake or denitrification but often examination of the distribution of N each year can help solve the puzzle (Fig. 6). Much of the decrease in

residual N when comparing the two years was for $\text{NO}_3\text{-N}$ and EON, which were both significantly lower in 2013 (Table 4). The amount of soil $\text{NH}_4\text{-N}$ in 2012 and 2013 were not significantly different between the two years in the Forest agro-ecosystem (Table 4). Enhanced $\text{NO}_3\text{-N}$ uptake by plants or enhanced denitrification in 2013 relative to 2012 might explain the differences. As for the ~75% decrease in EON between 2012 and 2013 (Table 4), that may be a result of microbial hydrolysis of organic N to inorganic N.

Current theory suggests that DON in soil solution is comprised of extracellular enzymes and active bacteria which should be possible considering the pore size of filters (0.7 μm) compared to the diameter of bacteria (~ 0.2 μm). If the soil microbial community remained in a maintenance rather than growth phase in these soils then expectations are that an increase in EON would not be observed following the initial mineralization of EON and subsequent immobilization of $\text{NH}_4\text{-N}$. Examination of the percent distribution of N species within TEN for the two years revealed that the proportion of $\text{NO}_3\text{-N}$ within TEN had remained constant between 2012 and 2013 collections (Fig. 6). The change in distribution of $\text{NH}_4\text{-N}$ and EON between 2012 and 2013 helps to elucidate potential mechanisms where a 23% decrease in EON and 23% increase in $\text{NH}_4\text{-N}$ proportions to TEN between years 2 & 3 of the trial suggests mineralization of EON but no microbial uptake of the $\text{NH}_4\text{-N}$ (Fig. 6).

In the Transition agro-ecosystem concentrations of both TEN and EON were significantly lower in 2013 when compared to the 2012 soils but soil $\text{NO}_3\text{-N}$ and $\text{NH}_4\text{-N}$ concentrations between the two years were not significantly different (Table 6). The

proportion of the individual N species within TEN did show some changes (Fig. 6). No EON was detected in the Transition zone soil in 2013 so the proportion of EON in TEN was 0%, much lower than the 52% EON within TEN observed in 2012 (Fig. 6). The EON lost appeared to have been mineralized or mineralized and nitrified in 2013 contributing 37% to $\text{NH}_4\text{-N}$ and 15% to $\text{NO}_3\text{-N}$ thus increasing their proportions within TEN in 2013 compared to 2012 (Fig. 6).

In the Guinea Savannah there were no significant differences in the concentrations of soil $\text{NO}_3\text{-N}$ and $\text{NH}_4\text{-N}$ when comparing 2012 and 2013 samples (Table 4). There were, however, significant differences in the concentrations of TEN and EON between the two years (Table 4). Concentrations of EON in 2013 was a negligible 0.2 mg kg^{-1} compared to 5.2 mg kg^{-1} in 2012 (Table 4). Examination of the proportion of EON in TEN revealed that 17% of EON was lost between the 2012 and 2013 samples and the proportion of $\text{NO}_3\text{-N}$ increased by 6% and $\text{NH}_4\text{-N}$ by 11% between the two years (Fig. 6).

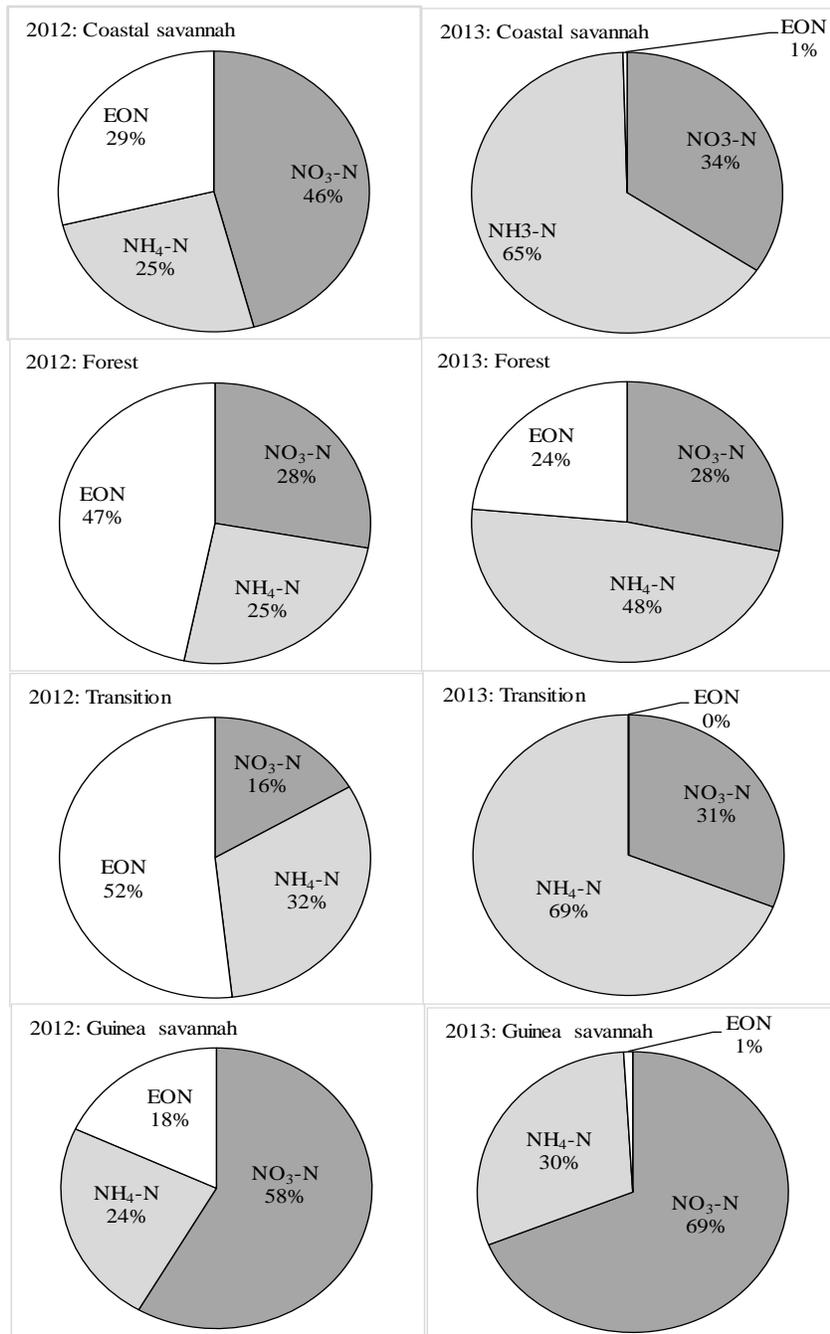


Fig. 6. Soil N species distribution. 0.1 M HCL extracted soil nitrogen species distribution in the 4 agro-ecosystems used in this study for soils collected in 2012 and 2013 in the tillage and cropping study.

In a tillage and cropping study in south-central Texas, USA comprising no tillage vs traditional tillage and a monoculture vs rotational cropping, the proportion of N species within TEN extracted with cold water extracts ranged from 21-34% EON, 8-18% NH₄-N and 55-68% NO₃-N (Carrillo-Gonzalez et al., 2013). Proportionally, NO₃-N represented a lower percent of TEN in all the Ghanaian agro-ecosystems except the Guinea Savannah. NH₄-N represented a similar but slightly higher percent of TEN for the 2012 soils compared to the Carrillo-Gonzalez et al. (2013) study. The percent of TEN as NH₄-N for the 2013 soils were 2-3X higher when compared to the Carrillo-Gonzalez et al. (2013) study. The proportion of EON in TEN in agricultural soils is generally on the low side but the soils collected in 2012 had comparable %EON to the Carrillo-Gonzalez (2013) study while the soils collected in 2013 did not. The Carrillo-Gonzalez et al. (2013) study had been ongoing for almost 30 years when samples were retrieved and the distribution of N species (EON and NH₄-N) within TEN for the 2012 samples were similar to those reported by Carrillo-Gonzalez et al. (2013). The NO₃-N proportion of TEN were much lower in the Ghanaian study for both 2012 and 2013 soils which might suggest that in a tillage and cropping study that has been on-going for 30 years NO₃-N will accumulate.

2.4.1.3. C-results: EOC

Univariate analysis of variance revealed a significant effect of agro-ecosystem on EOC concentrations ($p < 0.001$). There was no tillage, cropping, or interaction effects on EOC concentrations across all four climatic zones. The Forest agro-ecosystem had the highest

EOC concentrations followed by the Guinea Savannah. Both the Coastal Savannah and Transition had similarly lower concentrations of EOC (Fig 7).

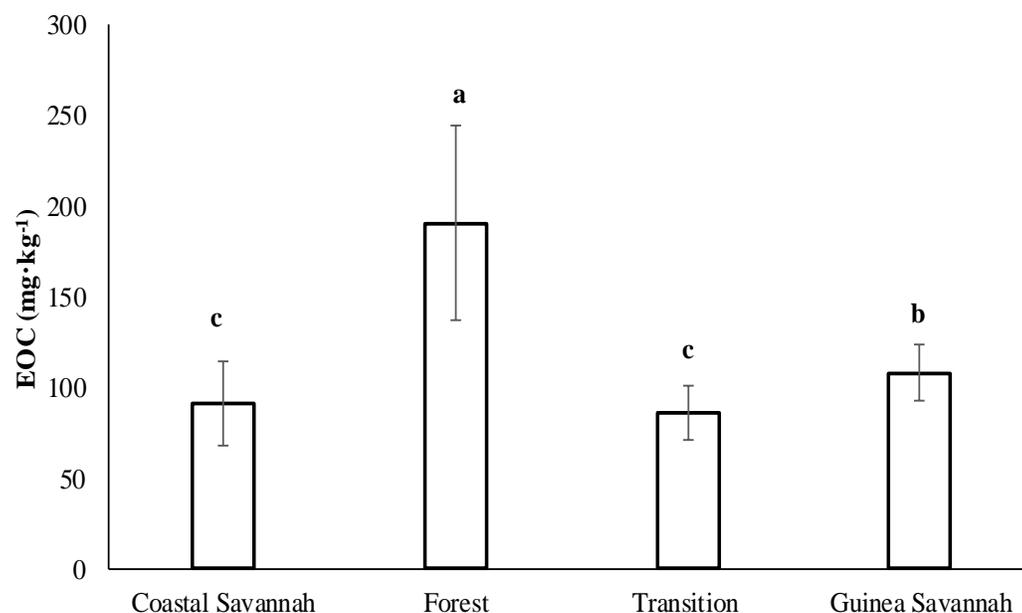


Fig. 7. Mean soil extractable organic carbon (EOC) concentrations. Error bars are standard deviation. Differences in lower case letters indicate a significant difference between agro-ecosystem types. Data shown is untransformed data but the Duncan's new multiple range test was performed on transformed data.

The use of EOC is a method introduced here for quantifying the dissolved organic carbon (DOC) that can be readily extracted with a weak acid (0.1M HCl). Dissolved organic matter (DOM) is defined as the soluble organic matter that can pass through a 0.45 μm filter (Thurman, 1985). DOM contains organic C, organic N, organic

P and organic S (Chapter 4). Its production and decline depends on biotic factors such as plant litter, root exudate releases, and microbial biomass (Kalbitz et al., 2000). These biological activities are heavily dependent on adequate moisture (Marschner & Kalbitz, 2003). Therefore, it is no surprise that the Forest zone would have the greatest EOC concentrations of all the agro-ecosystems evaluated because of the favorable climatic factors such as high annual rainfall, even rainfall distribution, high plant biomass production, and warm temperatures that support an active microbial community. The second highest EOC concentration in the Guinea Savannah may likely be attributed to the lack of decomposition of the DOM that accumulated in this region and the finer soil texture that may have served to protect DOM from microbial processes.

2.4.1.4. P-results: Orthophosphate

Univariate analysis of variance revealed that agro-ecosystem ($p < 0.001$) and tillage ($p = 0.03$) had a significant effect on Bray 1 extractable soil $\text{PO}_4\text{-P}$ concentrations. In addition, there was a tillage x cropping system effect ($p = 0.009$). The Coastal Savannah and Transition zone had significantly higher $\text{PO}_4\text{-P}$ than the Forest and Guinea Savannah (Fig. 8).

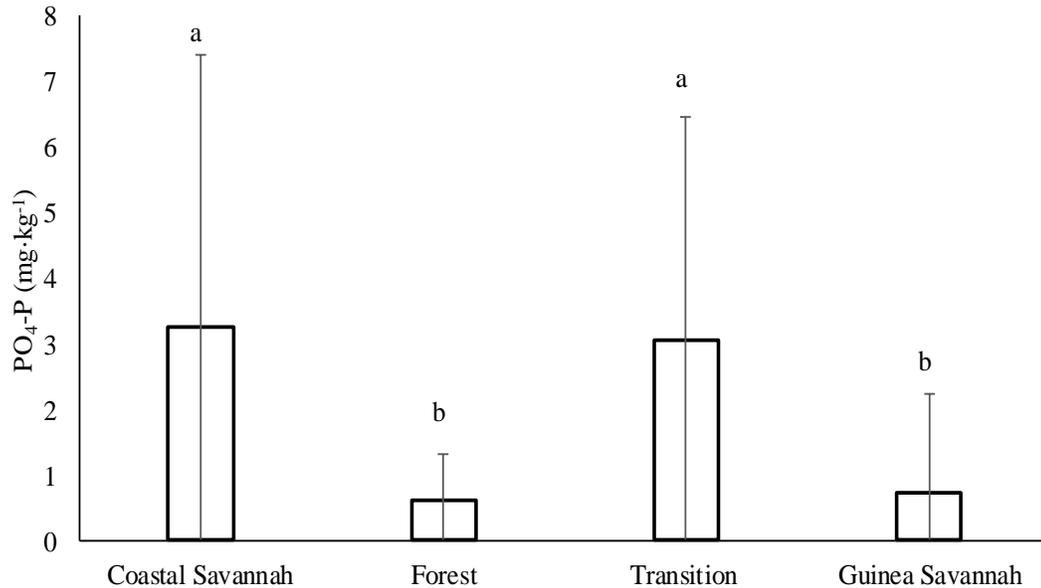


Fig. 8. Mean extractable soil $\text{PO}_4\text{-P}$ concentrations. Error bars are standard deviation. Differences in lower case letters indicate a significant difference between agro-ecosystem types. Data shown is untransformed data but the Duncan's new multiple range test was performed on transformed data.

Soils in the Forest and Guinea Savannah are silty loams with a pH of 6.27 and 5.34 respectively, and have low concentrations of $\text{PO}_4\text{-P}$ compared to the sandy soils in the other agro-ecosystems. The monovalent H_2PO_4^- form of phosphorus, which was measured in this study, is the most soluble form of P in near neutral to slightly acidic soils and most absorbed by plants (Troeh and Thompson, 2005). The portion of soil P adsorbed to minerals is considered labile and easily extractable in the lab or by plants (Foth and Ellis, 1997). Soils with clays and silts tend to have greater water holding capacity than sandier textured soils and are, therefore, more likely to contain H_2PO_4^- in

solution. It is likely that the Forest, with both had both ample annual rainfall and finer soil textures, exhibited lower concentrations of residual $\text{PO}_4\text{-P}$ due to higher plant uptake of solubilized H_2PO_4^- . Similarly in the Guinea Savannah the fine soil texture may have also had stronger P adsorption that may have contributed to labile P stocks accessed by crops. Meanwhile, findings demonstrated that the agro-ecosystems with coarse textured soils (Coastal Savannah and Transition) had greater residual H_2PO_4^- than agro-ecosystems with finer soil textures and poorer soil water retention. P movement to plant root zones depends primarily on diffusion (Foth and Ellis, 1997). Because soluble P can usually only be accessed by plants when soil solution is in close proximity to roots (Foth and Ellis, 1997) the poor water holding capacity in these sandier soils may have limited plant uptake of solubilized H_2PO_4^- in the Coastal Savannah and Transition resulting in greater residual H_2PO_4^- in these coarse textured soils.

Furthermore, P is generally considered to be limiting in tropical agricultural soils and this may be due to the amount of positively charged iron, zinc and manganese hydroxides found in highly weathered tropical soils in W. Africa (Verbree et al., 2014). $\text{PO}_4\text{-P}$ adsorbs strongly to soil minerals (Nodvin et al., 1986) which will affect its availability to plants for uptake.

2.4.1.5 Summary of agro-ecosystem effects on extractable soil nutrients

The Guinea and Coastal Savannahs had the highest concentrations of inorganic N (Table 5). The Forest soils had the highest concentrations of extractable organic C and N. Meanwhile the Coastal Savannah and Transition agro-ecosystems were highest in PO₄-P concentrations.

Agro-ecosystems with a prior land use that provided high carbon inputs such as leaf litter and root exudates in the Forest or Transition zone would have lower extractable soil NO₃-N because the carbon available in the soil could not be utilized as a substrate by the soil microbial community without NH₄-N which would leave little NH₄-N to be nitrified for plant uptake. Additionally, in agro-ecosystems with higher organic matter contributions NO₃-N may have been immobilized during the wet growing seasons until the next rewetting in the following rainy season where a rapid mineralization would have been possible. These scenarios support the significantly lower observed extractable soil NO₃-N in the Forest and Transition agro-ecosystems in this study.

Based on visual comparison with baseline year data there did not seem to be much change in nutrient status of each agro-ecosystem between the baseline year in 2011 before trials were established and the third trial year (Table 6).

Table 5. Summary of agro-ecosystems with the highest soil nutrients in trial year 3. Shaded cells indicate the agro-ecosystem that had the greatest soil nutrient compared to the other zones.

	Coastal Savannah	Forest	Transition	Guinea Savannah
NO ₃ -N				
NH ₄ -N				
EON				
EOC				
PO ₄ -P				

Table 6. Baseline soil nutrient data

	Coastal Savannah	Forest	Transition	Guinea Savannah
Depth (cm)	0-10	0-10	0-10	0-10
pH	6.25	6.27	4.80	5.34
Org. C %	0.36	2.45	0.35	0.58
OM %	0.62	4.22	0.60	1.00
N %	0.04	0.21	0.03	0.06
P (ppm)	18.2	3.0	14.0	6.3
K (ppm)	70.3	70.3	160.7	67.0
Texture	Loamy sand	Silty loam	Loamy sand	Silty loam

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Understanding soil nutrient differences between the agro-ecosystems provides fundamental information as to limitations of each climatic zone and may help in predicting which treatments will have the best impact on the soil nutrient status in each region. This is important for the local subsistence farmer that needs empirical information on what crop management methods to adopt in his region. To this aim, the next phase of this investigation focuses on identifying those within-zone treatment effects on soil nutrients.

2.4.2. Coastal Savannah Treatment Effects

Twelve combinations of tillage and crop rotation were used in this analysis. Not every tillage and cropping combination had a significant effect on 0.1 M HCl extractable nutrients and so data is presented for each agro-ecosystem nutrient where a significant effect of tillage, cropping or their interactions were revealed by univariate analysis of variance and further by means separation testing. Two of the extractable soil nutrients, $\text{NO}_3\text{-N}$ and $\text{NH}_4\text{-N}$, demonstrated significantly different concentrations among treatments. No other soil nutrients evaluated had significant findings.

2.4.2.1. Coastal Savannah: $\text{NO}_3\text{-N}$

Soil extractable $\text{NO}_3\text{-N}$ concentrations had normal distribution after log transformation in the Coastal Savannah based on a Shapiro-Wilks test ($p = 0.34$) but the treatment groups did not have equal population variance, and ANOVA assumptions were only partially met. Univariate analysis of variance revealed that the type of crop management system had a significant effect on soil extractable $\text{NO}_3\text{-N}$ concentrations (p

= 0.006). No significant tillage or tillage x cropping interaction effects were observed for NO₃-N in the Coastal Savannah (Fig. 9).

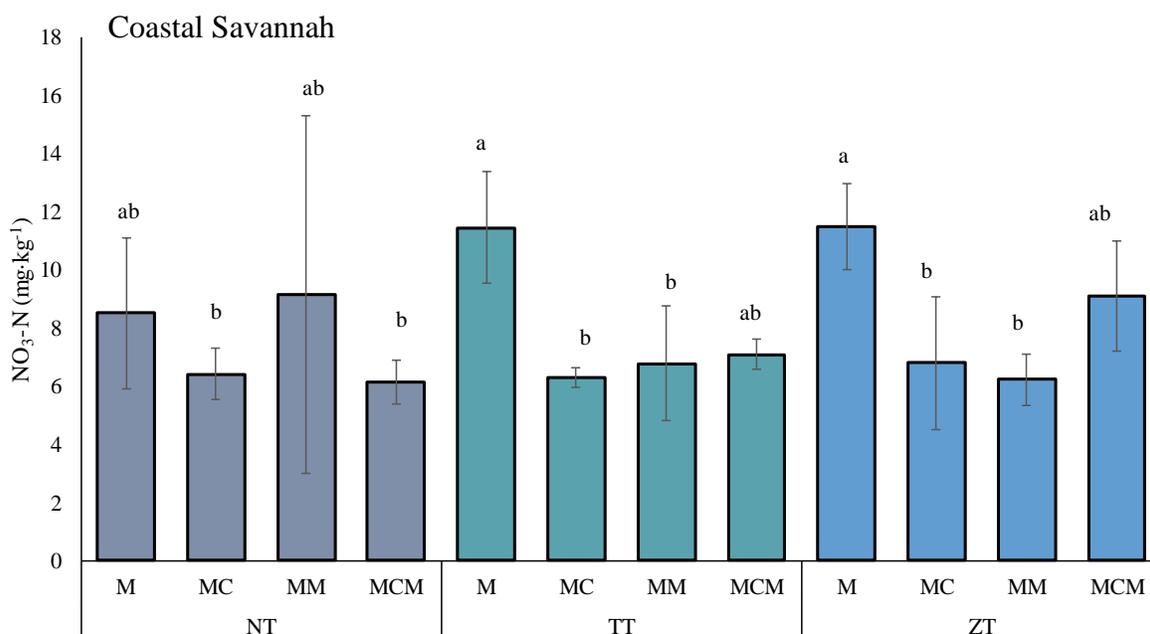


Fig. 9. Mean extractable soil NO₃-N for each treatment in the Coastal Savannah. The main plot factors are tillage methods abbreviated as follows: no-till (NT), traditional tillage (TT), and zonal till (ZT). The sub-plot factors are cropping systems, abbreviated as follows: continuous maize (M), maize-cowpea (MC), maize-mucuna (MM), and maize-cowpea-mucuna relay (MCM). Error bars are standard deviation. Differences in lower case letters indicate a significant difference between treatments. Data shown is untransformed data but the Duncan's new multiple range test was performed on transformed data ($\alpha < 0.05$).

Tillage and cropping treatment combinations were compared using the Duncan's new multiple range test. Two treatments (TT-M and ZT-M) had significantly higher soil extractable NO₃-N concentrations than 6 other treatment combinations. Both treatments with high soil extractable NO₃-N featured a maize-only cropping system, while none of

the low extractable NO₃-N treatments included a maize-only cropping system (Fig. 9; Table 7).

Table 7. Fertilizer treatment TT-M and ZT-M had higher soil NO₃-N concentrations than the following 6 other fertilizer treatments.

		NT-MC
		NT-MCM
TT-M	>	TT-MC
ZT-M		TT-MM
		ZT-MC
		ZT-MM

In addition, 2 of the 3 treatments with a maize-only crop rotation were high in soil extractable NO₃-N. Therefore there seems to be an association of maize monocrop with high NO₃-N in the Coastal Savannah. These means comparison results complement the ANOVA findings that cropping system had a significant effect on NO₃-N concentrations in the Coastal Savannah. Tillage was not found to be significant according to ANOVA. In addition visual comparison of treatment bars do not show any significant differences between the 3 tillage methods (Fig. 9).

The fact that the maize-only cropping system had significantly higher soil extractable NO₃-N concentrations compared to the maize-cowpea rotations was surprising considering the ability of legumes to fix N. Other studies have reported that maize cropping systems featuring a legume rotation had greater mineral N than

continuous maize cropping systems (Rao and Mathuva, 2000; Riedell et al., 2009). However another study in the Transition region of Ghana compared a maize-only cropping system with various legume and herbaceous crop rotations and found similar results to this study (Adjei-Nsiah et al., 2007). After year 1 and 2 cropping seasons there was no significant effect on soil pH, organic carbon, or total N by cropping system, despite there being differences in yield. This may be due to mineralization-immobilization patterns. Residues from mucuna and cowpea rotations could have caused net immobilization of N, as was the case in an East African crop residue study. In a leaching tube investigation that compared crop residue degradation in Malawan soils, Sakala et al. (2000) found that the residue combination of maize plus pigeonpea led to net immobilization. A similar interaction between maize stover and cowpea or mucuna could have resulted in the lower $\text{NO}_3\text{-N}$ observed from these treatments in this study.

The lack of tillage factor effects may be due to the minimal differences in soil disturbance between the tillage methods evaluated due to experimental design and sampling strategy. For example, the traditional tillage and zonal till both featured hand hoeing to a similar depth. The only difference being that traditional till featured hoeing throughout the plot, while the zonal till involved tilling only in the row to be planted. However, because soil sampling also took place only in the crop row there were essentially no differences in the samples from zonal till plots and traditional till plots. Therefore it is not surprising that Univariate analysis of variance or Duncan's means separation test found no significant differences between tillage methods.

Soils collected in 2012 from the same plots showed no significant effect of tillage, cropping or their interactions on 0.1M HCl extractable NO₃-N concentrations which were just two years after treatments commenced (Davies et al., 2014). NO₃-N concentrations in the Davies et al. (2014) study ranged from 9.9 to 19.8 mg kg⁻¹ in the TT-MM and NT-M respectively, slightly higher than observed in the current study.

2.4.2.2. Coastal Savannah: NH₄-N

Univariate analysis of variance revealed a significant interaction of cropping and tillage on extractable NH₄-N concentrations in the Coastal Savannah ($p = 0.04$). The significant interaction occurred for the traditional tillage only ($p < 0.05$). Transformed NH₄-N data had normal distribution ($p = 0.41$) and equal population variance, therefore assumptions for ANOVA were met. Means comparison revealed that only 1 treatment had significantly higher NH₄-N than others.

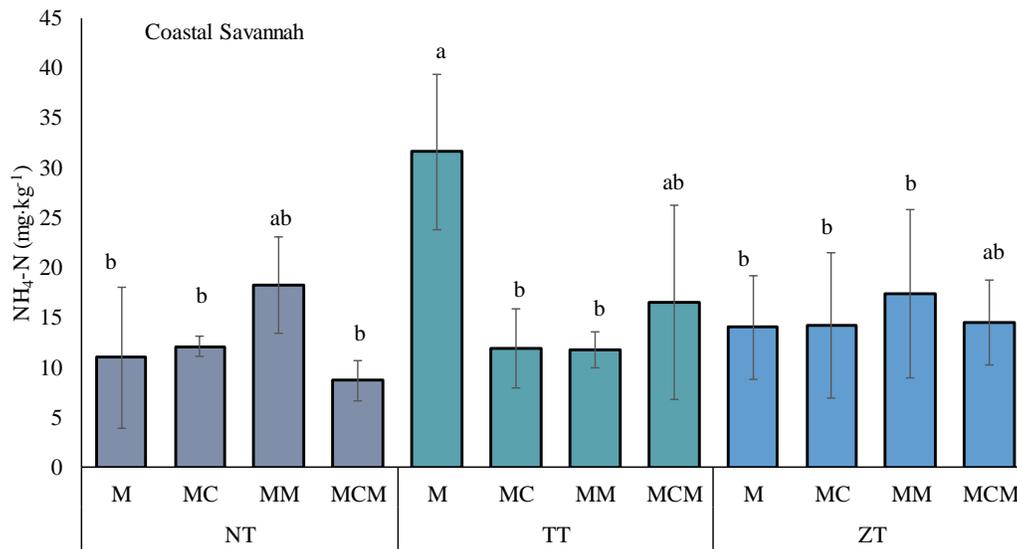


Fig. 10. Mean extractable soil NH₄-N for each treatment in the Coastal Savannah. The main plot factors are tillage methods abbreviated as follows: no-till (NT), traditional tillage (TT), and zonal till (ZT). The sub-plot factors are cropping systems, abbreviated as follows: continuous maize (M), maize-cowpea (MC), maize-mucuna (MM), and maize-cowpea-mucuna relay (MCM). Error bars are standard deviation. Differences in lower case letters indicate a significant difference between treatments. Data shown is untransformed data but the Duncan's new multiple range test was performed on transformed data ($\alpha < 0.05$)

Only one treatment was significantly different. The traditional tillage, via hand hoe, in a maize-only cropping system (TT-M) had significantly higher NH₄-N than 8 of the other treatments (Fig. 10). The TT-M treatment was similar in NH₄-N concentrations to 3 other treatments (NT-MM, TT-MCM and ZT-MCM) (Fig. 10). There did not seem to be a clear correlation between management differences and soil nutrients between TT-M and the low NH₄-N treatments. For example, of the 4 treatments featuring a traditional tillage crop rotation TT-M was significantly higher than 2 of them. This

indicated that traditional tillage was not consistently higher in $\text{NH}_4\text{-N}$ concentrations. In terms of cropping systems, of the 3 treatments featuring a maize-only cropping system the TT-M treatment was significantly higher than the other 2. It seems that the TT-M was an isolated treatment with higher $\text{NH}_4\text{-N}$. No treatments sharing either its tillage method nor cropping systems were also significantly higher in $\text{NH}_4\text{-N}$ concentrations. The lack of more significant differences between treatments suggests that despite the interaction effects suggested by ANOVA, Duncan's means comparison testing did not demonstrate a consistent correlation between either cropping system or tillage method and $\text{NH}_4\text{-N}$ concentration. This could be due to delayed soil response to management changes as this is only Year 3 of the field trials. It is possible that correlations become more pronounced in later years of the investigation.

Concentrations of $\text{NH}_4\text{-N}$ at the same site for soils collected in 2012 ranged from 5.6 to 7.2 mg $\text{NH}_4\text{-N}$ kg^{-1} soil and no effect of tillage or cropping was reported (Davies et al., 2014). Soils from a tillage and cropping experiment in Texas, USA extracted with DDW ranged from 3.6 to 3.8 mg $\text{NH}_4\text{-N}$ kg^{-1} soil and had a significant effect of cropping and interaction of cropping system x tillage was reported after a 30 year treatment (Carrillo-Gonzalez et al., 2013). Concentrations of $\text{NH}_4\text{-N}$ in this study were somewhat higher than reported for other studies and ranged from 8.7 to 31.6 mg kg^{-1} from the NT-MCM and TT-M respectively (Fig. 10).

2.4.3. Forest Treatment Effects

In the Forest agro-ecosystem none of the extractable soil nutrients measured were significantly different among treatments. Univariate analysis of variance revealed no significant effects of tillage, cropping system or their interaction on $\text{NO}_3\text{-N}$, $\text{NH}_4\text{-N}$, EON, EOC, or $\text{PO}_4\text{-P}$. Furthermore, numerous transformations failed to achieve normal distribution for the data within the forest agro-ecosystem. Davies et al. (2014) reported significant effects of tillage and cropping on DON and DOC but not $\text{NO}_3\text{-N}$ and $\text{NH}_4\text{-N}$ for soils collected at the same sites in 2012.

2.4.4. Transition Treatment Effects

In the Transition zone two extractable soil nutrients were significantly affected by treatments: $\text{NO}_3\text{-N}$ and $\text{PO}_4\text{-P}$.

2.4.4.1. Transition: $\text{NO}_3\text{-N}$

$\text{NO}_3\text{-N}$ did not pass the Shapiro-Wilks test of normality ($p = 0.000$). However, from visual inspection of the Quantile-Quantile (Q-Q) plots the data looked reasonably normal in distribution. In addition to lacking normality, the treatments also did not have equal variance for $\text{NO}_3\text{-N}$ concentration. Because the assumptions for ANOVA were not met, conclusions are drawn with caution.

Univariate analysis of variance revealed that the cropping system factor had a significant effect on extractable soil $\text{NO}_3\text{-N}$ ($p = 0.03$). Means comparison with Duncan's revealed that the NT-MM and NT-MCM treatments had higher $\text{NO}_3\text{-N}$ concentrations than 5 of the other treatment types (Fig. 11).

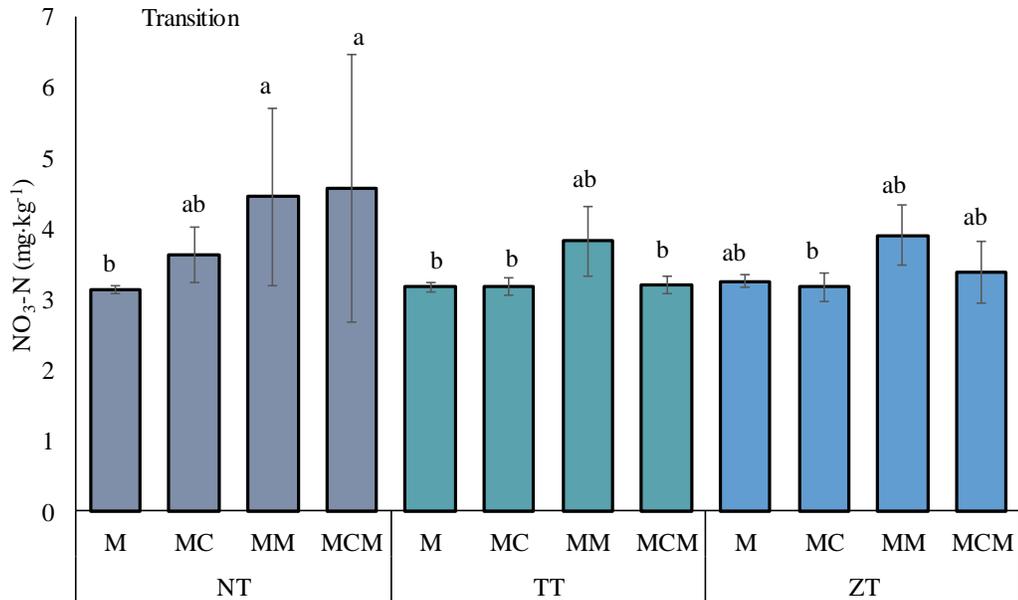


Fig. 11. Mean extractable soil NO₃-N for each treatment in the Transition. The main plot factors are tillage methods abbreviated as follows: no-till (NT), traditional tillage (TT), and zonal till (ZT). The sub-plot factors are cropping systems, abbreviated as follows: continuous maize (M), maize-cowpea (MC), maize-mucuna (MM), and maize-cowpea-mucuna relay (MCM). Error bars are standard deviation. Differences in lower case letters indicate a significant difference between treatments. Data shown is untransformed data but the Duncan's new multiple range test was performed on transformed data ($\alpha < 0.05$).

The two treatments with high NO₃-N concentrations both featured a mucuna crop in the rotation and were both no-till. Of the 5 significantly low extractable NO₃-N treatments 4 featured no mucuna crop coupled with some form of soil disturbance (Table 8). Comparison of treatment means seems to suggest that inclusion of mucuna and also lack of soil disturbance was associated with greater extractable NO₃-N concentrations in

this region. In addition, based on visual evaluation of treatment means graphs, the no-till treatments appear to have greater NO₃-N concentrations than treatments of traditional till or zonal till. Means comparison seemed to suggest that tillage type also influenced NO₃-N concentrations, which supports the ANOVA finding that the cropping system factor had a significant effect NO₃-N.

Table 8. Fertilizer treatments NT-MM and NT-MCM had higher soil NO₃-N concentrations than the following 5 other fertilizer treatments.

		NT-M
		TT-M
NT-MM	>	TT-MC
NT-MCM		TT-MCM
		ZT-MC

In 2012, Transition zone soils extracted with 0.1 M HCl had NO₃-N concentrations ranging from 2.6 to 3.9 mg kg in the TT-MC and ZT-M and TT-MM treatments respectively (Davies et al., 2014). These authors also reported a significant effect of cropping within the TT and ZT tillage treatments (Davies et al., 2014). Overall concentrations of NO₃-N in this study were slightly higher than concentrations of NO₃-N in the 2012 soils in the Transition agro-ecosystem. Both this study and the study of Davies et al. (2014) had an order of magnitude lower NO₃-N when compared to NO₃-N extracted with water in 30 year tillage and cropping study in Texas USA (Carrillo-

Gonzalez et al., 2013). This illustrates the high accumulation that can occur with a long term study when the effects of tillage, cropping and their interactions can be readily observed.

2.4.4.2. Transition: $PO_4\text{-P}$

ANOVA assumptions were not fully met for $PO_4\text{-P}$ data in the Transitional zone. The data was non-normal ($p = 0.009$) and the treatments did not have equal variance. Univariate analysis of variance revealed that tillage ($p = 0.02$), cropping system ($p = 0.02$), and their interaction ($p = 0.04$) had a significant effect on extractable soil $PO_4\text{-P}$ concentrations. Only the no tillage plots had a significant interaction with crop type (ANOVA; $p < 0.05$). Means comparison revealed several treatment differences.

First, the NT-MC treatment had significantly higher $PO_4\text{-P}$ than all treatments with the exception of NT-M and NT-MCM (Fig 12). Of the 9 treatments that were significantly lower in $PO_4\text{-P}$ concentrations, 8 of them included some form of soil disturbance (Table 9). Visual analysis of the treatment graph also confirms that no-till treatments appear higher than either traditional till or zonal till. Based on ANOVA, Duncan's New Multiple Range test, and a visual assessment it seems that tillage method had a significant effect on $PO_4\text{-P}$. Specifically, that no-till was associated with greater $PO_4\text{-P}$ concentrations.

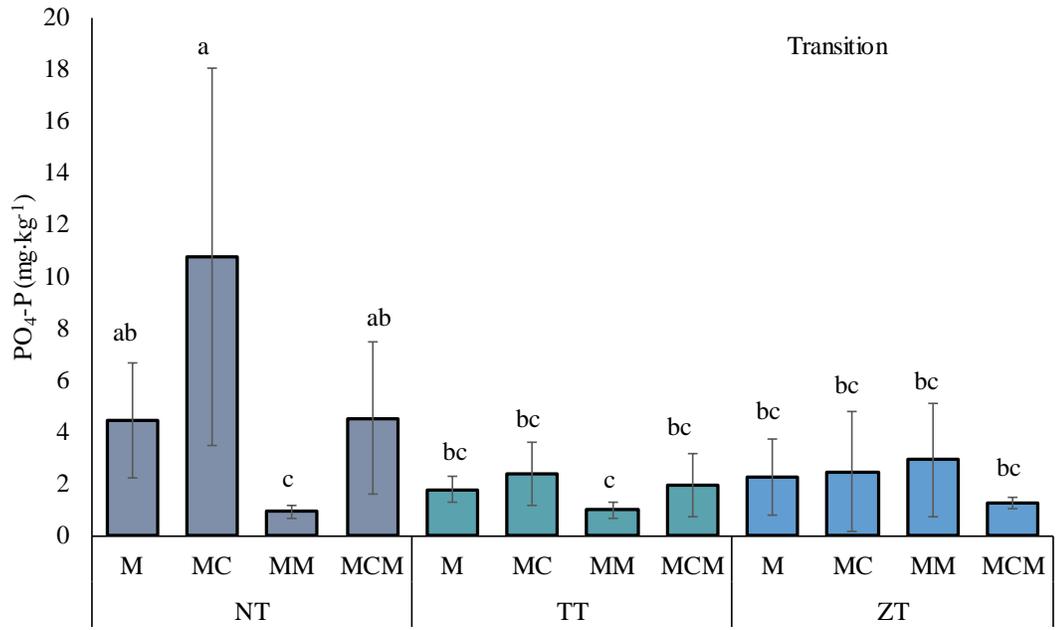


Fig. 12. Mean extractable soil PO₄-P for each treatment in the Transition. The main plot factors are tillage methods abbreviated as follows: no-till (NT), traditional tillage (TT), and zonal till (ZT). The sub-plot factors are cropping systems, abbreviated as follows: continuous maize (M), maize-cowpea (MC), maize-mucuna (MM), and maize-cowpea-mucuna relay (MCM). Error bars are standard deviation. Differences in lower case letters indicate a significant difference between treatments. Data shown is untransformed data but the Duncan's new multiple range test was performed on transformed data ($\alpha < 0.05$).

Table 9. Fertilizer treatment NT-MC had higher soil NO₃-N concentrations than the following 9 other fertilizer treatments.

		NT-MM
		TT-M
		TT-MC
		TT-MM
NT-MC	>	TT-MCM
		ZT-M
		ZT-MC
		ZT-MM
		ZT-MCM

The second major difference in the PO₄-P concentrations among treatments is that 2 treatments (NT-MM and TT-MM) were significantly lower in PO₄-P concentrations than 3 other treatments (NT-M, NT-MC, and NT-MCM). The 2 low performing treatments both included the maize-mucuna rotation, whereas the higher PO₄-P treatments did not. In addition, of the 3 maize-mucuna treatments 2 of them were determined to have significantly low PO₄-P concentrations and the other was similarly low in PO₄-P concentrations. This seems to suggest that the maize-mucuna rotation is associated with low soil extractable PO₄-P in the Transition. These results seem to suggest two major points: (1) no-till is associated with greater PO₄-P, and (2) maize-mucuna cropping systems have lower PO₄-P than the higher performing NT treatments in the Transition.

Davies (2014) reported PO₄-P concentrations ranging from 4.4 to 8.0 mg kg⁻¹ for soils collected in the Transition in 2012 from the NT-MM and ZT-M respectively. He found no significant effect of tillage and cropping on PO₄-P concentrations for Year 2 of the study. Overall PO₄-P concentrations were lower in this study compared to those in in 2012 (Davies, 2014), possibly due to improved crop productivity from multiple years of fertilization that may have stimulated greater crop uptake of P in 2013.

2.4.5. Guinea Savannah Treatment Effects

This data was not normally distributed which may play a role in the lack of differences detected by parametric tests. The only extractable soil nutrient that demonstrated any treatment effect was for PO₄-P in the Guinea Savannah. A significant tillage x cropping interaction was noted ($p = 0.01$)

2.4.5.1. Guinea Savannah: PO₄-P

Univariate analysis of variance indicated that cropping system ($p = 0.049$) and the interaction of tillage and cropping system ($p = 0.01$) had a significant effect on soil PO₄-P in this agro-ecosystem. ANOVA for individual tillage treatments revealed that cropping had a significant effect on PO₄-P in the zonal tillage ($p = 0.042$) and the no tillage ($p = 0.039$). Means comparisons revealed that the NT-M treatment had significantly higher extractable soil PO₄-P concentrations than all other treatments except for TT-CM (Fig. 13). One treatment had exceptionally greater PO₄-P concentrations than the other treatments. Data was evaluated for potential outliers, but the treatments were consistent among all reps; there did not seem to be an outlier.

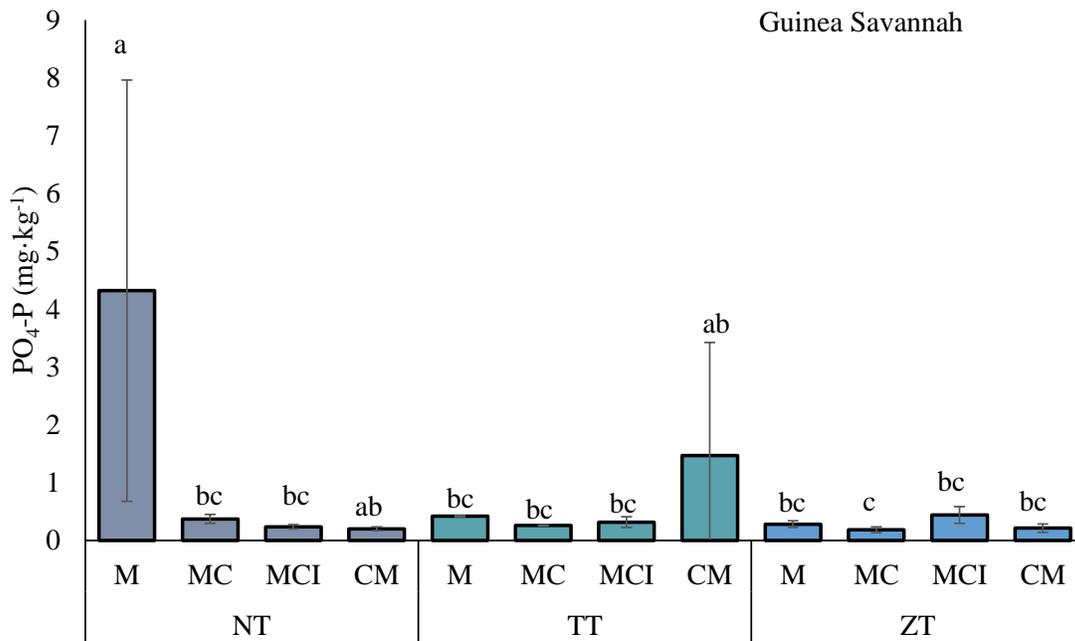


Fig. 13. Mean extractable soil PO₄-P for each treatment in the Guinea Savannah. The main plot factors are tillage methods abbreviated as follows: no-till (NT), traditional tillage (TT), and zonal till (ZT). The sub-plot factors are cropping systems, abbreviated as follows: continuous maize (M), maize-cowpea (MC), maize-mucuna (MM), and maize-cowpea-mucuna relay (MCM). Error bars are standard deviation. Differences in lower case letters indicate a significant difference between treatments. Data shown is untransformed data but the Duncan's new multiple range test was performed on transformed data ($\alpha < 0.05$).

Unlike in the Transition zone, the PO₄-P results of the Guinea Savannah do not seem to reveal a tendency of the no-till treatments to have higher PO₄-P. For example, only the NT-M had high PO₄-P. All the other treatments featuring no-till had similar concentrations of PO₄-P compared to the majority of the other tillage types. The ZT-CM treatment was significantly lower in PO₄-P than several treatments including NT-M,

MT-CM, and TT-CM. Considering the lack of normality in the data, it is not clear if the PO₄-P demonstrated a clear management affect.

2.5. CONCLUSION

Widespread adoption of certain tillage methods or cropping systems throughout Ghana is unlikely to result in enhanced extractable soil nutrients, according to this study. The Coastal Savannah agro-ecosystem tended to display higher concentrations of inorganic-N (NO₃-N and NH₄-N) when compared to all other agro-ecosystems, whereas the Forest agro-ecosystem tended to display extractable higher organic N and C. Prior land use coupled with soil texture in an agro-ecosystem will influence whether N is found in the organic or inorganic form. Results after three years of treatments indicated maize monocrop was associated with greater soil extractable NO₃-N in the Coastal Savannah, no-till was associated with greater soil NO₃-N and PO₄-P in the Transition, while the maize-mucuna rotation in the Transition was associated with higher NO₃-N and lower PO₄-P. In addition, NO₃-N and PO₄-P were the soil nutrients that responded most strongly to management changes, while EON and EOC concentrations revealed no differences among treatments within a given agro-ecological zone.

One limitation of this investigation was the strategy of sampling within a crop row. As mentioned previously, this method of sampling did not adequately capture differences between traditional and zonal tillage. Future investigations should consider a sampling strategy that can better detect differences between zonal and traditional till, such as random sampling or sampling both between rows and within rows.

Based on the results of this investigation, more years of the current crop management treatments would be very useful before making recommendations to subsistence farmers. Soil collection and analysis in the 5th and final year of this may reveal more differences in nutrient concentrations by treatment within each agro-ecosystem. To the extent possible these results should be linked with crop yield and biomass data. A medium-term crop management investigation with soil fertility and yield results is likely to provide highly valuable information to smallholder farmers in these regions.

3. MAXIMIZING USE OF FERTILIZER AND ORGANIC ADDITIONS IN GHANANIAN SUBSISTENCE AGRICULTURE

3.1. INTRODUCTION

Mineral fertilizers and organic inputs are essential to enhancing nutrient depleted tropical soils. Currently, many Sub-Saharan African (SSA) countries promote the same mineral fertilizer application rates throughout the country, irrespective of regional climates and cropping systems. Such universal fertilizer rates are often an inefficient use of expensive inputs. Moreover, previous research investigating the effects of mineral and organic inputs on soil nutrients and crop yield in tropical regions have found mixed results, generally due to variations in climate, physical conditions, and agronomic practices (Srinivasarao et al., 2012). These mixed results indicate the need for locally tested fertilizer recommendation rates.

3.1.1. Role of Mineral Fertilizers in SSA Agriculture

Fertilizers are critical to the future of SSA farming. Indeed half of the entire world's yield gains over the past 75 years are due to fertilizer use (Braun and Roy, 1983). Fertilizers have the potential to influence crop yield, plant nutrient status, nutrient cycles, soil water storage, bulk density, pH, and a number of other agronomic related factors. There are numerous ways to classify fertilizers: mineral versus organic, based on fertilizer analysis, or by method of preparation (Troeh and Thompson, 2005). The most common N fertilizer sources are anhydrous ammonia, urea, ammonium nitrate and slow

release fertilizer (Foth and Ellis, 1997). Some of the common P fertilizers are superphosphate, triple superphosphate, ammonium phosphate, and rock phosphate (Foth and Ellis, 1997). Potassium fertilizers and micro-nutrient fertilizers are also common but beyond the scope of this study (Foth and Ellis, 1997).

Manufactured fertilizers are very common and extremely important to successful crop production. Even in SSA where access to expensive fertilizer may be inconsistent for some farmers and where other forms of fertilizer are often used, manufactured fertilizer is still immensely necessary. Previous research in nutrient depleted tropical soils has shown that conservation agriculture (CA) practices are most effective at improving soil fertility and yield when combined with mineral fertilizers (Ouédraogo et al., 2007; Srinivasarao et al., 2012; Nyamangara et al., 2013; Logah et al., 2011). One such example is from a 20 year soil amendment study on Alfisols in southern India (Srinivasarao et al., 2012). In this study organic inputs and mineral fertilizers were applied at different rates: 100% of the recommended dose of fertilizer (RDF) (20:40:40 N, P₂O₅, K₂O), 50% RDF + groundnut shells, 50% RDF + farmyard manure, farm yard manure alone, and a control. They found the highest soil organic carbon (SOC) and yield was from the plots treated with 50% RDF+ groundnut shells and the plots treated with 50% RDF + farmyard manure (Srinivasarao et al., 2012). Such a result suggests that it is the combination of mineral fertilizers plus organic inputs that yields the greatest soil fertility improvements.

Other findings from Srinivasarao et al., (2012) are also useful. The application of fertilizer alone (100% RDF treatment) served only to maintain the soil C stocks at the antecedent level, preventing nutrition depletion over the 20 year study period, but not improving overall soil fertility (Srinivasarao et al., 2012). The manure alone application did improve C sequestration at a higher degree than the fertilizer alone, but not as well as the combination of organics and fertilizers (Srinivasarao et al., 2012). This suggests that, at least in a groundnut based cropping system, to achieve long term yield sustainability N-P-K fertilizer plus organic residues are needed.

Where Srinivasarao et al. (2012) found that the addition of organics alone improved C levels slightly, other CA experiments in nutrient-depleted zones have reported a decline in soil fertility from the addition of organic inputs without a complimentary application of mineral fertilizers (Nyamangara et al., 2013; Ouédraogo et al., 2007). This effect was shown in a tillage, amendment, and cropping system study on Arenosols in four agro ecological zones of Zimbabwe (Nyamangara et al., 2013). In this study, the effect of both the mulching treatment plus the legume cropping system led to maize grain yield declines of 28-48 percent (Nyamangara et al., 2013). However, when mineral fertilizer was added, the effect of the mulching + legumes was an increased yield of 7 to 69 percent (Nyamangara et al., 2013). This and other studies indicate that for some cropping systems, adopting CA without providing adequate amounts of N-P-K can lead to worsening yield and soil infertility. The Srinivasarao et al. (2012) and

Nyamangara et al. (2013) studies illustrate the importance of fertilizer to a dryland cropping system.

3.1.2. Residues and Organic Inputs

Organic inputs are another key factor of CA. Organic inputs such as crop residues, green manures, and animal manures have the potential to improve soil moisture, nutrient availability, and structure (Singh et al., 2007; Sommer et al., 2011). Organic inputs provide nutrients and energy to the microbial community. Once processed by the microbial community the resulting nutrients support plant growth, as well as enhance water holding capacity and improve soil structure. Research results have evidenced increased crop biomass and yields in mulched crops, particularly on degraded soils under dry conditions (Bationo and Buerkert, 2001; Buerkert et al., 2002 as cited by Lahmar et al., 2012). The potential for this CA practice to have positive impacts on SSA agriculture seems high. However, the quality of the residue is critical to the effectiveness of this practice (Singh et al., 2007; Sakala et al., 2000; Ouédraogo et al., 2006).

For poorer quality residues, that is, those with high carbon to nitrogen (C:N) ratios, the capacity for N immobilization may exceed the amounts of plant available N (Singh et al., 2007). Kaizzi et al. (2004) suggested that incorporating high-quality legumes such as *Mucuna pruriens* in farming systems may greatly improve nutrient balances at a much lower cost than typical mineral fertilizers. In a Ugandan farm study, Nkonya et al. (2005) found that households with livestock had higher soil nutrient balances, indicating a positive relationship between animal manures and soil fertility.

By many accounts organic residues is the most critical component of CA for degraded semi-arid tropic lands and may also be the most significant limiting factor for the effectiveness of other CA components such as no-till (Lahmar et al., 2012; Lal, 2007). Yet organic resources are especially scarce in SSA cropping systems because of the wide and varied uses of crop residues and the low biomass production of many crops. For example, crop residues may have higher value uses than mulching (Lahmar et al., 2012), such as feed for livestock and household purposes (Valbuena et al., 2014) and may at times fetch higher prices than grain on local markets in the Sahel (Lahmar et al., 2012). Therefore, identifying cropping systems that produce high amounts of residue and have economic value is critical.

3.2. OBJECTIVES & HYPOTHESIS

The objective of this experiment was to determine the effect of mineral fertilizer and organic inputs and their interactions on soil nutrient status in the four agro-ecosystems of Ghana.

Hypothesis 1. The addition of inorganic P will significantly improve soil fertility, quantified as 0.1 M HCl extractable N and C and Bray extractable $\text{PO}_4\text{-P}$, in all climatic zones because P tends to be a limiting nutrient in highly weathered soils of the tropics.

Hypothesis 2. The combination of mineral fertilizer and compost in moderate and high rainfall regimes will significantly improve soil fertility, quantified as 0.1 M HCl extractable N and C and Bray extractable $\text{PO}_4\text{-P}$, due to microbial mineralization of

compost when enough N-P-K mineral fertilizer is present and suitable soil moisture is available.

Hypothesis 3. In low rainfall regimes regions, the addition of compost will not significantly improve soil fertility, quantified as 0.1 M HCl extractable N and C and Bray extractable PO₄-P, to due to inability of the microbial community to mineralize N in the absence of adequate soil moisture.

3.3. METHODOLOGY

3.3.1. Experimental Sites

The project was established January 2011 in the Coastal Savannah (5°42'6N and 0°17'15W) Forest (6°34'11N and 1°51'26W), Forest-Guinea Savannah Transition (7°26'12.6"N and 1°29'31.6"W), and Guinea Savannah (9°31'18N and 0°55'9W) of Ghana.

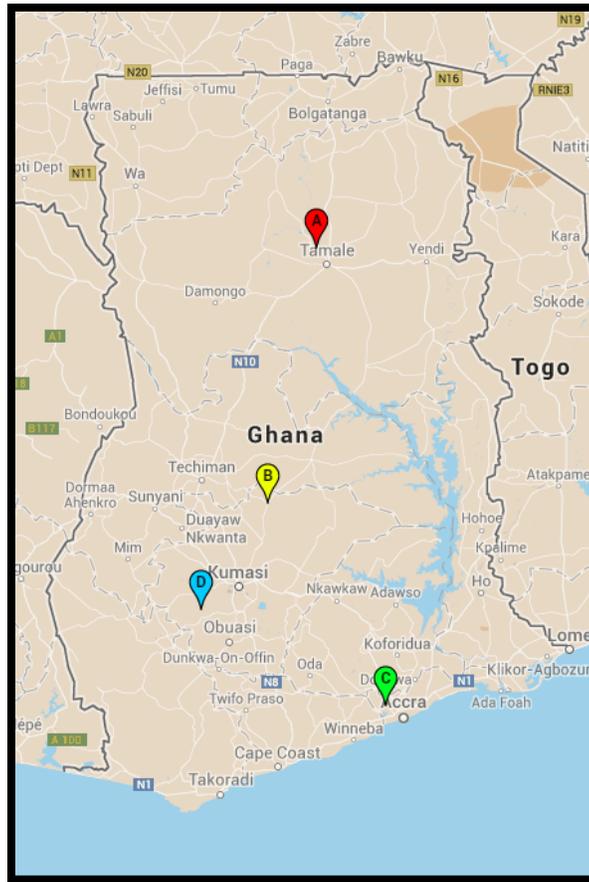


Fig. 14. Experimental plot locations in Ghana. “A” is the Guinea Savannah zone site; “B” is the Transition zone site; “D” is the Forest zone site; and “C” is the coastal savannah zone site. Source: Developed by Pitts, L.K.R. with Google. Accessible via hyperlink <https://www.google.com/maps/d/edit?mid=zVFhgsrtd3Aw.kgupxkJDLLmY>

3.3.1.1. Coastal Savannah

The Coastal Savannah experimental site is located in the Ga West District near the town of Pokuase. It lies on the southern coast of Ghana, north of the Capital of Accra (Fig. 14). The research plot lies adjacent to a wooded area and is at a shoulder position in the landscape. Mean annual rainfall in the Coastal Savannah is 800 mm (Oppong-

Anane, 2001). At this site the primary growing season is 100-110 days long and runs from March to June, while the minor growing season is about 60 days starting in October (Oppong-Anane, 2001).

Soils at this site were classified using the World Reference Base (WRB) soil classification system as a Haplic Lixisol by the Ghanaian government's Soil Research Institute (SRI). Haplic Lixisols are soils with clay-enriched subsoils that exhibit high base saturation and low-activity clays (WRB, 2006). The Haplic prefix qualifier indicates a typical expression of Lixisol features, with no other distinctive characteristics (WRB, 2006). The local soil series is Adawso developed over granite. Measured pH of the site was 5.94-6.25 (Kofi Boa, personal communication, December 2013).

Field observations indicated coarse-textured soils, with large size sand particles. The slope of the site is less than 4%. Unlike other agro-ecosystems where a great variety of insect species were observed, in the Coastal Savannah termites and ants were the only signs of a robust living soil community. Also of note is that this site was previously under cultivation for many years prior to starting the trials.

3.3.1.2. Forest

The Forest experimental site is located in Amansie West District near the town of Ahwerewa. It is North-West of the Coastal Savannah site (Fig. 14). This plot is nestled in the interior of a lush cocoa plantation and natural wooded area, approximately 50 m off a main road. Mean annual rainfall in the Forest site is 1500 mm (Oppong-Anane, 2001). There are two growing seasons. The major season of 150-200 growing days lasts

from March to July, and a minor season of about 90 days starts in September or October (Oppong-Anane, 2001; Obuobie et al., 2006).

The soils of this site are a Leptic Lixisols which are soils with an accumulation of clay with a high base saturation and low-activity in the subsoil layer (WRB, 2006).

Unlike the Coastal Savannah, the Forest site soils have a Leptic qualifier, indicating continuous rock starting within 100 cm of the soil surface (WRB, 2006). The local soil series is Amuni developed over phyllite. Soil pH values measured prior to initiation of the trials were 5.56-6.27 (Kofi Boa, personal communication, December 2013).

Field observations of the Forest site include that insects were prolific, with evidence of beetles, centipedes, lady bugs, and ants which is usually a sign of good soil moisture and a healthy soil ecosystem. This site was historically tilled, but had been fallow 2-4 years prior to the start of this trial. This site had the greatest slope between 6-7%.

3.3.1.3. Transition

The Transition site is located in the Ejura-Sekodumase District near the town of Ejura-Adiembra. It is northeast of the Forest experimental site (Fig. 14). This site occupies a summit position on the landscape. Adjacent to the plot are farmlands and a road bordered by about 20 m of grass vegetation. Mean annual rainfall is 1300 mm (Oppong-Anane, 2001). The Transition site has a major growing season with 200-220 growing days and a minor season of 60 growing days (Oppong-Anane, 2001).

The Transition site crops were planted into a Leptic Lixisol, the same soil as the Forest site. This site has the most acidic soils of all four agro-ecosystems with measured baseline pH values from 4.7-4.8 (Kofi Boa, personal communication, December 2011). Slope at this site is approximately 3%.

3.3.1.4. Guinea Savannah

The Guinea Savannah lies in northern Ghana, outside the city of Tamale. This plot is located in the Tolon-Kumbungu District near town of Kumbungu-Kukuo (Fig. 14). Surrounding the Guinea Savannah experimental plots is farmland and a major road less than 5 m from the plots.

Mean annual rainfall at this site is 1100 mm (Oppong-Anane, 2001). This is the only experimental site with one growing season which starts in April or May and ends in September or October and has 180-200 growing degree days. The soils of this site are Pisoplinthic Plinthisols. Pisoplinthic Plinthosols are soils with an accumulation of iron under hydromorphic conditions (WRB, 2006). The Pisoplinthic prefix qualifier indicates a Pisoplinthic horizon starting within 100 cm of the soil surface (WRB, 2006). This plot was the flattest with a slope of less than 2%. Baseline pH values measured prior to initiation of the experiment ranged from 5.3-5.56 (Kofi Boa, personal communication, December 2013).

3.3.2. Fertilizer Treatments

Triple superphosphate (TSP), urea, and compost fertilizer treatments were laid out in a maize (*Zea mays*) monoculture based split-plot design. TSP was the main plot

treatment (Table 10). Urea and compost were randomly assigned to the sub-plot treatments. There were three replications for each treatment combination. The rates of TSP and urea were adjusted after the first year of treatments due to poor crop response. In 2012 TSP fertilizer rates were raised from 0, 6.5 and 13 kg ha⁻¹ to 0, 20 and 40 kg ha⁻¹ and urea application rates were raised from 0, 45 and 90 kg ha⁻¹ to 0, 70 and 140 kg ha⁻¹. The compost rates were maintained at 0, 3 and 6 Mg ha⁻¹ but the source was changed from Ecofertilizer (3.2% N, 3.3% P₂O₅ and 4.5% K₂O) to Asaase Nufusuo (cocoa (*Theobroma cacao*) husk) (3.2% N, 3.2% P₂O₅ and 1.3% K₂O) since the Ecofertilizer was suspected to be immobilizing N due to high input C:N ratios based on treated plants looking very pale in the first year. These changes were maintained for the 2013 growing season when the fields were sampled for the year of study evaluated in this chapter.

Table 10. Factors treatments

Plot	Treatment	Rate
Main Plot	TSP	0 kg ha ⁻¹
		20 kg ha ⁻¹
		40 kg ha ⁻¹
Sub-Plot (A)	Urea	0 kg ha ⁻¹
		70 kg ha ⁻¹
		140 kg ha ⁻¹
Sub-Plot (B)	Compost	0 t ha ⁻¹
		3 Mg ha ⁻¹
		6 Mg ha ⁻¹

3.3.3. Plot Management

Fields were prepared for planting by hand slashing indigenous vegetation and spraying with glyphosate herbicide in all agro-ecosystems. Mineral fertilizers were point placed in hand dug seed holes. Compost was surface applied as a ring around each plant. Two seeds were placed per hole and covered with soil. Seed spacing was 40 cm and row spacing was 80 cm. The cropping system for all four agro-ecosystems was continuous maize. Fields were maintained by hand weeding or hand hoeing. Rain gauges were installed at all sites in the latter part of May of 2011.

3.3.4. Soil Collection and Processing

Soil samples were collected in December 2013 from all sites. All plots were composite sampled to a 0-15 cm depth. Three cores were taken from the middle row of each plot using a 2-cm diameter auger and placed in cotton soil bags. Samples were laid out to air dry within 4-48 hours of collection. Prior to shipment from Ghana to the NAWA lab at Texas A&M University, cotton bags were placed in sealed zip-lock plastic bags contained within sealed Tupperware containers.

On arrival at Texas A&M University the soils were logged into the NAWA laboratory soils database where they were given a unique ID number to track their analysis and chain of custody as required by BL2 protocols. Soils were further air-dried prior to gently breaking up any large soil peds using a mortar and pestle before sieving through a 2-mm sieve.

3.3.5 Soil Extractions

To quantify nitrogen and carbon concentrations, 3.5 g of soil was combined with 30 mL of 0.1 M HCl and shaken for two hours at 500 rpm on a rotary shaker. Samples were then centrifuged for 15 minutes at 19,974 g-force and filtered using a Whatman GF/F filter (nominal pore size 0.7 μm) to remove any floating organic material in the supernatant. Extracts were diluted with ultrapure water to ensure enough sample was available for chemical analysis. Extracts were analyzed between 1 and 24 hours after extraction.

To quantify orthophosphate-P, 3 g of soil was combined with 21 mL Bray 1 solution and shaken for 1 minute on a rotary shaker (Bray, 1945). Samples were then centrifuged for 5 min at 2,809 g-force and filtered with Whatman GF/F filters (nominal pore size 0.7 μm) to remove any floating organic material. Extracts were analyzed between 1 and 24 hours after extraction.

3.3.6 Chemical Analyses

Chemical analyses were performed to quantify $\text{NO}_3\text{-N}$, $\text{NH}_4\text{-N}$, EON, TEN, EOC, and $\text{PO}_4\text{-P}$ concentrations. Extractable organic carbon (EOC) and total extractable nitrogen (TEN) was measured using a high temperature Pt-catalyzed combustion with a Shimadzu TOC-VCSH and Shimadzu total measuring unit TNM-1 (Shimadzu Corp. Houston, TX, USA). EOC was measured as non-purgeable carbon, which entailed acidifying the sample (250 μL 2 M HCl) and sparging for 4 min with C-free air. $\text{NH}_4\text{-N}$ was analyzed using the phenate hypochlorite method with Na nitroprusside enhancement

(USEPA method 350.1) and NO₃-N using Cd-Cu reduction (USEPA method 353.3). PO₄-P was analyzed using the ascorbic acid, molybdenum blue method (APHA 1992). Colorimetric methods were performed using a Westco Scientific Smartchem Discrete Analyzer (Westco Scientific Instruments Inc. Brookfield, CT, USA). Sample replicates, blanks, NIST (National Institute of Standards and Technology) traceable and check standards were run every 12th sample to monitor instrument precision. Instrument lower detection concentrations were 0.1 mg L⁻¹ for NO₃-N, NH₄-N and PO₄-P, 0.5 mg L⁻¹ for TEN and EOC and 0.7 mg L⁻¹ for EON.

3.3.7. Statistical Analyses

Prior to performing statistical analyses, data were reviewed for outliers and samples were re-run as necessary. Data were evaluated for meeting the following key assumptions of analysis of variance (ANOVA):

1. There are k simple random samples from k populations.
2. The k samples are independent of each other; that is, the subjects in one group cannot be related in any way to subjects in a second group.
3. The populations are normally distributed.
4. The populations have the same variance; that is, each treatment group has population variance s^2 .

To meet the assumptions of ANOVA, data were tested for normality using the Shapiro-Wilks test. Soil nutrient concentrations from each agro-ecosystem were evaluated separately for a p-value greater than 0.05, meaning that the data was not

significantly different from a normal distribution curve. Normality tests revealed that the data was not normally distributed. Several transformations were performed, a logarithmic function was ultimately chosen because it was the most effective at achieving normal distribution for the majority of the data. To ensure equal population variance the largest treatment or sample standard deviation was confirmed as being no more than twice as large as the smallest sample standard deviation.

Univariate analysis of variance with a general linear model (GLM) procedure was conducted using SPSS v. 16 software to determine differences in extractable nutrient concentrations ($\text{NO}_3\text{-N}$, $\text{NH}_4\text{-N}$, TEN, EON, EOC, and $\text{PO}_4\text{-P}$) across all agro-ecosystems of Ghana. Fixed factors were as follows: 1) agro-ecosystem, 2) TSP, 3) urea, and 4) compost. Interaction effects were as follows: 1) agro-ecosystem x TSP, 2) agro-ecosystem x urea, 3) agro-ecosystem x compost, 4) TSP x urea, 5) TSP x compost, 6) urea x compost, 7) agro-ecosystem x TSP x urea, 8) agro-ecosystem x TSP x compost, 9) agro-ecosystem x urea x compost, 10) TSP x urea x compost, 11) agro-ecosystem x TSP x urea x compost.

Univariate analysis of variance was also used to determine factor effects on the concentrations of $\text{NO}_3\text{-N}$, $\text{NH}_4\text{-N}$, EON, TEN, EOC, and $\text{PO}_4\text{-P}$ within an agro-ecosystem. The fixed factors were 1) TSP, 2) urea, and 3) compost. Interaction effects were: 1) TSP x Urea, 2) TSP x Compost, 3) Urea x Compost, and 4) TSP x Urea x Compost. Differences in individual treatment combinations were determined with Duncan's New Multiple Range Test ($\alpha < 0.05$). If any interactions occurred for any

nutrient then these were examined further using ANOVA within each individual fertilizer in turn. Significant effects of TSP or urea and compost and their interactions (univariate analysis of variance) were determined at $p < 0.05$.

3.4. RESULTS & DISCUSSION

Mineral fertilizers are critical to the improvement of Sub-Saharan African farming. The importance of mineral fertilizers in combination with CA cannot be overstated. Previous research in nutrient depleted tropical soils has shown that CA practices are most effective at improving soil fertility and yield when combined with mineral fertilizers (Ouédraogo et al., 2007; Srinivasarao et al., 2012; Nyamangara et al., 2013; Logah et al., 2011). The current study sought to examine the effect of soil amendments across Ghana, West Africa and within agro-ecosystems of Ghana on residual soil nutrient concentrations using a 0.1 M HCl extract. Examination of the statistical analyses showed that hypothesis 1 which stated that the addition of inorganic-P would significantly improve soil fertility, quantified as HCl-extractable N and C and Bray extractable $\text{PO}_4\text{-P}$, in all climatic zones because P tends to be a limiting nutrient in highly weathered soils of the tropics failed to be rejected. Indeed, the addition of TSP did have a significant effect on at least one soil nutrient evaluated in all four agro-ecological zones evaluated.

Hypothesis 2 which stated that “the combination of mineral fertilizer and compost in moderate and high rainfall regimes will significantly improve soil fertility, quantified as HCl-extractable N and C and Bray extractable $\text{PO}_4\text{-P}$, due to microbial

mineralization of compost when enough N-P mineral fertilizer is present and suitable soil moisture is available” was not accepted. It was expected that moderate and high rainfall regimes, such as the Forest and Transition agro-ecosystems, would show soil fertility gains under a combination of mineral fertilizer plus compost. This was not true. In the Forest, the zone with the highest effective rainfall of this study, neither urea, compost, nor their interaction had a significant effect on any nutrients. Only TSP had an impact on DOC, leading to a rejection of hypothesis 2.

Hypothesis 3 which stated “in low rainfall regime regions, the addition of compost will not significantly improve soil fertility, quantified as HCl-extractable N and C and Bray extractable PO₄-P, due to the inability of the microbial community to mineralize N in the absence of adequate soil moisture” was also not accepted. Compost did in fact have a significant effect on NO₃-N and PO₄-P concentrations in the Guinea Savannah.

3.4.1. Results Across Agro-ecosystems

Data across all agro-ecosystems were aggregated and evaluated for factor effects from agro-ecosystem, TSP, urea, compost and their interactions. Agro-ecosystem had a significant effect on the concentrations of all soil nutrients evaluated. Whereas TSP, urea, compost, and their interactions were only significant for some of the soil nutrients measured.

3.4.1.1. N-results: NO_3 -N, NH_4 -N and EON

Univariate analysis of variance revealed that soil extractable NO_3 -N was significantly affected by the agro-ecosystem type ($p < 0.0001$), urea ($p < 0.0001$), and compost amendments ($p < 0.0001$). Duncan's new multiple range tests showed that NO_3 -N concentrations were equally high in both the Coastal and Guinea Savannahs, while the Forest and Transition zones had successively lower NO_3 -N concentrations (Fig. 15). Lower NO_3 -N concentrations were observed in the Forest and Transition agro-ecosystems, which are zones with higher effective rainfall and greater crop biomass production than the other agro-ecosystems. The higher OM contributions in these zones may have led to net immobilization of available N as the microbial community utilized carbon substrate. Additionally, greater plant uptake of this highly mobile, plant-available N form may have led to lower residual NO_3 -N concentrations in the soil.

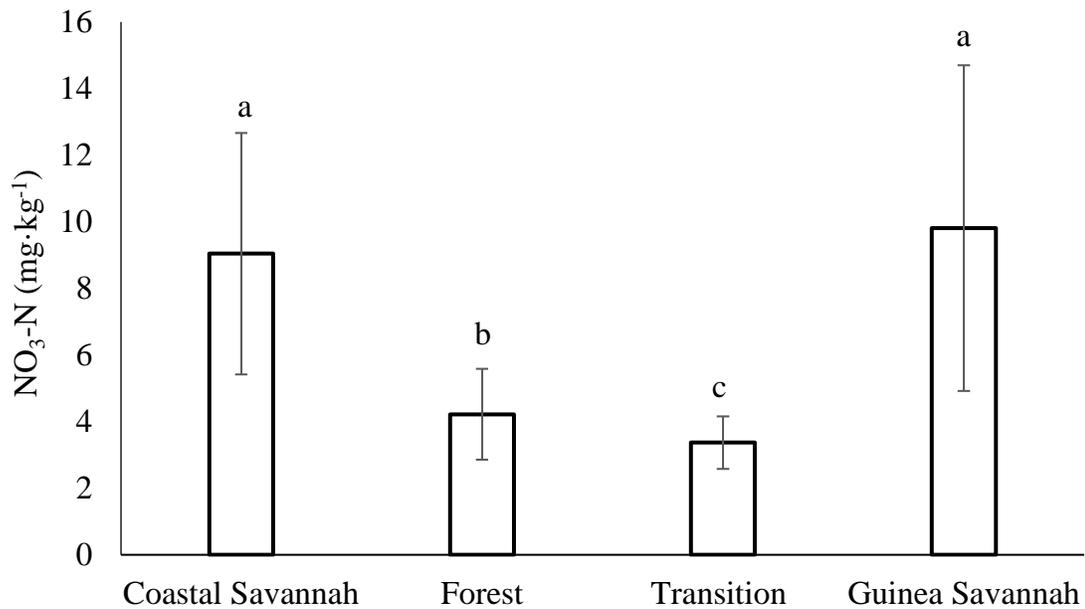


Figure 15. Mean extractable soil NO₃-N concentrations. Means shown are raw data. Error bars are standard deviation. Differences in lower case letters indicate a significant difference at $\alpha < 0.05$. Duncan's new multiple range test was performed on transformed data.

The Coastal and Guinea Savannahs, with lower rainfall and biomass production, had higher concentrations of NO₃-N than the other agro-ecosystems. The limited biomass production of these zones may have provided insufficient C sources to lead to microbial immobilization of inorganic-N, thereby allowing for higher residual NO₃-N concentrations. The sandy soil texture and uneven rainfall distribution specifically in the Coastal Savannah may have also inhibited microbial utilization of NO₃-N by providing inadequate soil conditions for an active microbial community. Another possibility for the higher NO₃-N may be the result of crop growth. The precipitation constraints of both the

Guinea and Coastal Savannahs may have presented a greater limiting factor to plant growth than soil nutrients, thereby reducing crops utilization soil available $\text{NO}_3\text{-N}$, resulting in higher residual $\text{NO}_3\text{-N}$ concentrations. Either of these scenarios supports the finding of significantly higher extractable soil N in the Coastal Savannah.

Agro-ecosystem type also had a significant effect on concentrations of $\text{NH}_4\text{-N}$ ($p < 0.0001$). No other factors were determined to have a significant effect on $\text{NH}_4\text{-N}$ concentrations. Means comparison showed that the Coastal Savannah and Forest agro-ecosystems had equally high amounts of $\text{NH}_4\text{-N}$, while the Transition and Guinea Savannah agro-ecosystems had equally low concentrations (Fig. 16).

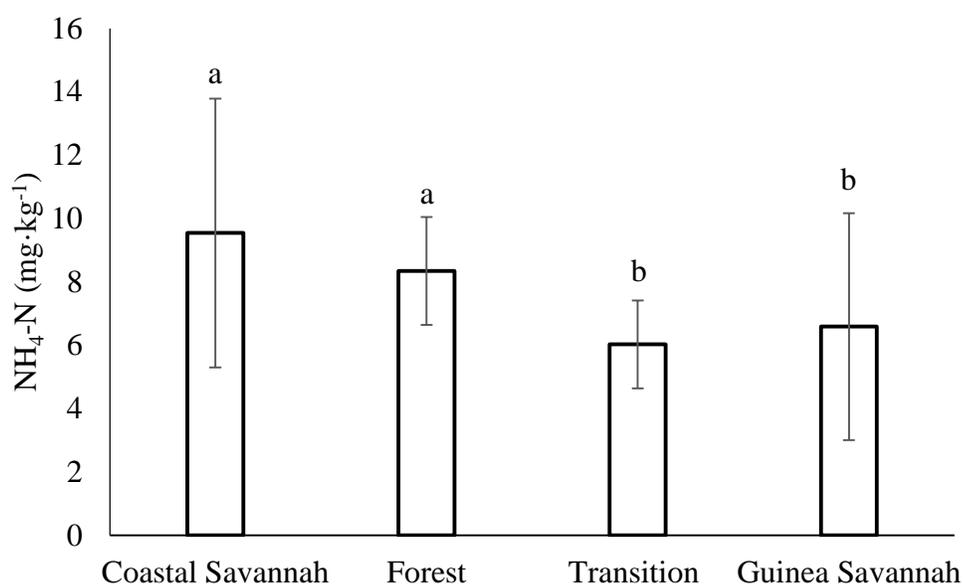


Fig. 16. Mean extractable soil $\text{NH}_4\text{-N}$ concentrations. Means shown are raw data. Error bars are standard deviation. Differences in lower case letters indicate a significant difference at $\alpha < 0.05$. Duncan's new multiple range test was performed on transformed data.

A significant effect on EON concentrations was also observed among agro-ecosystem types ($p < 0.0001$). Means comparison testing found the Forest agro-ecosystem to have higher EON concentration than the Transition and Coastal Savannah, but a similar concentration to the Guinea Savannah (Fig. 17). The Guinea Savannah soils were only greater in EON concentrations than the Coastal Savannah (Fig. 17).

EON is that easily solubilized N pool associated with soil biomass and soil organic matter (Ros et al., 2009) that responds similarly to wetting and drying as does DOC (Xiang et al., 2008). Meta-analysis of DON and DOC research by Ros et al. (2009) found that DON concentrations were lower with soil $\text{pH} > 6$ and lower in sandy soils. Ros et al. (2009) determined that higher DON concentrations were observed during spring/summer seasons, and under grassland when compared to arable land. The findings of the current study are somewhat consistent with Ros et al.'s meta-analysis (2009). The Forest zone likely accumulated the highest EON concentrations due to its high biomass production which both provided an OM source and stimulated microbial activity and its loamy soil texture which allowed for greater adsorption of EON than the sandy soils of the other agro-ecosystems.

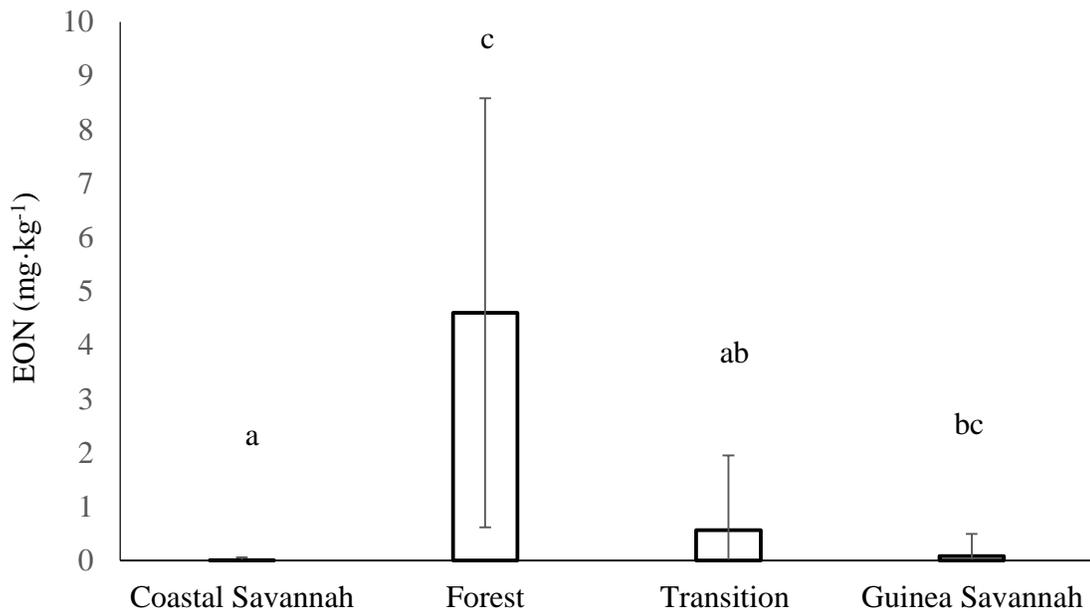


Fig. 17. Mean soil extractable organic nitrogen (EON) concentrations. Means shown are raw data. Error bars are standard deviation. Differences in lower case letters indicate a significant difference at $\alpha < 0.05$. Duncan's new multiple range test was performed on transformed data.

That the Forest had significantly higher EON concentration is consistent with much of the Ros et al. (2009) meta-analysis findings. Because EON is closely linked to organic matter and microbes, it follows that the agro-ecosystem with the highest annual rainfall, greatest biomass production, and loamiest texture is likely to have the most active microbial community and provide for the greatest accumulation of organically bound nitrogen.

The distribution of N species in an ecosystem gives some indication of anthropogenic impacts to that ecosystem which is typically evaluated using the DON:

TDN ratio (Pellerin et al., 2006). Relatively undisturbed ecosystems generally have a DON:TDN ratio of 65 or greater, whereas for urban ecosystems, particularly downstream of a waste water treatment facility, the DON:TDN ratio will be near 10 (Aitkenhead-Peterson et al., 2009). The 0.1 M HCl extracted DON:TDN ratios of soil in the current study ranged from 0 in the savannah agro-ecosystems to 27 in the Forest agro-ecosystem (Fig. 18).

These are extremely low proportions of EON in the agro-ecosystem studied and suggest that although the applications of nitrogen are both organic that the urea is rapidly mineralized in all the agro-ecosystems. The organic nitrogen in the compost is likely in larger organic N molecules comprising amines rather than the easily mineralized amino acids and so the results here are unusual. Examination of the nitrogen distribution in soils collected in 2012 revealed that the DON:TDN ratios for the 4 agro-ecosystems were 1 in the coastal savannah, 20 in the Forest, 10 in the Transition and 5 in the Guinea Savannah compared to 0 in the coastal savannah, 27 in the Forest, 0.06 in the Transition and 0 in the Guineas savannah for soils collected in 2013. A further unusual occurrence was the proportion of total N in the form of $\text{NH}_4\text{-N}$ when comparing the two soil collections. In the 2012 soil collections, $\text{NH}_4\text{-N}$ extracted using the same standard operating procedure comprised 38%, 34%, 6% and 46% of total N compared to 51%, 49%, 61% and 60% in the Coastal Savannah, Forest, Transition and Guinea Savannah agro-ecosystems respectively (Fig. 18).

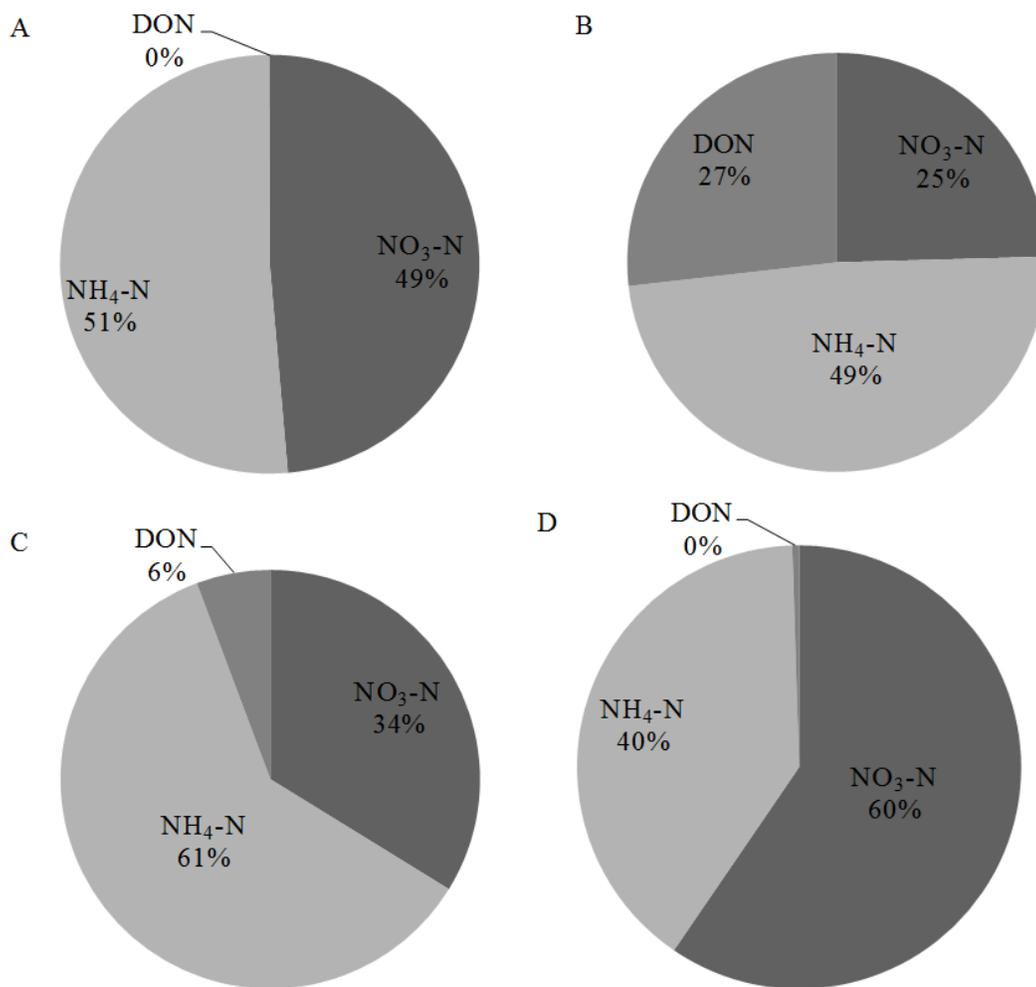


Figure 18. Distribution of nitrogen species in the four agro-ecosystems. A) Coastal Savannah, B) Forest, C) Transition and D) Guinea Savannah.

While EON concentrations were relatively low in the 2012 collection they were barely detectable in the 2013 soil collections except for the Forest agro-ecosystem (Fig. 18). Effects on DON concentrations can also be determined when examining its relationship with DOC (Carrillo-Gonzalez et al., 2013). In a temperate agricultural soil

in Texas, USA the type of extract used to extract soil nutrients tended to decouple the DON vs. DOC relationship but then so did monoculture crops (Carrillo-Gonzalez et al., 2013). Given that this African study was conducted on a maize only monoculture with traditional tillage, it is not surprising that the EON vs EOC relationship is decoupled driven by particularly low extraction of EON. Further discussion on this subject is found in Chapter 4.

3.4.1.2. C-results: EOC

Carbon, measured as EOC, was significantly affected by agro-ecosystem type ($p < 0.0001$). Univariate analysis of variance did not reveal any other factor effects on EOC concentration. Means separation testing showed that the Forest agro-ecosystem had the highest EOC concentration. The Guinea Savannah had the second highest concentration of EOC, followed by the Transition and Coastal Savannah agro-ecosystems which had equally low concentrations (Fig. 19).

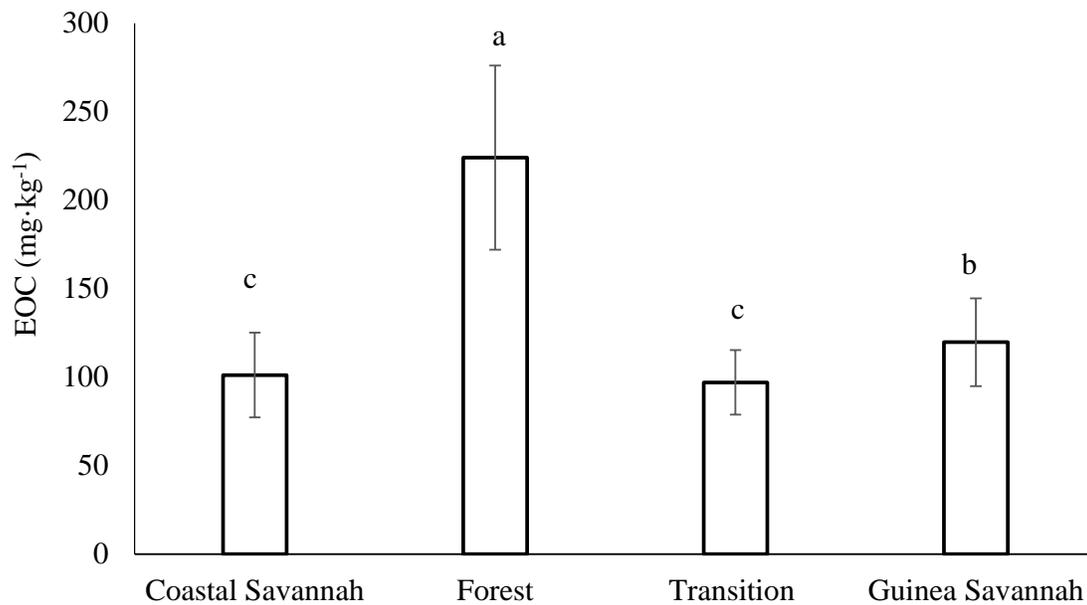


Fig. 19. Mean soil extractable organic carbon (EOC) concentrations. Means shown are raw data. Error bars are standard deviation. Duncan's new multiple range test was performed on transformed data. Differences in lower case letters indicate a significant difference at $\alpha < 0.05$ based on Duncan's test.

EOC production and loss in soil depends on biotic factors such as decomposition, root exudate releases, and microbial turnover (Kalbitz et al., 2000; Aitkenhead-Peterson et al., 2003). These biological activities mediated by soil microbes are heavily dependent on those conditions that will support microbial life such as adequate moisture (Marschner & Kalbitz, 2003). Therefore, it is no surprise that the soil of the Forest agro-ecosystem would have the highest EOC concentrations of all the agro-ecosystems evaluated because of the favorable climatic factors that support an active microbial community to facilitate C cycling, such as high annual rainfall, even rainfall distribution, high plant biomass production, and warm temperatures. There is also a difference in the

lability of DOC when comparing soils obtained from temperate forest and agricultural soils (McDowell et al., 2006) and soils obtained from urban sub-tropical remnant forests and turf grasses (Cioce and Aitkenhead-Peterson, 2015). Water-extracted litter from a Norway spruce forest floor had significantly higher biodegradability (58%) when compared to agricultural soil (30-50%) and agricultural soil had significantly higher biodegradability compared to soil derived from the O horizon of a Norway spruce forest (10%) (McDowell et al., 2006). Thus, if DOC biodegradability is lower in a forest soil which is probably due to greater aromatic C, then recovery of EOC from these soils will also be higher. Many more aromatic-C compounds are likely to be in forest soils which are not readily degraded in addition, and compost supplements are also associated with increasing aromatization in DOM (Chefetz et al., 1998; Said-Pullicino et al., 2007; Caricasole et al., 2010). Cioce and Aitkenhead-Peterson (2015) also reported significantly higher biodegradability from sub-tropical soils under turf grass (52-66%) when compared to soils under wetland forests (23-24%) in south central Texas, USA.

The second highest EOC concentration was observed in the Guinea Savannah and may likely be attributed to wetting and drying effects on C dynamics (Xiang et al., 2008; Guo et al., 2012). The Guinea Savannah was sampled in December well after the October end of the monomodal rainy season. The soils were observed to be very dry during sample collection in the Guinea Savannah. Xiang et al. (2008) demonstrated that extended dry periods increased DOC in surface and subsurface soils. This could be caused by a number of microbial mechanisms that are not well understood. Accumulated

OM in the Guinea Savannah region may have been protected by the finer soil texture (clayey) so that the amount of OM that was decomposed was protected by the soil structure. This protection may have reduced the bioavailability of DOM such as DOM trapped in small pores and thus inaccessible to soil microbes or alternatively through adsorption to soil minerals (Marschner et al. 2008). In general hydrophobic DOM is adsorbed to soil minerals and while it is somewhat biodegradable it is not as biodegradable as DOM in soil solution (Marschner et al., 2008).

3.4.1.3. P-results: Orthophosphate

Phosphorus is generally considered to be limiting in tropical agricultural soils and this may be due to the amount of positively charged iron, zinc and manganese hydroxides found in highly weathered tropical soils in W. Africa (Verbree et al., 2014). PO_4^{3-} adsorbs strongly to soil minerals (Nodvin et al., 1986) which will affect its availability to plants for uptake. Agro-ecosystem ($p < 0.0001$), TSP ($p < 0.0001$), and compost ($p < 0.0001$) had a significant effect on $\text{PO}_4\text{-P}$ concentrations across all agro-ecosystems. Phosphate, measured as $\text{PO}_4\text{-P}$, was highest in the Coastal Savannah, followed by the Transition agro-ecosystem. The Forest and Guinea Savannah had equally low $\text{PO}_4\text{-P}$ concentrations (Fig. 20). The monovalent H_2PO_4^- form of phosphorus is the most soluble in near neutral to slightly acidic soils and most absorbed by plants (Troeh and Thompson, 2005). Finer soil textures such as clays have greater water holding capacity and are, therefore, more likely to contain H_2PO_4^- in solution. Findings in the current study showed that the agro-ecosystems with coarse textured soils (Coastal

Savannah and Transition) had higher concentrations of PO₄-P than agro-ecosystems with finer soil textures. This could likely be due to higher plant uptake of solubilized PO₄-P in the Forest agro-ecosystem, which had both ample annual rainfall and finer soil texture to make P available to plant roots. Meanwhile the agro-ecosystems with coarse soil textures may have retained more PO₄-P because of poor soil water retention and therefore lower plant uptake of PO₄-P.

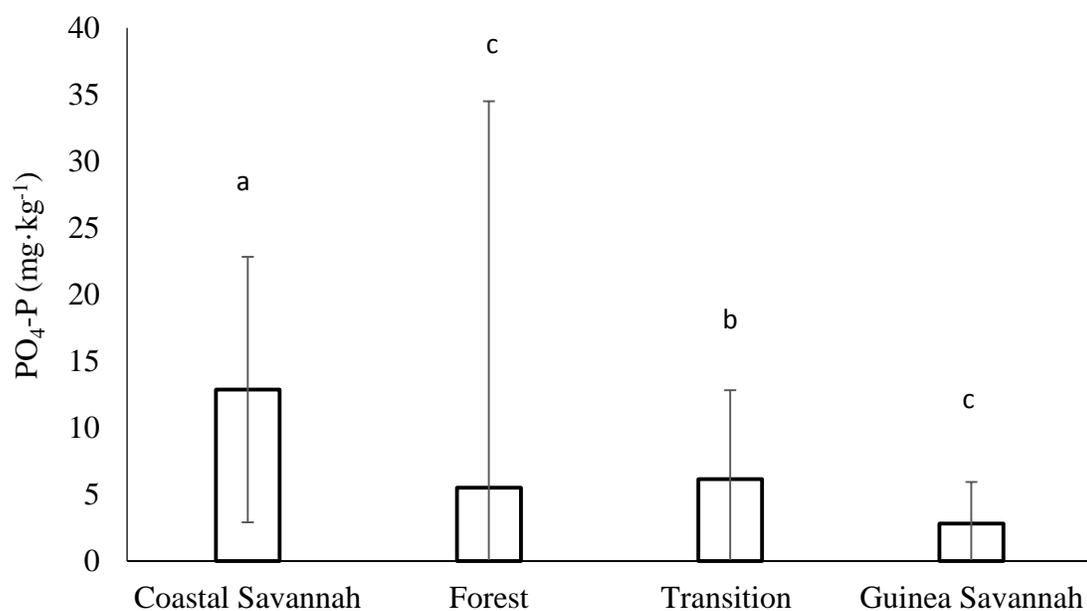


Fig. 20. Mean extractable soil PO₄-P concentrations. Means shown are raw data. Error bars are standard deviation. Differences in lower case letters indicate a significant difference at $\alpha < 0.05$. Duncan's new multiple range test was performed on transformed data.

Temperature and pH also exert an influence on soluble H_2PO_4^- (Troeh and Thompson, 2005), but temperature was relatively similar for all of the agro-ecosystems. Conversely, pH varied among agro-ecosystems and was much lower in the Transition and Guinea Savannah, although this factor did not appear to have an effect on soil $\text{PO}_4\text{-P}$ concentrations (Fig. 20). Therefore, it is likely that rainfall and soil texture led to differences in $\text{PO}_4\text{-P}$ concentrations.

3.4.1.4. Summary of agro-ecosystem results

The Coastal Savannah and Guinea Savannah had the greatest inorganic-N, while the Forest had the greatest EON and EOC concentrations. Inorganic P concentrations were greatest in the Coastal Savannah (Table 11). Although it is useful to understand the soil nutrient status of each agro-ecosystem, the primary interest of this study was in understanding within agro-ecosystem variability because this information is more useful to smallholder farmers.

Table 11. Summary of agro-ecosystems with the highest concentration of each soil nutrient evaluated. Shaded grids indicate agro-ecosystems that had the highest concentration of each soil constituent at an alpha < 0.05

	Coastal Savannah	Forest	Transition	Guinea Savannah
$\text{NO}_3\text{-N}$				
$\text{NH}_4\text{-N}$				
TEN				
EON				
EOC				
$\text{PO}_4\text{-P}$				

Fifteen combinations of soil amendments were used in each of agro-ecosystems evaluated but not all treatment combinations had a significant effect on soil C, N and P concentrations. The following section will concentrate on only those nutrients that were significantly affected by soil amendment treatments within each agro-ecosystem.

3.4.2. Coastal Savannah Treatment Effects

In the Coastal Savannah, soil NO₃-N and PO₄-P concentrations were significantly different among treatments.

3.4.2.1. Coastal Savannah: NO₃-N

Soil extractable NO₃-N had normal distribution after log transformation in the Coastal Savannah. Because the transformed data was normally distributed but the treatment groups did not have equal population variance, the ANOVA assumptions were only partially met. ANOVA revealed that of the fertilizer inputs evaluated, TSP (p = 0.040) and urea (p = 0.013) had a significant effect on concentrations of soil NO₃-N in Coastal Savannah soils. Duncan's means comparison revealed further differences in the concentration of NO₃-N among the fertilizer treatment combinations (Fig. 21).

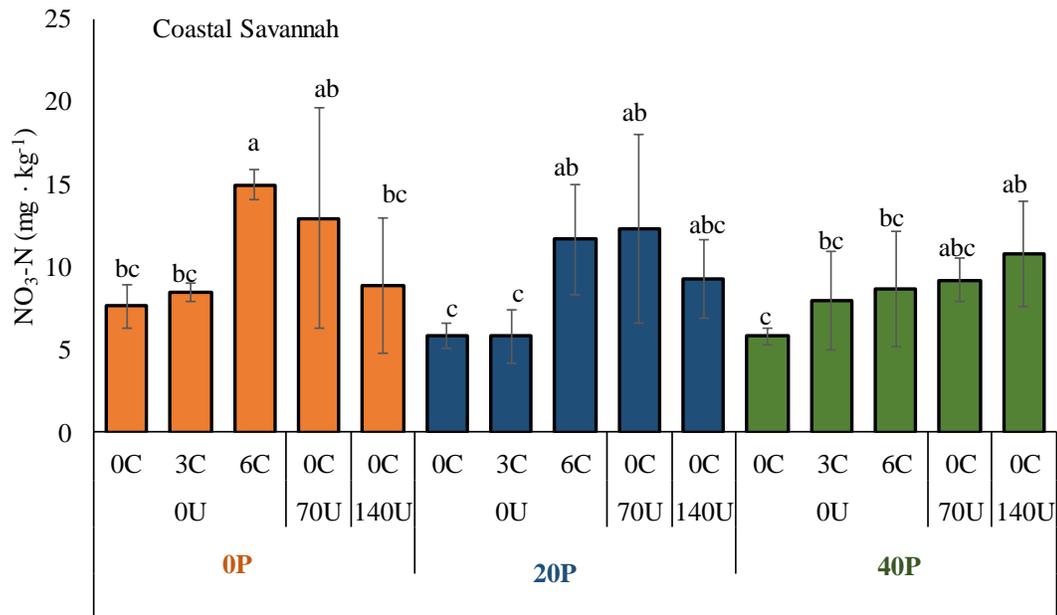


Fig. 21. Mean extractable soil NO₃-N for each treatment in the Coastal Savannah. Error bars are standard deviation. Differences in lower case letters among treatments indicate a significant difference at $\alpha < 0.05$. Duncan's new multiple range test was performed on transformed data.

Significantly higher NO₃-N was extracted from the 0P-0U-6C treatment compared to 8 other treatments (Fig. 21). Five of these 8 low residual NO₃-N treatments had some amount of TSP, whereas the high NO₃-N treatments had no TSP (Table 12). This seems to indicate that TSP may have had an inverse relationship with NO₃-N concentrations. Indeed, a visual analysis of figure 21 suggests that the 5 treatments with 0 kg P ha⁻¹ TSP are slightly higher in NO₃-N than the other treatments, especially when compared to the 40 kg P ha⁻¹ treatments.

Table 12. Fertilizer treatment 0P-0U-6C had higher soil NO₃-N concentrations than 8 other fertilizer treatments.

		0P-0U-0C
		0P-0U-3C
		0P-140-0C
		20P-0U-0C
0P-0U-6C	>	20P-0U-3C
		40P-0U-0C
		40P-0U-3C
		40P-0U-6C

Incorporation of an N source also seems to lead to higher NO₃-N concentrations. Again, visual assessment of NO₃-N concentrations between treatments shows a clear tendency for urea and high compost treatments to have higher soil extractable NO₃-N (Fig. 21). Confirming this visual analysis, Duncan's means comparison provides statistically significant evidence of such a trend. For example, 4 treatments (40P-140U-0C, 20P-0U-6C, 20P-70U-0C, and 0P-70U-0C), all which feature an N or compost source had higher soil NO₃-N than 3 treatments that had no urea additions (20P-0U-0C, 20P-0U-3C, and 40P-0U-0C). Such a tendency suggests that the application of urea may be correlated with higher soil NO₃-N concentrations in the Coastal Savannah. Furthering this notion is the ANOVA result that found urea to have a significant effect on soil extractable NO₃-N. Therefore, based on visual assessment of graphs, Duncan's testing,

and ANOVA, the 0 kg ha⁻¹ TSP + N additions, preferably in the form of urea, seems to correlate with greater soil extractable NO₃-N in the Coastal Savannah.

The availability of the amino functional group from the urea molecule to cleavage by extracellular enzymes would be higher than the release of N from more complex amines in the compost. Halvorson and Reule (1994) found that mineral fertilizer such as ammonium nitrate fertilizer (NH₄NO₃) rates above 90 kg N ha⁻¹ led to increased soil NO₃-N concentrations in the 0 to 180 cm depth zone of a rainfed grain crop rotation on Mollisols in Central Great Plains USA. Availability of NH₄⁺ is one prerequisite to nitrification especially in tilled soils where there is plenty of aeration. Several factors such as soil pH, soil moisture, and availability of biodegradable C can influence microbial utilization of NH₄⁺ possibly resulting in nitrification. Ammonium nitrate fertilizer utilized by Halvorson and Reule (1994) would have dissociated in the soil solution thus supplying a readily available supply of NO₃⁻ for the plant roots and NH₄⁺ for microbial use either as a substrate or, in a low soil C situation as would likely be found in a savannah soil, conversion to NO₃⁻.

Nitrate-N concentrations in the Coastal Savannah soils collected in 2012 ranged from 8.3 to 25.3 mg kg⁻¹ (Davies, 2014) compared to NO₃-N from soils collected in the current study which ranged from 5.8 to 15.0 mg kg⁻¹.

3.4.2.2. Coastal Savannah: Orthophosphate-P

Transformed PO₄-P data showed normal distribution in the Coastal Savannah, but did not have equal population variance. Therefore, ANOVA assumptions were only partially met. Univariate analysis of variance demonstrated that TSP ($p = 0.031$) and compost ($p < 0.0001$) additions had a significant effect on soil extractable PO₄-P concentrations in the Coastal Savannah.

Means comparison with Duncan's new multiple range test revealed differences among the 15 individual fertilizer combinations (Fig. 22). Highest recovery of PO₄-P was observed for the 40P-0U-0C treatment, which was significantly different than 5 of the 15 fertilizer combinations (0P-0U-3C, 0P-140U-0C, 20P-0U-0C, 20P-140U-0C, 40P-70U-0C, and 40P-140U-0C).

Although ANOVA did not detect statistically significant TSP x compost nor TSP x urea interaction effects there does seem to be a relationship between TSP combined with an N-source and residual PO₄-P. Four of the 5 treatments that were lower in PO₄-P concentrations included N fertilizer source, while the high PO₄-P concentration treatment (40P-0U-0C) had no N inputs. Based on both the ANOVA and means separation testing it appears that the combination of TSP with an N source may lead to significantly lower soil extractable PO₄-P in Coastal Savannah soils. Such a conclusion is drawn cautiously as all ANOVA assumptions were not met and there few significant differences among treatments. This could indicated that soils are simply slow to respond to changes in management or it could be a type I error.

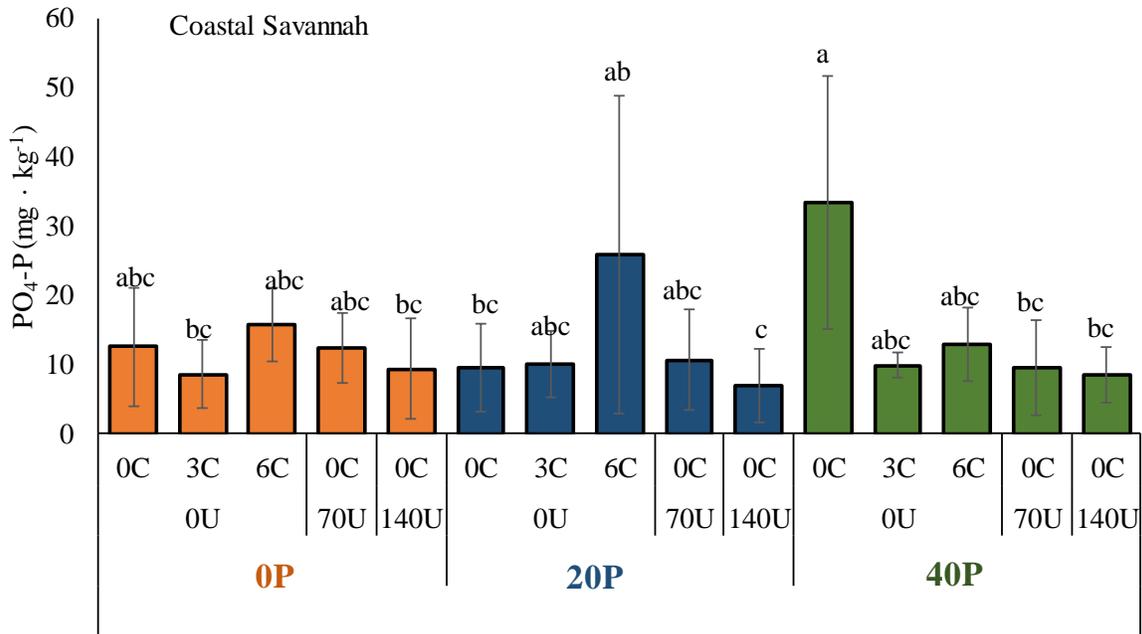


Fig. 22. Mean extractable soil PO₄-P for each treatment in the Coastal Savannah. Error bars are standard deviation. Differences in lower case letters among treatments indicate a significant difference at $\alpha < 0.05$. Duncan's new multiple range test was performed on transformed data.

Extractable PO₄-P ranged from 6.9 to 33.4 mg kg⁻¹ 20P-140U-0C and 40P-0U-0C treatments respectively (Fig. 22). For the soils collected in 2013 PO₄-P ranged from 13.6 to 68.8 mg kg⁻¹ for soils collected in 2012 from the 0P-140U-0C and 40P-0N-3C treatments respectively (Davies, 2014). Pooling treatments for the individual collection years, 27.8 and 13.1 mg kg PO₄-P was extracted for collection years 2012 and 2013 respectively illustrating a drop in extractable PO₄-P when comparing the two years.

Both sets of soils were extracted with a Bray extract and so the effect of treatment combinations and an additional year of treatment may have had an effect on PO₄-P concentrations.

3.4.3. Forest Treatment Effects

Experimental treatments in the Forest zone resulted in few significant findings. DOC was the only soil nutrient affected by fertilizer treatments.

3.4.3.1. Forest: EOC

Univariate analysis of variance found that application of TSP had a significant effect on EOC extracted ($p = 0.05$). However, it should be noted that the EOC data was not normally distributed. Further investigation of differences between individual treatment means showed that just one treatment (40P-0U-3C) had significantly higher EOC concentrations than all other treatments with the exception of 0P-0U-0C, 20P-140U-0C and 40P-0U-6C (Fig 23).

The 4 treatments with high EOC concentrations in the Forest zone did not seem to follow a particular trend. For example, most of these treatment combinations had different rates of TSP fertilizer and one had no TSP, even though ANOVA suggested that TSP had a significant effect on EOC (Fig. 23). Also of note is that several of the treatments with high EOC concentrations had an N source. However this did not seem to follow a trend either. For example, the 20P-140U-0C treatment had significantly more EOC concentrations than the treatments with the exact same rate 140 kg N ha⁻¹ (40P-140U-0C and 0P-140U-0C).

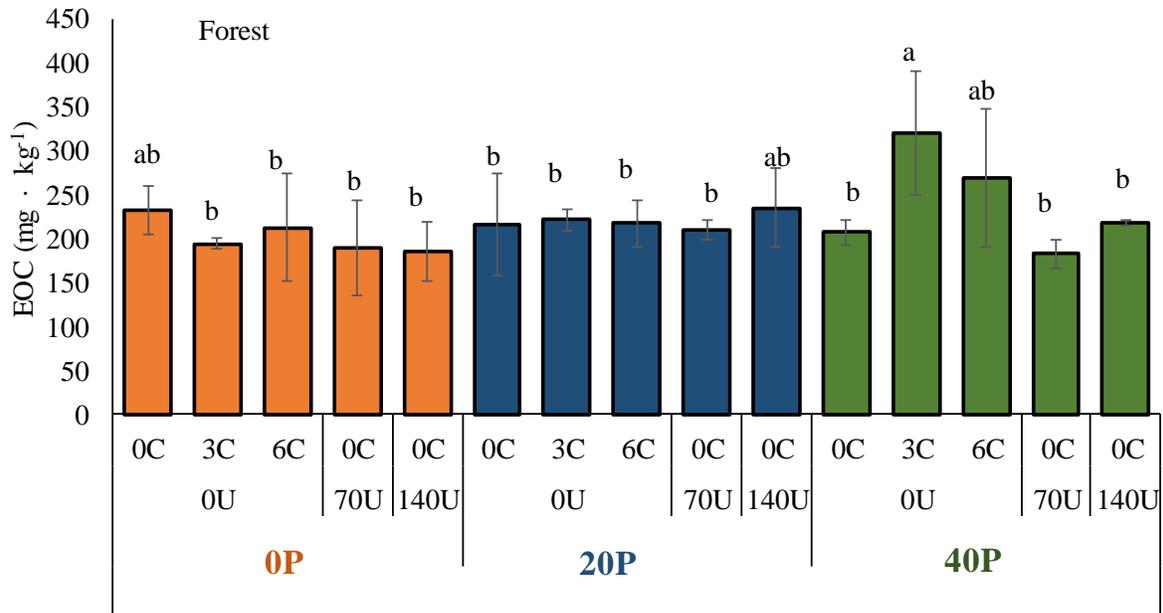


Fig. 23. Mean soil extractable organic carbon (EOC) for each treatment in the Forest. Error bars are standard deviation. Differences in lower case letters among treatments indicate a significant difference at $\alpha < 0.05$. Duncan's new multiple range test was performed on transformed data.

Overall it is difficult to see a clear correlation between individual fertilizer combinations and concentrations of EOC, this is supported the lack of statistically significant differences between most of the treatments. Therefore, based on the lack of normality and the significant differences between treatment means, it is suggested that at year 3 of this fertility trial soils in this zone do not have a clear response to treatments.

Concentrations of EOC ranged from 80.0 mg kg^{-1} to 150 mg kg^{-1} in the 40P-140U-0C and the 40P-0U-3C treatment combinations in 2012 (Davies et al., 2014)

compared to 183 mg kg⁻¹ to 320 mg kg⁻¹ in the 0P-70U-0C and 40P-0U-3C treatment combinations in 2013. In both years addition of urea appeared to depress residual EOC whereas a combination of TSP and compost enhanced residual EOC. The doubling of EOC concentrations between 2012 and the current study is encouraging as EOC is highly correlated with %OC in soils suggesting that % soil C sequestration is responding to soil amendments.

Even though urea, once the amino groups are cleaved, can be quantified as EOC; the functional carboxyl group (CO) is released allowing it to bind with a hydroxyl group (OH) readily available in these high pH forest soils forming COOH which will readily form carboxylic acids and esters. These smaller C molecules derived from urea may be more labile and act as a primer for other EOC mineralization by soil microbes resulting in lower residual EOC with urea addition.

The traditional method of clearing forests for agriculture in West Africa was slash and burn which would lead to relatively stable C in the form of charcoal in the soil (Verbree et al., 2014). In neighboring southern Mali, Verbree et al. (2014) reported high incidences of charcoal in some of the soil they analyzed which can be an issue because it can elevate soil Zn (Scholes and Andreae, 2000; Mishra and Chaudhury, 1994; Namgay et al., 2010). Elevated metals in soil solution such as Zn³⁺, Mn³⁺ and Fe³⁺ would attract the generally negative DOC molecule resulting in stable organo-metal complexes and perhaps more stable and unrecoverable EOC. The major benefit of trees with agriculture was described by Verbree et al. (2014) where the roots bring deep micronutrients such as

Zn³⁺ and Fe³⁺ to the surface soil when plant leaf litter falls to the ground releasing these metals.

Above- and below-ground litter in the form of leaf litter, root exudate, and root death and decay can have a significant effect on DOC concentrations in soil solution (Aitkenhead-Peterson et al., 2003). Root exudate, thought to be wholly biodegradable is significantly affected by N addition in Norway spruce (Aitkenhead-Peterson and Kalbitz, 2005) where zero (0 mg L⁻¹) or high (100 mg L⁻¹) N in soil solution can have a negative effect on soil microbial function resulting in larger DOC molecules as measured by the humification index (Zsolnay et al., 1999; Kalbitz and Geyer, 2001). Not surprising, in the current study EOC was significantly higher in the Forest agro-ecosystem but most interesting was why some applications of fertilizer promoted higher EOC concentrations than others.

3.4.4. Transition Treatment Effects

In the Transition zone, fertilizer treatment had a significant effect on three of the soil nutrients quantified: NO₃-N, EOC, and PO₄-P. Data were normally distributed for all three soil nutrients.

3.4.4.1. Transition: $\text{NO}_3\text{-N}$

Univariate analysis of variance showed that application of urea had a significant effect on $\text{NO}_3\text{-N}$ in the Transition zone ($p = 0.03$). Means comparison with Duncan's New Multiple Range test revealed that only the 40P-140U-0C treatment had significantly higher $\text{NO}_3\text{-N}$ concentrations when compared to the than the 0P-0U-3C treatment (Fig. 24). The main differences between these two fertilizer combinations is that one has a TSP plus urea fertilizer combination, whereas the other has only compost. Evaluation of the treatment differences the 40P-140U-0C treatment was similar to all other treatments except for 0P-0U-3C, including those treatments with no urea such as the control, 0P-0U-6C, 40P-0U-6C, and others (Fig. 24). Therefore it does not seem reasonable to conclude that the addition of urea was consistently associated with higher $\text{NO}_3\text{-N}$ concentrations. Nitrate-N concentrations ranged from 2.3 mg kg^{-1} to 5.8 mg kg^{-1} in the 2012 soil collections (Davies, 2014) compared to a range of 2.6 mg kg^{-1} to 4.1 mg kg^{-1} for the current study.

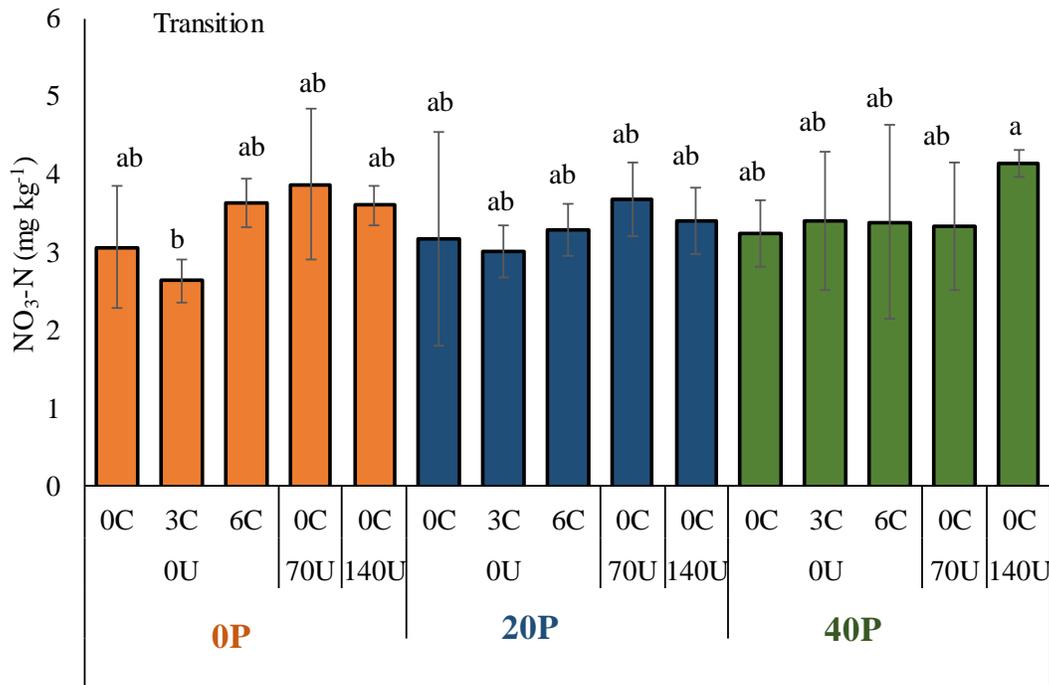


Fig. 24. Mean extractable soil NO₃-N for each treatment in the Transition. Error bars are standard deviation. Differences in lower case letters among treatments indicate a significant difference at $\alpha < 0.05$. Duncan's new multiple range test was performed on transformed data

3.4.4.2. Transition: EOC

Univariate analysis of variance revealed that compost addition had a significant effect on EOC in the Transition zone ($p = 0.001$). The 20P-0U-6C treatment combination was significantly higher than the 0P-0U-0C treatment (Fig. 25) but EOC concentration were not significantly different for the other treatment combinations (Fig. 25).

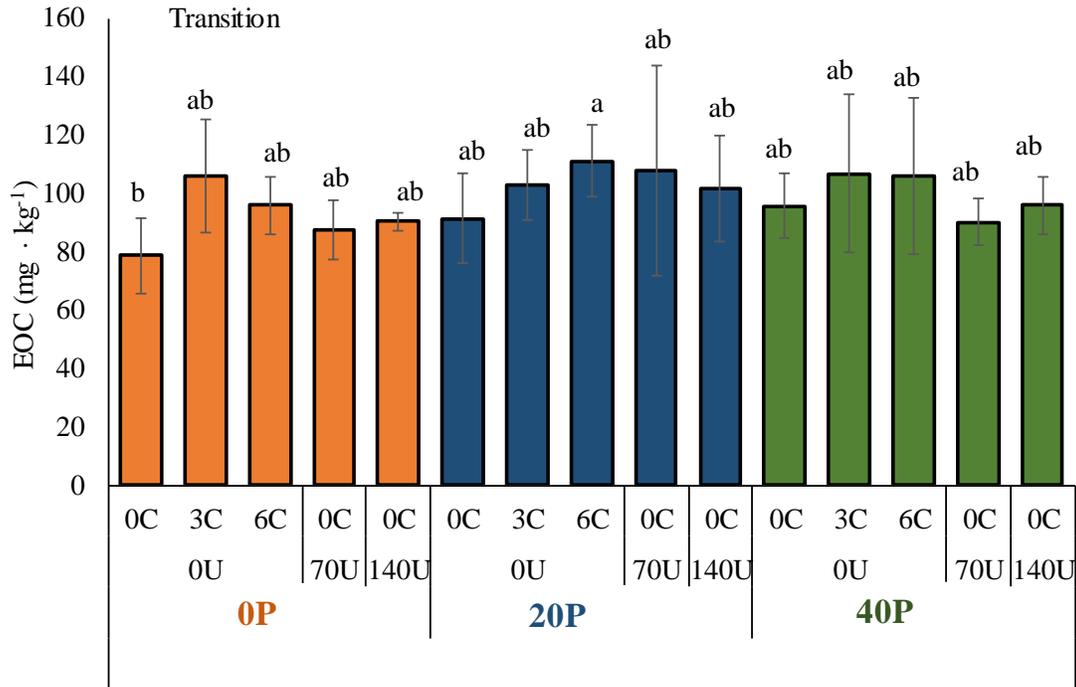


Fig. 25. Mean soil extractable organic carbon (EOC) for each treatment in the Transition. Error bars are standard deviation. Differences in lower case letters among treatments indicate a significant difference at $\alpha < 0.05$. Duncan's new multiple range test was performed on transformed data.

EOC concentrations ranged from 51 mg kg⁻¹ to 82 mg kg⁻¹ in the 2012 soil collections (Davies, 2014) compared to 88 to 111 mg kg⁻¹ in the current study. Recoverable EOC concentrations in the Transition agro-ecosystem show an increase in 2013 collected relative to those collected in 2012 indicating increased soil C sequestration but the increase between the two years was not as great in the Transition zone.

3.4.4.3. Transition: PO₄-P

Univariate analysis of variance showed that TSP ($p = 0.02$) was significantly affected PO₄-P concentrations in the Transition agro-ecosystem soils. Means separation analysis revealed that treatment 40P-0U-3C had significantly higher PO₄-P than of the 6 treatment combinations (Table 13). Three of those 6 treatments have no TSP include: 0P-0U-0C, 0P-70U-0C and 0P-140U-0C (Fig. 26).

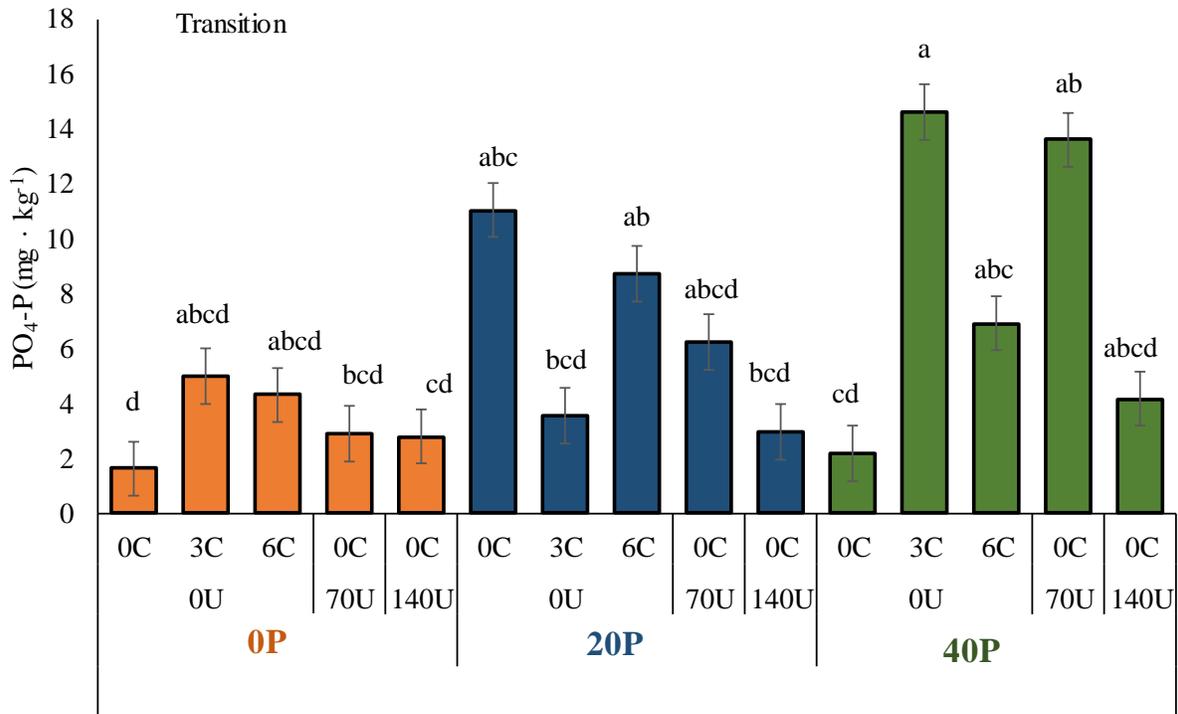


Fig. 26 Mean extractable soil PO₄-P for each treatment in the Transition. Error bars are standard deviation. Differences in lower case letters among treatments indicate a significant difference at $\alpha < 0.05$. Duncan's new multiple range test was performed on transformed data.

Table 13. Fertilizer treatment 40P-0U-3C had higher soil PO₄-P concentrations than 8 other fertilizer treatment combinations.

		0P-0U-0C
		0P-70U-0C
		0P-140U-0C
40P-0U-3C	>	20P-0U-3C
		20P-140U-0C
		40P-0U-0C

Concentrations of PO₄-P in the Transition agro-ecosystem ranged from 1.6 mg kg⁻¹ to 14.6 mg kg⁻¹ in the 0P-0U-0C and the 40P-0U-3C respectively compared to a range of 4.5 mg kg⁻¹ to 58.8 mg kg⁻¹ in the 0P-140U-0C and 40P-0U-0C treatment combinations respectively from the 2012 collected soils (Davies, 2014). It is evident that addition of TSP will increase recoverable PO₄-P from soils but the large decline in residual PO₄-P between the two years in the Transition agro-ecosystem is an enigma. PO₄-P concentrations in soils also declined between the two years in the Coastal Savannah agro-ecosystem.

3.4.5. Guinea Savannah Treatment Effects

Univariate analysis of variance showed that three of the soil nutrients evaluated had significant differences in concentrations as a result of fertilizer inputs in the Guinea Savannah: NO₃-N, EOC and PO₄-P.

3.4.5.1. Guinea Savannah: $\text{NO}_3\text{-N}$

$\text{NO}_3\text{-N}$ concentrations in the Guinea Savannah were significantly affected by applications of TSP ($p = 0.04$) and urea ($p = 0.01$). There were no interaction effects of TSP, urea or compost on recoverable $\text{NO}_3\text{-N}$ concentrations.

Log transformed $\text{NO}_3\text{-N}$ data in the Guinea Savannah did not have normal distribution based on Shapiro-Wilks ($p = 0.02$). Additionally there was not equal population variance. Therefore conclusions from the univariate analysis of variance for $\text{NO}_3\text{-N}$ should be considered with caution. Means separation tests indicated that the 0P-0U-6C treatment had significantly higher $\text{NO}_3\text{-N}$ concentration than 3 other treatment combinations: the 0P-0U-0C, 40P-0U-3C, and 40P-0U-0C (Fig. 27). No significant differences in other treatment combinations were observed.

Concentrations of $\text{NO}_3\text{-N}$ in soil collected in 2012 from the Guinea Savannah ranged from 8.0 to 23.0 mg kg^{-1} in the 0P-0U-0C and 0P-140U-0C treatment combinations respectively. In the current study concentrations of $\text{NO}_3\text{-N}$ ranged from 3.0 to 4.1 mg kg^{-1} in the 20P-0U-3C and 40P-140P-0C treatments respectively. This high reduction in residual $\text{NO}_3\text{-N}$ in the Guinea Savannah was not reflected in the Coastal Savannah or Transition agro-ecosystems where $\text{NO}_3\text{-N}$ concentrations between the two years were comparable. NO_3^- is a very mobile ion and does not adsorb to soil minerals (Nodvin et al., 1986) nor is it generally recognized as an ion for microbial uptake, its fate being plant uptake or loss through leaching or runoff. Differences in annual rainfall between the two years might suggest either loss through leaching but may also indicate

greater plant uptake and thus lower concentrations in the soil. More likely though the reason lies with doubling of EOC between the two years, higher DOC concentrations in a soil sometimes results in lower NO₃-N concentrations because when labile DOC is available soil microbes will utilize available NH₄-N rendering it less available for nitrification. The inverse but non-significant correlation ($R = 0.03$) between NO₃-N and EOC in the Guinea Savannah may support this assumption.

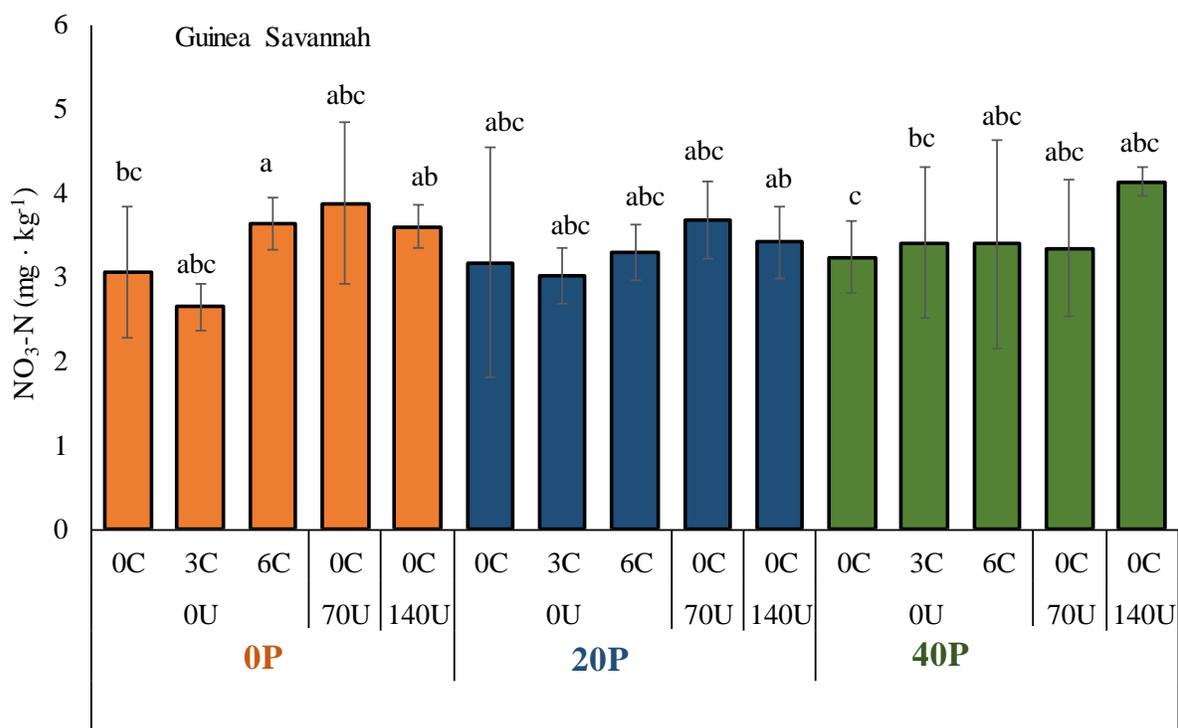


Fig. 27. Mean extractable soil NO₃-N for each treatment in the Guinea Savannah. Error bars are standard deviation. Differences in lower case letters among treatments indicate a significant difference at $\alpha < 0.05$. Duncan's new multiple range test was performed on transformed data.

3.4.5.2. *Guinea Savannah: EOC*

Application of compost had a significant effect on EOC concentrations in the Guinea Savannah soils ($p = 0.01$) but application of TSP, urea or interactions of fertilizer treatments had no significant effect on EOC concentrations. There was normal distribution but unequal population variance for the EOC data. Duncan's means separation test did not show any fertilizer treatment combinations to be significantly different (data not shown).

Concentrations of EOC in soils collected in 2012 from the Guinea Savannah ranged from 40.0 to 77.0 mg kg⁻¹ in the 0P-0U-0C and the 0P-0U-6C treatment combinations respectively (Davies, 2014). These EOC concentrations were much lower than in soils of the current study which ranged from 106 to 144 mg kg⁻¹ in the 40P-0U-0C and the 20P-0N-3C treatment combinations respectively. As found with the Forest and Transition agro-ecosystem EOC, soils collected and processed in 2013 had higher residual EOC when compared to soils collected and processed in 2012 indicating an increase in organic matter.

3.4.5.3. *Guinea Savannah: PO₄-P*

Univariate analysis of variance showed that TSP ($p = 0.03$) and compost ($p < 0.001$) had a significant effect on PO₄-P concentrations in soils of the Guinea Savannah. Addition of urea or fertilizer interactions had no significant effect on PO₄-P. Log transformed PO₄-P data was normally distributed using the Shapiro-Wilks test ($p = 0.14$), but did not have equal population variance. Duncan's New Multiple Range

separation Test indicated that the 40P-0U-3C treatment had significantly higher PO₄-P concentrations than seven of the other treatments (Fig. 28).

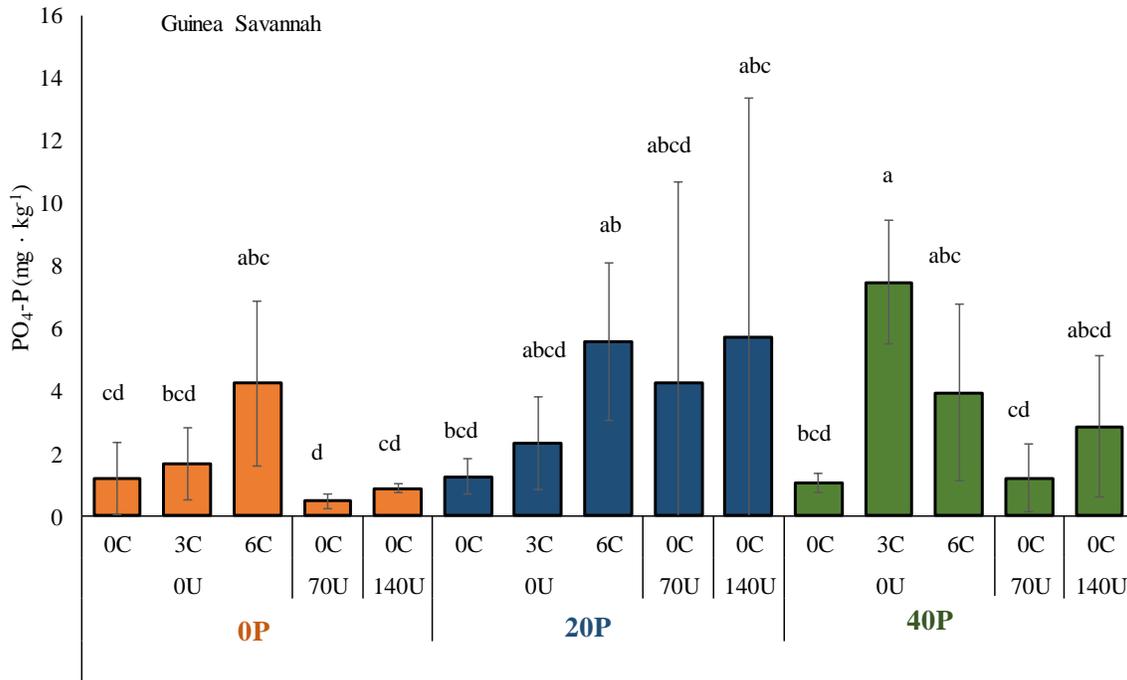


Fig. 28. Mean extractable soil PO₄-P for each treatment in the Guinea Savannah. Error bars are standard deviation. Differences in lower case letters among treatments indicate a significant difference at $\alpha < 0.05$. Duncan's new multiple range test was performed on transformed data.

It is interesting to note that the 40P-0U-3C treatment that is significantly high in PO₄-P here, is the same fertilizer combination that had significantly higher PO₄-P concentrations in the Transition agro-ecosystem. Of the 7 fertilizer combinations that were significantly lower in PO₄-P concentrations, four had zero TSP which supports the

univariate analysis of variance finding that TSP had a significant effect on PO₄-P concentration but two of the lower PO₄-P soil concentrations had TSP applied at the 40 kg P ha⁻¹ rate (Table 14).

Table 14. Fertilizer treatment 40P-0U-3C had higher soil PO₄-P concentrations than 7 other fertilizer combinations.

		0P-0U-0C
		0P-0U-3C
		0P-70U-0C
40P-0U-3C	>	0P-140U-0C
		20P-0U-0C
		40P-0U-0C
		40P-70U-0C

Concentrations of PO₄-P in the Guinea Savannah ranged from 0.5 to 7.5 mg kg⁻¹ in the 0P-0U-0C and the 40P-0U-3C treatments respectively in the current study compared to 1.6 to 58.9 mg kg⁻¹ in the 0P-70U-0C and the 40P-0U-0C treatment combinations in soils collected in 2012 (Davies, 2014). Average pooled PO₄-P concentrations for 2012 were 7.9 mg kg⁻¹ compared to average pooled PO₄-P concentrations for 2013 which were 2.9 mg kg⁻¹. A similar decline in recoverable PO₄-P was also evident in the Coastal Savannah and Transition agro-ecosystems when comparing the two years.

3.5. CONCLUSION

Combinations of fertilizers had a significant effect on the concentrations of some nutrients in some of the Ghanaian agro-ecosystems but not others. It is important

therefore to identify which combination of fertilizer works best for each specific nutrient concentration. Soil testing is important to determine which nutrients are needed for agriculture in a broad sense (NPK) and knowledge of fertilizer combinations can help to increase specific N and P needs.

The most important finding is that a ‘one-size-fits-all’ approach to fertilization in the Ghanaian agro-ecosystems will not produce the same results with respect to increasing soil fertility. Differences in environmental factors such as annual rainfall and soil parent material and texture will affect residual soil nutrient concentrations response to fertilizer combinations. Soil scientists have advocated for decades the nutrient requirements based on soil type, cropping system, farm size and the availability of essential inputs (Lal, 1987).

4. COMPARISON OF WATER AND 0.1 M HCL EXTRACTS ON SOIL NUTRIENT EXTRACTION

4.1. INTRODUCTION

What fraction, and the appropriate ecological amount of C, N and P that is extracted from whole soil continues to be elusive for many soil scientists (Haney et al., 2006; Jones and Willett, 2006; Carrillo-Gonzalez et al., 2013). This is largely due to the question of whether the extracted C and N concentrations reflect plant available N, microbial available C and N or simply, just how much C and N is leachable from soil. Methods for the extraction of P are fairly well defined and will have minimal discussion in this chapter.

4.1.1. Ecological Use of Carbon and Nitrogen

4.1.1.1. Biotic fate of carbon

Dissolved organic carbon (DOC) or water extractable organic carbon (WEDOC) is recognized as an important substrate for both soil and surface water microbes for the immobilization of inorganic nitrogen and phosphorus (Yano et al., 1998, 2000; Kalbitz et al., 2003; McDowell et al., 2006; Cioce and Aitkenhead-Peterson, 2015). The biodegradability of this substrate carbon is frequently termed labile DOC (LDOC) or biodegradable DOC (BDOC) (Kalbitz et al., 2003; McDowell et al., 2006; Cioce and Aitkenhead-Peterson, 2015) and is most often reported as a percent of the DOC in the water sample retrieved (i.e. stream water or soil solution) or the water extracted solution

(i.e. plant material or soil). The percent biodegradable carbon tends to decrease with its travel through an ecosystem (Cioce and Aitkenhead-Peterson, 2012). For example, Cioce and Aitkenhead-Peterson (2012) examined biodegradability of various plant material, soils and surface waters in an urban ecosystem and reported ranges of water extractable BDOC between $75\pm 4\%$ for turf grass and $34\pm 3\%$ for forest leaf litter, $47\pm 1\%$ to $19\pm 3\%$ for water extracted soil under turf grass and woody species, respectively, and up to 10% biodegradability in surface waters. Cioce and Aitkenhead-Peterson's (2012) findings illustrated the decline in percent BDOC as ecosystem water moved through an urban ecosystem.

4.1.1.2. Abiotic fate of carbon

Carbon is primarily leached from vegetation and soils by precipitation, irrigation and throughfall (interaction with plant canopy). The pH and the chemistry of these water vectors varies at local, regional and global scales (Aitkenhead-Peterson et al., 2003; Pannkuk et al., 2011; Steele and Aitkenhead-Peterson, 2013). Furthermore, the differences in pH, ionic strength, and chemistry of precipitation, irrigation and throughfall water can influence the mass of leached carbon (Pannkuk et al., 2011; Steele and Aitkenhead-Peterson, 2013) and on soil microbial community composition (Holgate et al., 2011). Different pH and solute chemistries may also have an effect on the molecular weight distribution of DOC in soil A and B horizons (Aitkenhead-Peterson et al., 2003).

This natural DOC derived from leaching of plant and soil by solutes (rainwater and irrigation water) comprising very different chemistries (Steele and Aitkenhead-Peterson, 2013) is then used as a substrate as previously described (Section 4.1.1), adsorbed to soil minerals, leached to groundwater, or runs-off to surface waters. Adsorption of DOC to soil minerals has received a lot of attention over the last three decades in forested and urban ecosystems (Nodvin et al., 1986; Kaiser and Zech, 1997, 2000; Kaiser and Kalbitz, 2012; Aitkenhead-Peterson and Cioce, 2013). The use of the initial mass isotherm approach to DOC adsorption is particularly recommended for quantifying DOC adsorption (Nodvin et al., 1986). The mechanisms controlling the adsorption of DOC to soil minerals are for the most part speculative and include anion exchange, ligand exchange, water and cation bridging, hydrophobic interactions, van der Waals forces, dipole-dipole interactions, π - π bonds between aromatic or highly conjugated groups and hydrogen bonding of non-ionic compounds (Parfitt et al., 1977; Tipping, 1981; Jardine et al., 1989; Jekel, 1991; Pignatello, 1993; Gu et al., 1995). Soil solution DOC bound by ligand exchange is most likely to be protected against microbial metabolism and carbon bound by ion exchange is the most likely to be re-released (Aitkenhead-Peterson et al., 2003).

Several studies have examined the soil attributes likely responsible for DOC adsorption. For example, Jardine et al. (1989), David et al., (1989) and Kennedy et al. (1996) all suggested that soil pH was responsible for DOC adsorption to their forest soils with maximum adsorption occurring at an equilibrium solution pH of 4.5 (Jardine et al.

1989). A hyperbolic function is displayed in the relationship between DOC adsorption and the pH of equilibrium solution in the Jardine et al. (1989) study suggesting that with an equilibrium solution of pH 2 DOC, adsorption would be similar to that of an equilibrium solution of pH 5 to 6. Higher pH equilibrium solutions tended to impede the adsorption of DOC (Jardine et al., 1989). Kennedy et al. (1996) reported that at input pH of 3-4.5, DOC was weakly retained in Bhs horizon of a spodosol but strongly retained in its Bs and Cx horizons.

Generally DOC is attracted the positive charge of iron and aluminum and manganese oxides (McDowell and Wood, 1984; Jardine et al., 1989; Dalva and Moore, 1991; Qualls, 2000). But its adsorption can also be determined by the percent clay content of a soil (Nelson et al., 1990, 1993; Kaiser and Zech, 1998) and the amount of indigenous carbon already in the soil (Jardine et al., 1989; Kaiser et al., 2000).

4.1.1.3 Importance of C as DOC in soil

Carbon in soil is a continuum of molecule sizes with the DOC fraction operationally defined as that material which is smaller than 0.45 μm (Thurman, 1985). Many researchers consider DOC to be the fraction $< 0.7 \mu\text{m}$ (Aitkenhead-Peterson et al., 2003). Fellman et al. (2012) reported no significant difference in DOC concentrations filtered through 0.45 μm compared to 0.7 μm filters. Others opt for a much small pore size of $< 1 \mu\text{m}$ (Chow et al., 2005) depending on the Dalton weight fraction they are interested in. DOC in soil reflects the balance of productivity by the plants and mineralization by the microbes. Its adsorption to soil minerals tends to protect it from

further microbial decay (Marschner et al., 2008) and its presence will improve soil texture which will enhance soil water holding capacity.

4.1.1.4. Organic and inorganic N

Of the nitrogen species, it is generally accepted that microbes tend to utilize $\text{NH}_4\text{-N}$ and compete with plants for this resource. Use of, or microbial transformations of inorganic-N tends to increase with an increased supply of biodegradable DOC. It is not fully known whether soil microbes can utilize dissolved organic nitrogen (DON) or water extractable dissolved organic nitrogen (WEDON). Gregorich et al. (2003) examined biodegradability of DON in maize-cropped soils and reported that proportionally greater amounts of organic-N were metabolized relative to organic-C. Approximately 80% of water extractable DON was biodegradable in an agricultural fertilized soil with a maize-soy rotation and 60% was biodegradable in a manured maize monoculture (Gregorich et al., 2003)

Plants tend to utilize NO_3^- because it is mobile in the soil and synergistically promotes the uptake of K^+ , Ca^{2+} and Mg^{2+} . There is higher plant uptake efficiency with NO_3^- compared to NH_4^+ ; Legaz et al. (1996) reported that when N^{15} labeled NO_3^- and NH_4^+ was applied to citrus trees in sandy soil, fertilizer in the form of potassium nitrate had a plant uptake rate of 60% compared to 40% uptake for fertilizer added as ammonium sulfate. Nitrate is readily available to plants in aerated soils but in anaerobic soils NH_4^+ is more available to plants than NO_3^- . Organic nitrogen, most commonly in the form of urea ($(\text{NH}_2)_2\text{CO}$) is also applied to crops; but the urea needs to be converted

to ammonium by urease enzymes prior to plant uptake. DON is known to be preferentially taken up by certain plant species, particularly those in arctic tundra and boreal ecosystems (Kielland, 1994; Kielland et al., 2006, 2007; Lipson and Monson, 1998; Lipson et al., 1999, 2001). Plant uptake of organic N molecules has been studied for several years (Read, 1991; Lipson & Näsholm, 2001; Näsholm & Persson, 2001; Neff et al., 2003; Schimel & Bennett, 2004; Rentsch et al., 2007). In spite of the numerous studies reporting the capacities of trees and crop plants to absorb organic N compounds directly through roots, the issue is still a matter of intense debate. Many of these studies have used the amino acid glycine ($\text{NH}_2\text{CH}_2\text{COOH}$) and even with labeled N^{15} glycine there is a huge potential for the amino group to be cleaved enzymatically from the molecule allowing the amino group to be taken up rather than the whole amino acid. The ecological function of DON has remained elusive (McDowell 2003) but it is likely to be a measure of free enzymes and microbes, specifically bacteria in soil solution.

4.1.2. Extraction Methods for the Quantification of Soil Organic C and N

Organic carbon and nitrogen has been extracted from soil using many methods (Dou et al., 2007, 2008a, b; Jones and Willett, 2006; Suominen et al., 2003; Wright et al., 2007; Zsolnay and Gorlitz, 1994; Carrillo-Gonzalez et al. 2013). Chantigny (2003) concluded after examining a large dataset of dissolved organic matter (DOM) collected from ecosystems and processed using various sieving and extraction methods that it was difficult to make generalizations in terms of changes in land use and management

practices because the methodologies used to measure extractable organic matter were not comparable to each other. Indeed, Carrillo-Gonzalez et al. (2013) suggested that extract type had a larger effect on DOC, DON and dissolved inorganic nitrogen (DIN) concentrations than did tillage and cropping practices.

Methods used for quantifying extractable organic carbon (EOC) have included different 1) drying methods: air-dried or field moist, 2) shaking times (minutes to hours), 3) temperature, 4) soil:extractant ratios and 5) extractants. It is generally accepted that extracting field moist soil is best for EOC analysis (Jones and Willett 2006) because air-drying will result in a large flush of organic C and oftentimes N. Shaking times for extracting organic matter from soils have varied from minutes up 24 h (Carrillo-Gonzalez et al., 2013; Aitkenhead-Peterson et al., 2012). Jones and Willett (2006) found that after 6 h of shaking no more OC was extracted and recommended a shaking time of 1 hr. Temperatures while extracting OC have varied from 2° C to 20° C using the argument that at higher extraction temperatures, soil microbes will mineralize the OC. There have also been a large range of soil:extract ratios used (Jones and Willett 2006). Extractants used have included 4 mM CaSO₄ (Zsolnay and Gorriz, 1994; Carrillo-Gonzalez et al., 2013), 0.5 M K₂SO₄ (DeLuca and Keeney, 1994; Rochette and Gregorich, 1998, Gregorich et al., 1998; Jones et al., 2008; Carrillo-Gonzalez et al., 2013), 2M KI (Jones and Willett, 2006; Carrillo-Gonzalez et al., 2013), 1 M NaOH (Jones and Willett, 2006), 1 M HCl (Jones and Willett, 2006) and cold double distilled water or hot double distilled water (DDW) (Gregorich et al., 2003; Jones and Willett,

2006; Steele and Aitkenhead-Peterson, 2012; Carrillo-Gonzalez et al., 2013). Jones and Willett (2006) concluded after utilizing cold DDW, 2M KCl, 0.5 M K₂SO₄, 1 M NaOH or 1 M HCl as extractants with different extraction times (5 min – 24 h), extraction temperatures (2 and 20° C) and soil:extract ratios (1:1 to 1:100) that a standard procedure for the extraction of organic C and N should be proposed based on the very different recoveries of DOC and DON they observed. They proposed using either cold DDW, 2M KCl or 0.5 M K₂SO₄ at a 1:5 soil:extract ratio shaken for 1 h at 20° C on field moist, un-sieved soils. However, they also stressed that their different soils (Cambisol, Podzol, Gleysol) reacted very differently to the five extractants they used and that perhaps standardizing a method for extracting organic C and N would not be realistic.

The majority of studies examining extractable organic C and N have been for forest soils (Aitkenhead-Peterson et al., 2006; Jones and Willett, 2006) or soils under pasture (Jones and Willett, 2006) with relatively few studies examining extractable organic C and N in agricultural crop soils (Gregorich et al., 2003; Dou et al. 2007, 2008 a,b; Wright et al. 2007; Carrillo-Gonzalez et al. 2013; Davies et al., 2014). Carrillo-Gonzalez et al. (2013) used the same soil (fine silty, mixed, thermic Fluventic Ustochrept; pH 7.5-7.7) which was under different tillage and cropping treatments in which to extract organic C and N and inorganic-N. Unlike Jones and Willett (2006) they kept their soil processing, soil:extractant ratio, extracting temperature and time constant. They used field moist soil which was extracted within 24 hours of collection with a 1:5 soil:extract ratio shaken for 15 mins at 200 RPM at 20° C. Extracts used in the Carrillo-

Gonzalez et al. (2013) study were cold DDW, hot DDW, 10 mM CaCl₂, 2M KCl and 0.5 M K₂SO₄. Even though the soil used was the same soil series for their extraction experiment they found that land management practice (tillage and cropping) as well as extractant type had a significant effect on the concentration of organic C. Overall they reported that K₂SO₄ extracted the highest concentrations of organic C and either KCl, cold water or CaCl₂ the lowest, depending on cropping and tillage treatment. Gregorich et al. (2003) used hot and cold water DDW for the examination of soils under monoculture and dual crop rotation soils with and without either mineral fertilizer or manure addition. Davies et al. (2014) and Aitkenhead-Peterson et al. (2015) used 0.1 M HCl considering it to be a weak acid extract reflecting soil solution conditions in the Ghanaian soil they were examining.

4.1.3. Extraction Methods for the Quantification of Soil Inorganic-N

The most recognized method for extracting plant available NO₃⁻ is 2M KCl however this method presents problems if using ion chromatography to quantify NO₃⁻ because it is difficult to separate the Cl⁻ and NO₃⁻ peaks. Furthermore the 2M KCl extractant may produce a high peak of carbonate/bicarbonate just behind the nitrate peak for neutral to alkaline soils which will also affect separation of the nitrate peak. Carrillo-Gonzalez et al. (2013) examined cold and hot DDW, 10 mM CaCl₂, 2M KCl and 0.5 M K₂SO₄ for the extraction of NO₃-N from a soil comprising 45% silt, 43% clay and 12% sand and used colorimetric analysis (cadmium reduction) to quantify NO₃-N. They reported that under no-till conditions, cold water extracted significantly more NO₃-N

from soil when compared to 2M KCl but under conventional tillage there was no significant difference in the concentration of NO₃-N extracted among the 5 extractants used.

Haney et al. (2006) developed a new method for the extraction of soil P that could also simultaneously extract soil NO₃-N, NO₂-N and NH₄-N. The new extractant was named H³A (Haney, Haney, Hossner, Arnold) and was a combination of lithium citrate, citric acid, malic acid, oxalic acid, EDTA and DTPA. The extractant was fashioned to simulate root exudates. They tested the extract against commonly used extracts such as Olsen and Mehlich 3 for soil P and DDW and 1 M KCl for soil NO₃-N and NH₄-N. The recognition by Haney et al. (2006) that extracts used on soil should mimic conditions expected within the soil is a move away from the traditional standard methods generally used to extract inorganic-N.

4.2. OBJECTIVES

The objective of this study was to examine soils from four agro-ecosystems in Ghana under multiple tillage, cropping and soil amendment studies that were extracted using two extracts 1) cold ultrapure water and 2) 0.1 M HCl to determine if there was a significant difference in extraction of EOC, EON, NO₃-N and NH₄-N concentrations between the two extraction types.

4.3. METHODOLOGY

4.3.1. Site Descriptions

The agro-ecosystems used in this experiment were 1) Coastal Savannah, 2) Forest, 3) Forest-Guinea Savannah transitional region (Transition) and 4) Guinea Savannah. Soil in the coastal savannah agro-ecosystem was classified using the World Reference Base for soil resources (WRB, 2006) as a Haplic Lixisol formed on granite with a loamy-sand texture to 60 cm. Soil in the forest agro-ecosystem was classified as a Leptic Lixisol formed on phyllite with a silty-loam texture to 60 cm. Soil in the forest-Guinea Savannah transition agro-ecosystem was classified as Leptic Lixisol formed on sandstone with a loamy sand texture to 30 cm. Finally, soil in the Guinea Savannah agro-ecosystem was classified as a Pisolithic Plinthosol formed on shalestone with a silty loam texture to 60 cm (Davies et al., 2014).

Historically, mean annual rainfall differ between the four agro-ecosystems. In the coastal savannah, mean annual rainfall is 810 mm, in the forest agro-ecosystem, mean annual rainfall is 1500 mm. Mean annual rainfall in the forest-Guinea Savannah transition agro-ecosystem is 1300 mm. Lastly, the Guinea Savannah has a mean annual rainfall of 1100 mm. Apart from the differences in the mean annual rainfall, the distribution of precipitation across the four agro-ecosystems also differs. Each year there is a major (March-July) and minor (August- November) cropping season in the coastal savannah, forest and forest-Guinea savanna agro-ecosystem which is driven by the bimodal distribution of precipitation. In the Guinea Savannah, only one annual cropping

season occurs (August-November) due to the majority of precipitation falling between August and November.

4.3.2. Treatments

Three methods of tillage and four types of cropping systems were laid out in a split-plot design with three replications. The tillage types investigated were traditional till, zonal till, and no-till (Table 15). Traditional tillage (TT) corresponded with the traditional method of seed bed preparation of each region. For the Coastal Savannah, Transition, and Guinea Savannah that meant hoeing to approximately 10 cm deep, whereas in the Forest slash and burn with no mechanical disturbance was the traditional seed bed preparation. Zonal tillage (ZT) was defined as hoeing only in the row to be planted, and no-till (NT) was the absence of any form of mechanical disturbance, with the exception of the hole in which the seed is planted.

Table 15. Factor treatments in the Coastal Savannah, Forest, and Transition agro-ecosystems

Main Plot Factors	Sub-Plot Factors
No-Till (NT)	Maize (M)
Zonal Till (ZT)	Maize-cowpea (MC)
Traditional Till (TT)	Maize-mucuna (MM)
	Maize-Cowpea-mucuna (MCM)

The cropping systems were continuous maize (M), maize-cowpea rotation (MC), maize-mucuna rotation (MM), and a maize-cowpea-mucuna relay (MCM) for the Coastal Savannah, Forest, and Transition zones. In the northern Guinea Savannah zone, due to the single growing season, the cropping systems were altered to include: M, MC, maize and cowpea intercropping (MCI), and cowpea-maize rotation (CM) (Table 15). For the Guinea Savannah, the difference between MC and CM crop rotations is that under MC rotation maize started the cropping sequence in the first trial year followed by cowpea in the following year, whereas the CM crop rotation featured cowpea as the first crop followed by maize the next year (Table 16).

Table 16. Factor treatments in the Guinea Savannah agro-ecosystem

Main Plot Factors	Sub-Plot Factors
No-Till (NT)	Maize (M)
Zonal Till (ZT)	Maize-cowpea (MC)
Traditional Till (TT)	Maize-cowpea intercrop (MCI)
	Cowpea-Maize rotation (CM)

Triple superphosphate (TSP), urea, and compost fertilizer treatments were laid out in a split-plot design in a maize (*Zea mays*) monoculture cropping system with three replications. TSP was the main plot treatment (Table 17). Urea and compost were assigned to the sub-plot treatments.

Table 17. Factors treatments (revisited)

Plot	Treatment	Rate
Main Plot	TSP	0 kg ha ⁻¹
		20 kg ha ⁻¹
		40 kg ha ⁻¹
Sub-Plot (A)	Urea	0 kg ha ⁻¹
		70 kg ha ⁻¹
		140 kg ha ⁻¹
Sub-Plot (B)	Compost	0 t ha ⁻¹
		3 Mg ha ⁻¹
		6 Mg ha ⁻¹

The rates of TSP and urea were adjusted after the first year of treatments due to poor crop response. In 2012 TSP fertilizer rates were raised from 0, 6.5 and 13 kg ha⁻¹ to 0, 20 and 40 kg ha⁻¹ and urea application rates were raised from 0, 45 and 90 kg ha⁻¹ to 0, 70 and 140 kg ha⁻¹. The compost rates were maintained at 0, 3 and 6 Mg ha⁻¹ but the source was changed from Ecofertilizer (3.2% N, 3.3% P₂O₅ and 4.5% K₂O) to Asaase Nufusuo (cocoa (*Theobroma cacao*) husk) (3.2% N, 3.2% P₂O₅ and 1.3% K₂O) since the Ecofertilizer was suspected to be immobilizing N due to high input C:N ratios based on treated plants looking very pale in the first year. These changes were maintained for the 2013 growing season when the fields were sampled for the year of study evaluated in this chapter.

4.3.3. Soil Sampling and Processing

Soils were sampled in December 2013, three years after tillage and cropping and soil amendment treatments were initiated in January 2011. Soils were collected using a 2 cm diameter soil probe to a depth of 15 cm. Three soil cores were taken across the central row and bulked on site. Soils were air-dried and shipped to Texas A&M University for analysis. Larger soil peds were gently broken using a mortar and pestle prior to sieving to <2 mm. Soil samples (3.5 g) were dissolved in either 35 g of 0.1 M HCl (1:10 soil:HCl ratio) or 35 g cold UPW (1:10 soil:UPW ratio) and shaken for two hours at 500 rpm on a rotary shaker. Samples were then centrifuged for 15 minutes at 19,974 g-force and filtered using a Whatman GF/F filter (nominal pore size 0.7 μm) to remove any remaining floating organic material.

Soil extracts were analyzed after extraction for extractable organic C (EOC), total extractable N (TEN), $\text{NH}_4\text{-N}$ and $\text{NO}_3\text{-N}$. Extractable organic nitrogen (EON) was calculated by deducting $\text{NO}_3\text{-N}$ plus $\text{NH}_4\text{-N}$ from TEN.

4.3.4. Chemical Analyses

Extractable organic carbon (EOC) and total extractable nitrogen (TEN) were measured using a high temperature Pt-catalyzed combustion with a Shimadzu TOC-VCSH and Shimadzu total measuring unit TNM-1 (Shimadzu Corp. Houston, TX, USA). Extractable organic carbon (EOC) was measured as non-purgeable carbon, which entails acidifying the sample (250 μL 2 M HCl) and sparging for 4 min with C-free air. $\text{NH}_4\text{-N}$ was analyzed using the phenate hypochlorite method with Na nitroprusside

enhancement (USEPA method 350.1) and NO₃-N was analyzed using Cd-Cu reduction (USEPA method 353.3). All colorimetric methods were performed with a Westco Scientific Smartchem Discrete Analyzer (Westco Scientific Instruments Inc. Brookfield, CT, USA). Sample replicates, water blanks, NIST (National Institute of Standards and Technology) traceable and check standards were run every 12th sample to monitor instrument precision.

4.3.5. Statistical Analyses

A total of 40 samples from the four different agro-ecosystems, undergoing either tillage and cropping or soil amendments were extracted with both 0.1M HCl or UPW for this experiment to examine if there were significant differences in concentrations of NO₃-N, NH₄-N, EOC and EON that might be caused by the type of extractant used. Soils from each agro-ecosystem were grouped by either tillage and cropping treatment or soil amendment treatment with 4-5 soils examined from each agro-ecosystem for each treatment group. Univariate analysis of variance was performed on the tillage and cropping and on the soil amendment experiments separately to determine if there was a significant effect of agro-ecosystem, extract type, and tillage method, cropping system or soil amendment on concentrations of EOC, EON, NO₃-N, NH₄-N and TEN ($\alpha < 0.05$). Student 2-sample, 2-tailed t-tests ($\alpha < 0.05$) were used for each individual agro-ecosystem and either tillage and cropping or soil amendment in turn to determine if there was a significant difference in residual nutrient concentrations among the two extracts used.

Because of the large significant differences in N species when comparing their extraction rate after using UPW extracts or 0.1M HCl extracts for the current study, the previous year's soil collections from the same plots as used in this study (n = 40) and also extracted with 0.1 M HCl were examined using Student 2-sample, 2-tailed t-tests ($\alpha < 0.05$) to assess if there was a significant effect of extract on the concentration of N species. A further analysis of the distribution of N species within TEN was performed for samples collected in 2012 and samples collected in 2013 and the two years compared.

4.4. RESULTS

The objective of this research was to determine if there was a significant difference in DOC, DON and nutrient concentrations when using 0.1 M HCl compared to cold ultrapure water extract.

Data for the tillage and cropping experiment and for the soil amendment experiment were statistically analyzed separately.

4.4.1. Effect of Zone, Extract, and Treatment on N and C Concentrations for Tillage and Cropping

There was no significant effect of tillage practice on the determined concentrations of soil nitrogen or carbon and so the data were pooled and examined using agro-ecosystem, extract used and cropping as fixed factors in univariate analysis of variance.

4.4.1.1. *NO₃-N*

There was a significant effect of agro-ecosystem type ($p < 0.001$), type of extractant used ($p < 0.001$) and crop ($p < 0.01$) on the extracted concentrations of $\text{NO}_3\text{-N}$ (Table 18). An interaction was observed between agro-ecosystem x crop which had a significant effect on $\text{NO}_3\text{-N}$ concentrations ($p < 0.001$).

4.4.1.2. *NH₄-N*

Agro-ecosystem type ($p < 0.01$) and type of extract ($p < 0.001$) had a significant effect on $\text{NH}_4\text{-N}$ concentration from Ghanaian soils (Table 18). There was a significant interaction between agro-ecosystem x extract ($p < 0.05$) and between agro-ecosystem x crop ($p < 0.05$) on the concentrations of $\text{NH}_4\text{-N}$.

4.4.1.3. *EON*

There was a significant effect of agro-ecosystem type ($p < 0.01$), extract ($p < 0.001$) and crop ($p < 0.05$) and a significant interaction of agro-ecosystem type x crop on EON concentrations ($p < 0.05$: Table 18).

4.4.1.4. *TEN*

A significant effect of agro-ecosystem type ($p < 0.01$) and interaction of agro-ecosystem x crop ($p < 0.01$) was observed for the concentrations of TEN. Extract did not have a significant effect on TEN concentrations ($p = 0.22$).

4.4.1.5. *EOC*

The amount of EOC was significantly affected by agro-ecosystem type ($p < 0.001$). A significant interaction between agro-ecosystem x crop ($p < 0.05$) was

observed on the amount of DOC from Ghanaian soils (Table 18). Type of extract used had no significant effect on the concentrations of EOC ($p = 0.13$).

Table 18. Effects of zone, extract and cropping and their interactions on N species.

	NO ₃ -N			NH ₄ -N		
	Mean Square	F	Sig	Mean Square	F	Sig
Corrected Model	39.07	29.83	0.000	44.62	5.81	0.001
Intercept	1401.18	1069.52	0.000	1636.92	213.02	0.000
Zone	118.32	90.32	<0.001	89.97	11.71	<0.01
Extract	125.31	95.65	<0.001	393.06	51.15	<0.001
Crop	13.58	10.36	<0.01	20.71	2.70	0.09
Zone x Extract	0.87	0.67	0.59	27.61	3.59	<0.05
Zone x Crop	55.34	42.24	<0.001	29.04	3.78	<0.05
Extract x Crop	3.62	2.77	0.09	7.89	1.03	0.42
Zone x Extract x Crop	3.10	2.36	0.10	11.55	1.50	0.26

	EON			TEN		
	Mean Square	F	Sig	Mean Square	F	Sig
Corrected Model	44.69	4.85	0.003	84.65	2.89	0.03
Intercept	372.61	40.47	0.000	7701.29	263.11	0.00
Zone	76.59	8.32	<0.01	175.89	6.01	<0.01
Extract	211.51	22.97	<0.001	49.43	1.69	0.22
Crop	36.54	3.97	<0.05	37.80	1.29	0.32
Zone x Extract	14.81	1.61	0.24	9.27	0.32	0.81
Zone x Crop	35.70	3.88	<0.05	176.75	6.04	<0.01
Extract x Crop	9.56	1.04	0.41	29.68	1.01	0.42
Zone x Extract x Crop	4.04	0.44	0.84	6.15	0.21	0.97

Zone = Agro-ecosystem

4.4.2. Effect of Zone, Extract, TSP, Urea, and Compost on Residual Soil N and C Concentrations for the Soil Amendments Study

4.4.2.1. Application of TSP on the effect of N and C concentrations

Addition of TSP only had a significant effect on the amount of residual NO₃-N, but there was also a significant effect of agro-ecosystem ($p < 0.001$) and extract ($p < 0.001$) as well as an interaction of agro-ecosystem x TSP ($p < 0.001$) on the concentration of NO₃-N (Table 19). There was no effect of TSP application on any other N species or EOC.

Table 19. Univariate analysis of variance showing significant effects on NO₃-N

	NO ₃ -N		
	Mean Square	F	Sig
Corrected Model	69.80	15.77	0.0000
Intercept	1506.14	340.36	0.0000
Zone	339.75	76.78	<0.001
Extract	184.40	41.67	<0.001
TSP	93.18	21.06	<0.001
Zone *Extract	3.72	0.84	0.49
Zone*TSP	95.31	21.54	<0.001
Extract*TSP	0.20	0.04	0.96
Zone*Extract*TSP	0.42	0.10	0.99

Zone = Agro-ecosystem

4.4.2.2. Application of urea-N and compost on N and C concentrations

There was no effect of TSP on concentrations of N and C on NH₄-N, EON, TEN or EOC so the data was pooled and the sub-plot treatments of urea-N and compost application only were analyzed using univariate analysis of variance (Table 20). Sub-plot was also examined for NO₃-N (Table 20) because of the low sample number, the degrees of freedom were not large enough to examine agro-ecosystem, extract, main and sub-plot effects on residual NO₃-N.

4.4.2.3. NO₃-N

Agro-ecosystem ($p < 0.001$), extract used ($p < 0.001$), application of urea-N ($p < 0.01$) as well as an interaction of agro-ecosystem x urea-N ($p < 0.01$) all had a significant effect on the extraction of NO₃-N (Table 20).

4.4.2.4. NH₄-N

Agro-ecosystem ($p < 0.001$), type of extract ($p < 0.001$) and compost fertilization ($p = 0.05$) had a significant effect on NH₄-N concentrations. Significant interactions on residual NH₄-N were observed for agro-ecosystem x extract ($p < 0.05$), agro-ecosystem x urea-N ($p < 0.001$), agro-ecosystem x compost ($p < 0.01$) and extract x urea-N ($p < 0.01$; Table 19).

4.4.2.5. EON

Agro-ecosystem ($p < 0.01$) and extract ($p < 0.001$) had a significant effect on the concentrations of EON (Table 20). An interaction of agro-ecosystem x extract had a significant effect on EON concentrations ($p < 0.01$).

4.4.2.6. TEN

There was no effect of extract on residual TEN in the soil amendment soils analyzed ($p = 0.73$). There was however an effect of agro-ecosystem ($p < 0.01$), urea-N addition ($p < 0.05$) and an interaction of agro-ecosystem x urea-N on TEN concentrations (Table 20).

4.4.2.7. EOC

Only agro-ecosystem had a significant effect on the extraction of EOC from soils in the soil amendment experiment (Table 21). There was no significant effect of extractant type on EOC concentrations ($p = 0.07$).

Table 20. Effects of zone, extract, urea-N and compost addition and their interactions of N species.

	NO ₃ -N			NH ₄ -N		
	Mean Square	F	Sig	Mean Square	F	Sig
Corrected Model	50.95	3.99	0.004	9.83	22.62	0.000
Intercept	899.61	70.53	0.000	734.76	1690.73	0.000
Zone	205.66	16.12	<0.001	8.13	18.72	<0.001
Extract	134.59	10.55	<0.01	119.17	274.21	<0.001
Urea	91.52	7.18	<0.01	1.08	2.49	0.12
Compost	15.78	1.24	0.32	2.48	5.71	<0.05
Zone *Extract	3.25	0.25	0.86	1.56	3.58	<0.05
Zone*Urea	95.96	7.52	<0.01	9.59	22.07	<0.001
Zone*Compost	6.49	0.51	0.68	7.07	16.28	<0.002
Extract*Urea	0.10	0.01	0.99	4.99	11.48	<0.01
Extract*Compost	0.59	0.05	0.95	1.51	3.48	0.06
Zone*Extract*Urea	0.87	0.07	0.93	5.23	12.03	<0.01
Zone*Extract*Compost	0.94	0.07	0.97	2.05	4.72	<0.05
	EON			TEN		
	Mean Square	F	Sig	Mean Square	F	Sig
Corrected Model	23.20	7.51	0.000	45.27	1.82	0.12
Intercept	293.65	95.09	0.000	4720.85	189.81	0.000
Zone	32.15	10.41	<0.01	201.84	8.12	<0.01
Extract	232.10	75.16	<0.001	3.08	0.12	0.73
Urea	2.38	0.77	0.48	94.92	3.82	<0.05
Compost	4.38	1.42	0.27	46.00	1.85	0.19
Zone *Extract	21.39	6.93	<0.01	10.52	0.42	0.74
Zone*Urea	6.15	1.99	0.17	101.99	4.10	<0.05
Zone*Compost	1.81	0.59	0.63	26.02	1.05	0.40
Extract*Urea	0.64	0.21	0.82	8.49	0.34	0.72
Extract*Compost	1.34	0.43	0.66	0.82	0.03	0.97
Zone*Extract*Urea	1.65	0.53	0.60	1.38	0.06	0.95
Zone*Extract*Compost	1.80	0.58	0.64	3.07	0.12	0.94

Zone = Agro-ecosystem

Table 21. Effect of zone, extract and cropping on EOC and of zone, extract and urea-N or compost amendments on EOC.

<u>Tillage and Cropping</u>	EOC		
	Mean Square	F	Sig
Corrected Model	5260	4.45	0.005
Intercept	375046	317.11	0.000
Zone	17797	15.05	<0.001
Extract	3069	2.59	0.13
Crop	3286	2.78	0.09
Zone*Extract	1844	1.56	0.25
Zone*Crop	4735	4.00	<0.05
Extract*Crop	959	0.81	0.51
Zone*Extract*Crop	1174	0.99	0.47

<u>Soil Amendments</u>	EOC		
	Mean Square	F	Sig
Corrected Model	3286	2.21	0.060
Intercept	401357	270.40	0.000
Zone	15678	10.56	<0.001
Extract	5794	3.90	0.07
Urea	1404	0.95	0.41
Compost	1938	1.31	0.30
Zone *Extract	2015	1.36	0.30
Zone* Urea	2480	1.67	0.22
Zone*Compost	166	0.11	0.95
Extract* Urea	860	0.58	0.57
Extract*Compost	550	0.37	0.70
Zone*Extract* Urea	425	0.29	0.76
Zone*Extract*Compost	272	0.18	0.91

4.4.3. Within Zone Effects on Residual N and C Concentrations

4.4.3.1. NO₃-N

There was no significant difference in the concentration of NO₃-N concentrations extracted from soil under tillage and cropping treatments in the Forest or Guinea Savannah agro-ecosystems (Fig. 29A). The 0.1 M HCl solution extracted significantly more NO₃-N from the Coastal Savannah and Transition agro-ecosystem soils (Fig. 29A). Under soil amendments, the 0.1 M HCl extracted significantly more NO₃-N from soil than the UPW in the Coastal Savannah, Forest and Transition agro-ecosystems but there was no significant difference in NO₃-N extracted from the Guinea Savannah soil when comparing 0.1 M HCl with UPW extracts (Fig. 29B).

4.4.3.2. NH₄-N

Significantly higher residual NH₄-N was observed from soils in the Coastal Savannah, Forest and Guinea Savannah agro-ecosystems using 0.1 M HCl compared to UPW under tillage and cropping but there was no significant difference in the concentration of NH₄-N extracted from soil in the Transition agro-ecosystem (Fig. 30A). In the soil amendments study, 0.1 M HCl extracted significantly higher NH₄-N from all agro-ecosystems soil when compared to residual NH₄-N with UPW (Fig. 30B).

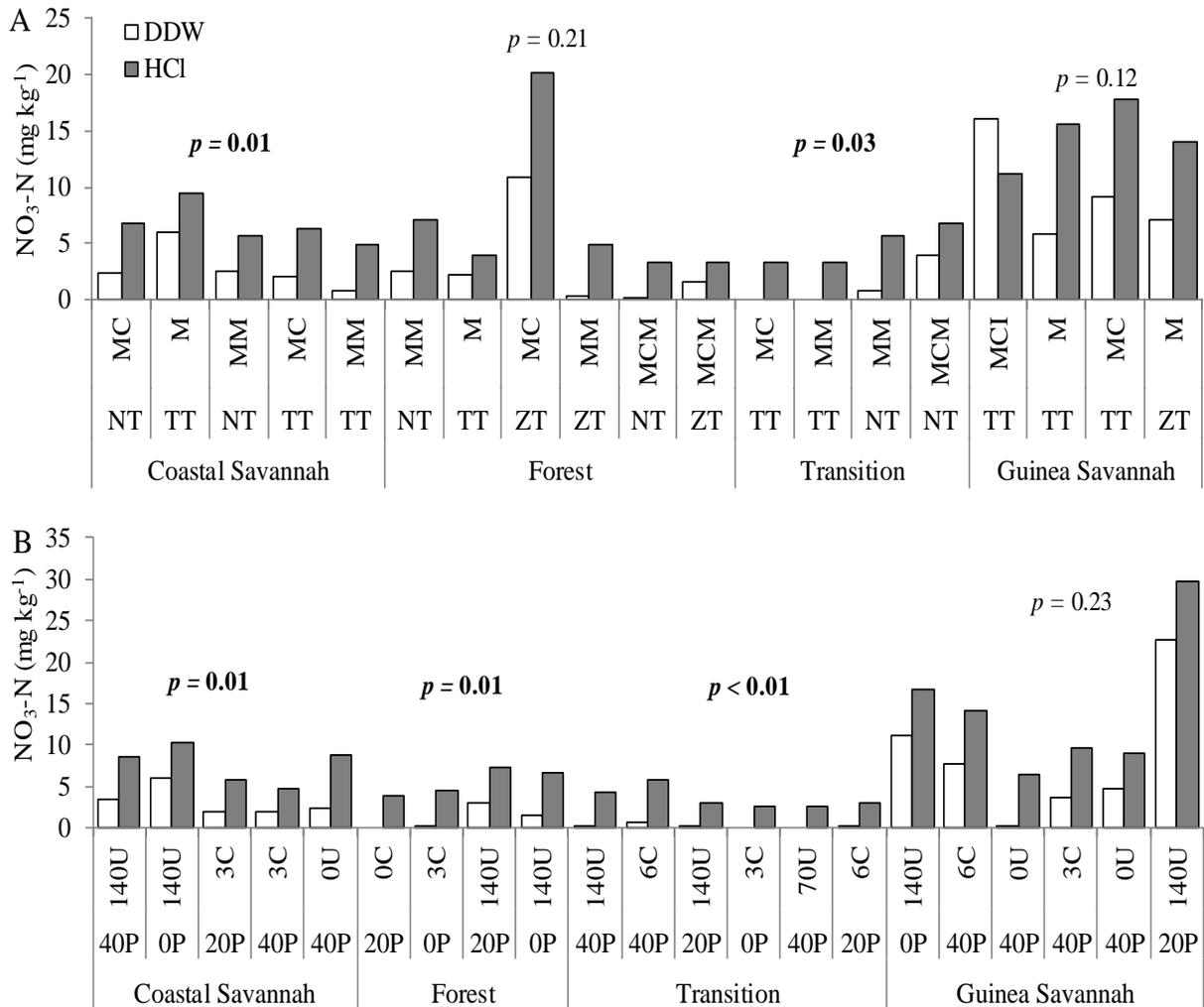


Fig. 29. Comparison of nitrate-N concentrations extracted using UPW versus 0.1 M HCl. (A) tillage and cropping treatments where NT-No Till, TT= Tradition Till and ZT=Zonal Till and M=Maize, MC=Maize-Cowpea rotation, MM=Maize-Mucuna rotation and MCM=Maize-Cowpea-Mucuna relay and (B) fertilizer treatments where 0P=0 kg ha⁻¹ Triple Super Phosphate (TSP), 20P= 20 kg ha⁻¹ TSP and 40P=40 kg ha⁻¹ TSP and 0U=0 kg ha⁻¹ Urea, 70U=70 kg ha⁻¹ Urea, 140U=140 kg ha⁻¹ Urea, 0C=0 kg ha⁻¹ Compost, 3C=3 Mg ha⁻¹ Compost and 6C=6 Mg ha⁻¹ Compost. Significant difference between UPW and HCl is bold ($\alpha < 0.05$).

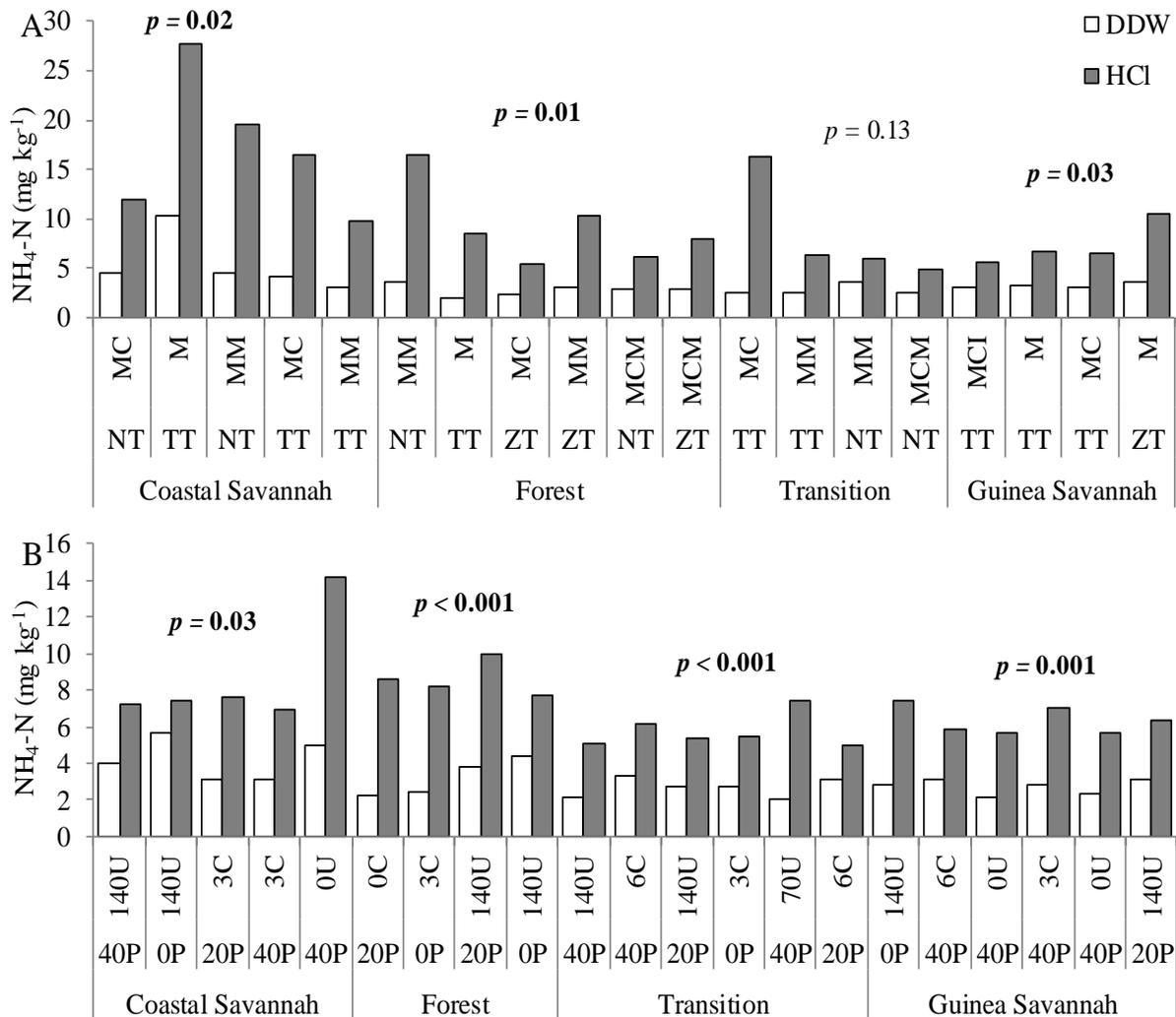


Fig. 30. Comparison of ammonium-N concentrations extracted using UPW versus 0.1 M HCl. (A) tillage and cropping treatments where NT-No Till, TT= Tradition Till and ZT=Zonal Till and M=Maize, MC=Maize-Cowpea rotation, MM=Maize-Mucuna rotation and MCM=Maize-Cowpea-Mucuna relay and (B) fertilizer treatments where 0P=0 kg ha⁻¹ Triple Super Phosphate (TSP), 20P= 20 kg ha⁻¹ TSP and 40P=40 kg ha⁻¹ TSP and 0U=0 kg ha⁻¹ Urea, 70U=70 kg ha⁻¹ Urea, 140U=140 kg ha⁻¹ Urea, 0C=0 kg ha⁻¹ Compost, 3C=3 Mg ha⁻¹ Compost and 6C=6 Mg ha⁻¹ Compost. Significant difference between UPW and HCl is bold ($\alpha < 0.05$).

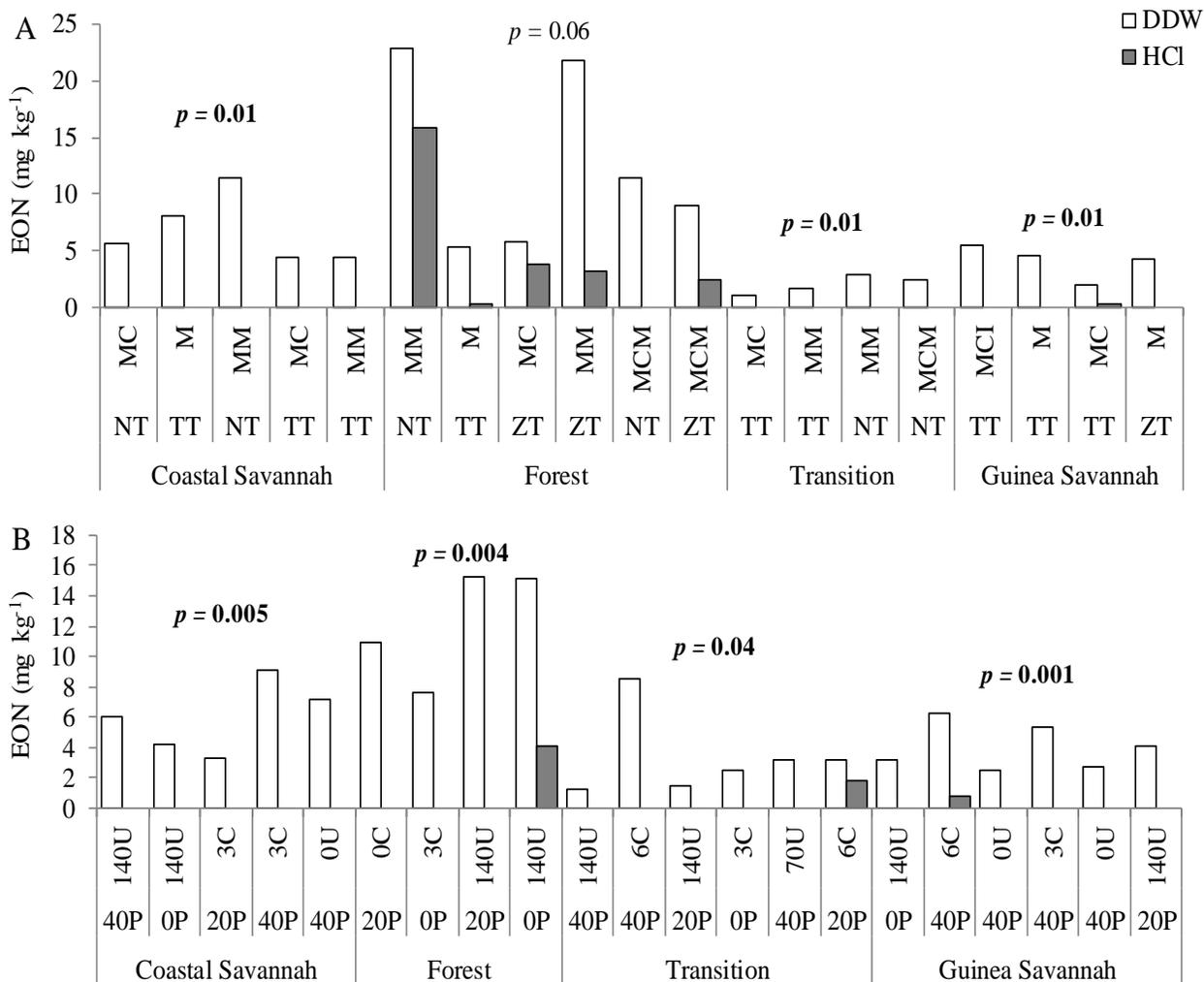


Fig. 31. Comparison of extractable organic N (EON) concentrations extracted using UPW versus 0.1 M HCl. (A) tillage and cropping treatments where NT=No Till, TT=Tradition Till and ZT=Zonal Till and M=Maize, MC=Maize-Cowpea rotation, MM=Maize-Mucuna rotation and MCM=Maize-Cowpea-Mucuna relay and (B) fertilizer treatments where 0P=0 kg ha⁻¹ Triple Super Phosphate (TSP), 20P= 20 kg ha⁻¹ TSP and 40P=40 kg ha⁻¹ TSP and 0U=0 kg ha⁻¹ Urea, 70U=70 kg ha⁻¹ Urea, 140U=140 kg ha⁻¹ Urea, 0C=0 kg ha⁻¹ Compost, 3C=3 Mg ha⁻¹ Compost and 6C=6 Mg ha⁻¹ Compost. Significant difference between UPW and HCl is bold ($\alpha < 0.05$).

4.4.3.3. *EON*

UPW extracted significantly higher EON from the Coastal Savannah, Guinea Savannah, and the Transition agro-ecosystems when compared to the 0.1 M HCl extraction method under the tillage and cropping experiment (Fig. 31A). In the soil amendment study UPW extracted significantly higher EON from all the agro-ecosystem (Fig. 31B).

4.4.3.4. *TEN*

There was no significant effect of extract used in any of the agro-ecosystems examined in either the tillage and cropping study or the soil amendment study ($p = 0.08-0.76$; Fig. 32A, B).

4.3.3.5. *EOC*

In the lower pH soils of the Transition ($p < 0.02$) and Guinea Savannah ($p < 0.03$) agro-ecosystems 0.1 M HCl extracted significantly higher EOC when compared to using UPW as an extract in the soil amendment study (Fig. 33B). There was no significant difference in EOC extraction when using UPW or 0.1 M HCl in the Coastal Savannah and Forest agro-ecosystems for the soil amendment study (Fig. 33B).

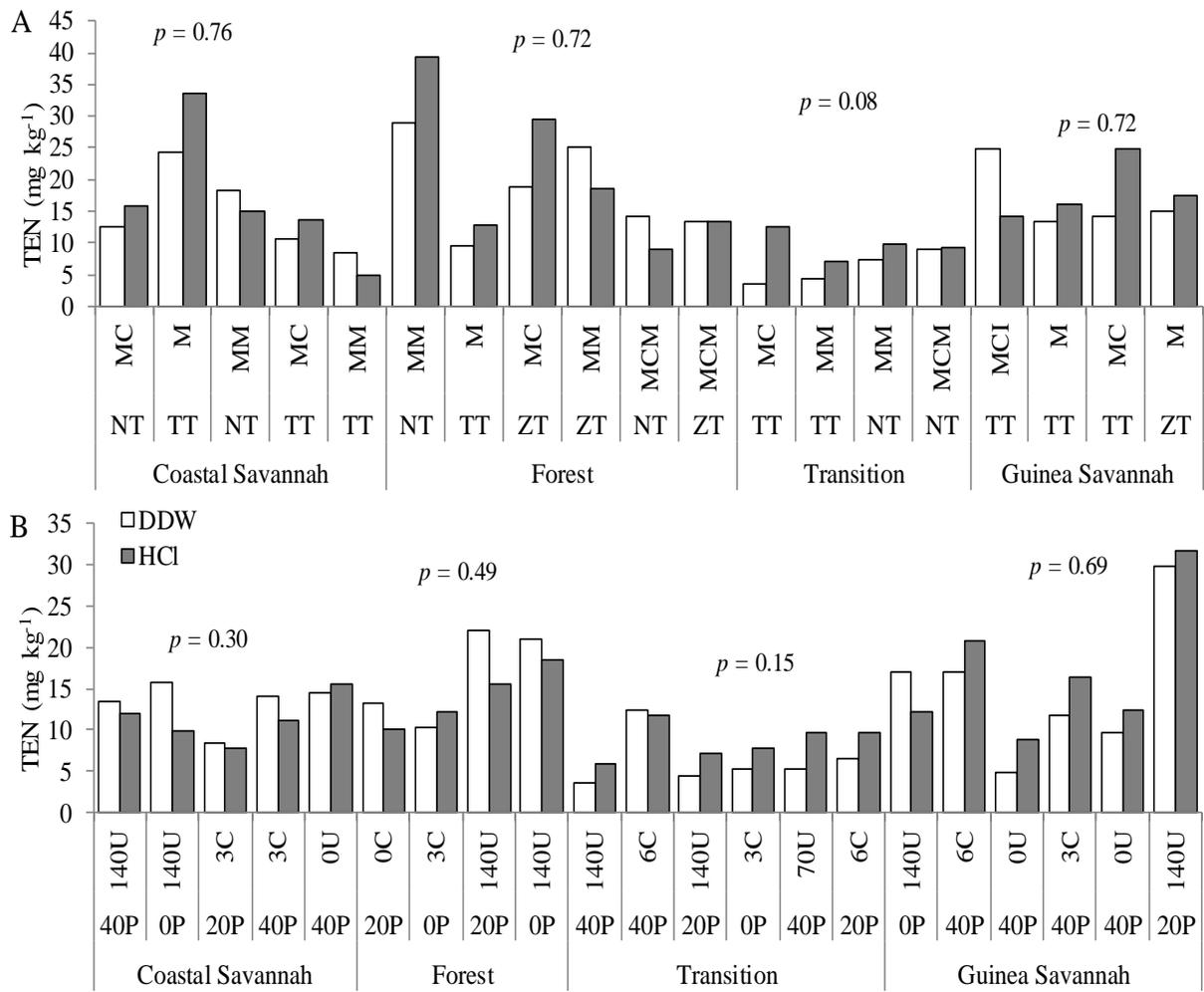


Fig. 32. Comparison of total extractable N (TEN) concentrations extracted using UPW versus 0.1 M HCl. (A) tillage and cropping treatments where NT=No Till, TT= Tradition Till and ZT=Zonal Till and M=Maize, MC=Maize-Cowpea rotation, MM=Maize-Mucuna rotation and MCM=Maize-Cowpea-Mucuna relay and (B) fertilizer treatments where 0P=0 kg ha⁻¹ Triple Super Phosphate (TSP), 20P= 20 kg ha⁻¹ TSP and 40P=40 kg ha⁻¹ TSP and 0U=0 kg ha⁻¹ Urea, 70U=70 kg ha⁻¹ Urea, 140U=140 kg ha⁻¹ Urea, 0C=0 kg ha⁻¹ Compost, 3C=3 Mg ha⁻¹ Compost and 6C=6 Mg ha⁻¹ Compost. Significant difference between UPW and HCl is bold ($\alpha < 0.05$).

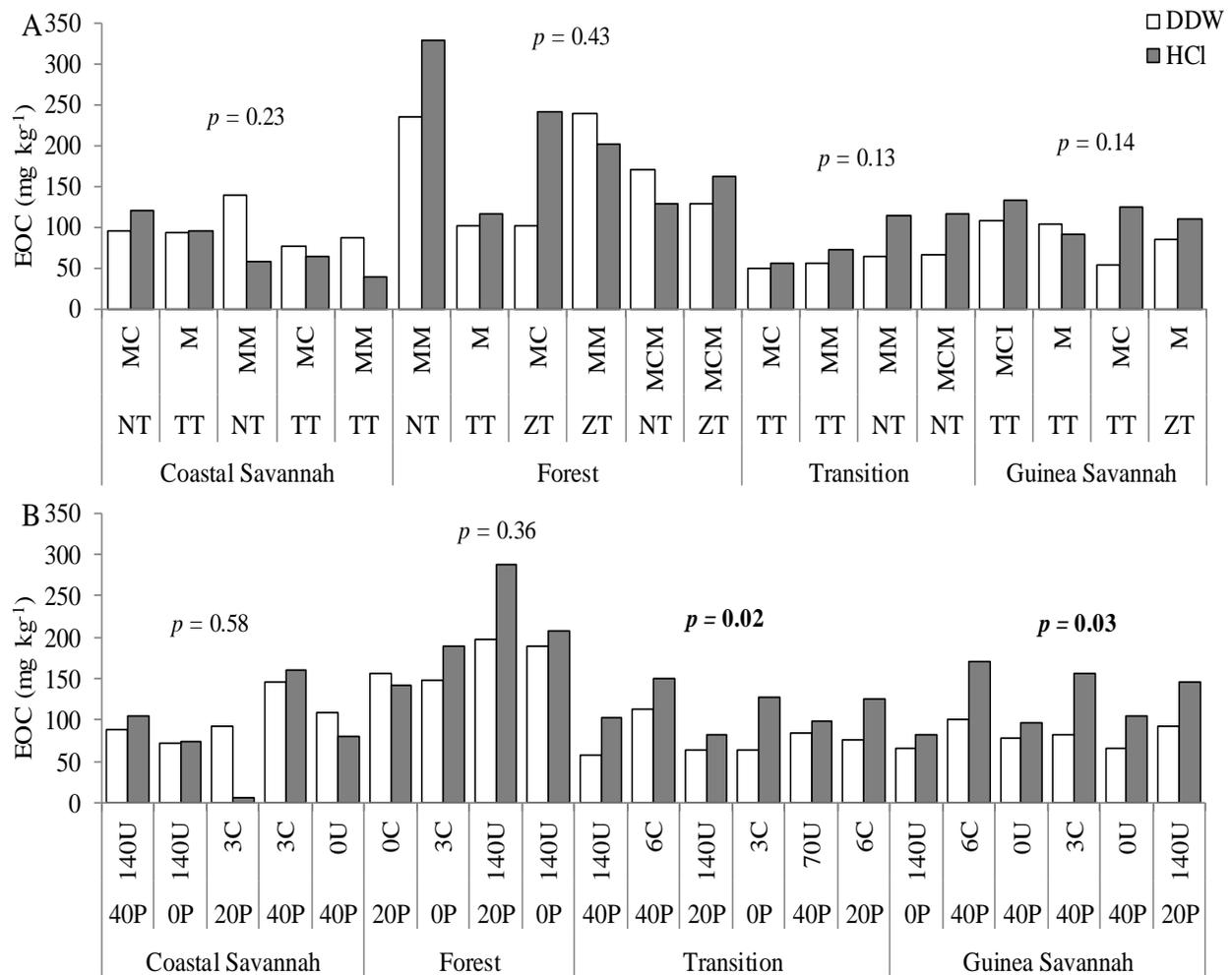


Figure 33. Comparison of extractable organic C (EOC) concentrations extracted using UPW versus 0.1 M HCl. (A) tillage and cropping treatments where NT=No Till, TT=Tradition Till and ZT=Zonal Till and M=Maize, MC=Maize-Cowpea rotation, MM=Maize-Mucuna rotation and MCM=Maize-Cowpea-Mucuna relay and (B) fertilizer treatments where 0P=0 kg ha⁻¹ Triple Super Phosphate (TSP), 20P= 20 kg ha⁻¹ TSP and 40P=40 kg ha⁻¹ TSP and 0U=0 kg ha⁻¹ Urea, 70U=70 kg ha⁻¹ Urea, 140U=140 kg ha⁻¹ Urea, 0C=0 kg ha⁻¹ Compost, 3C=3 Mg ha⁻¹ Compost and 6C=6 Mg ha⁻¹ Compost. Significant difference between UPW and HCl is bold ($\alpha < 0.05$).

4.4.4. Effect of Extract on the Distribution of N Species

Of the soils extracted with UPW in the tillage and cropping 49% was in the form of EON, 27% NO₃-N and 24% NH₄-N to make up 100% TEN (Fig. 34). When the tillage and cropping soils were extracted with 0.1 M HCl the N speciation changed considerably with 7% EON, 40% NO₃-N and 53% NH₄-N making up 100% TEN (Fig. 34). A similar occurrence was observed with the soil amendment soils. When extracted with UPW the speciation of N was 47% EON, 27% NO₃-N and 26% NH₄-N (Fig. 34) but when the same soils were extracted with 0.1 M HCl the distribution of N species changed to 2% EON, 52% NO₃-N and 46% NH₄-N (Fig. 34).

4.4.5. Effect of Extract Type on the Relationship Between DON and DOC

The relation between soil solution and surface water DOC and DON is usually strong and positive (Carrillo-Gonzalez et al., 2013). In both the crop and tillage and the soil amendment studies the UPW extracted soils maintained a strong and significant relationship ($R^2 = 0.92 - 0.98$; $p < 0.001$; Fig. 7A) but when extracted with 0.1 M HCl the relationship was decoupled (Fig. 7B).

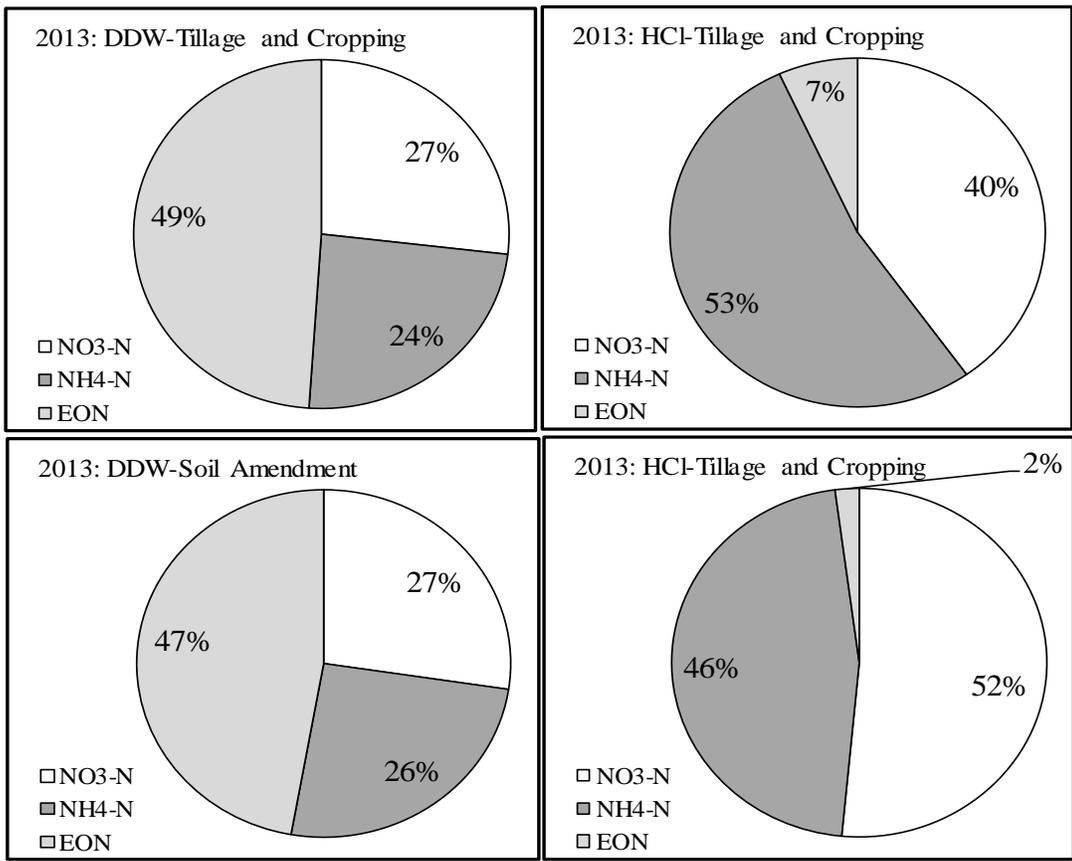


Fig. 34. Nitrogen percent speciation changes with extract used.

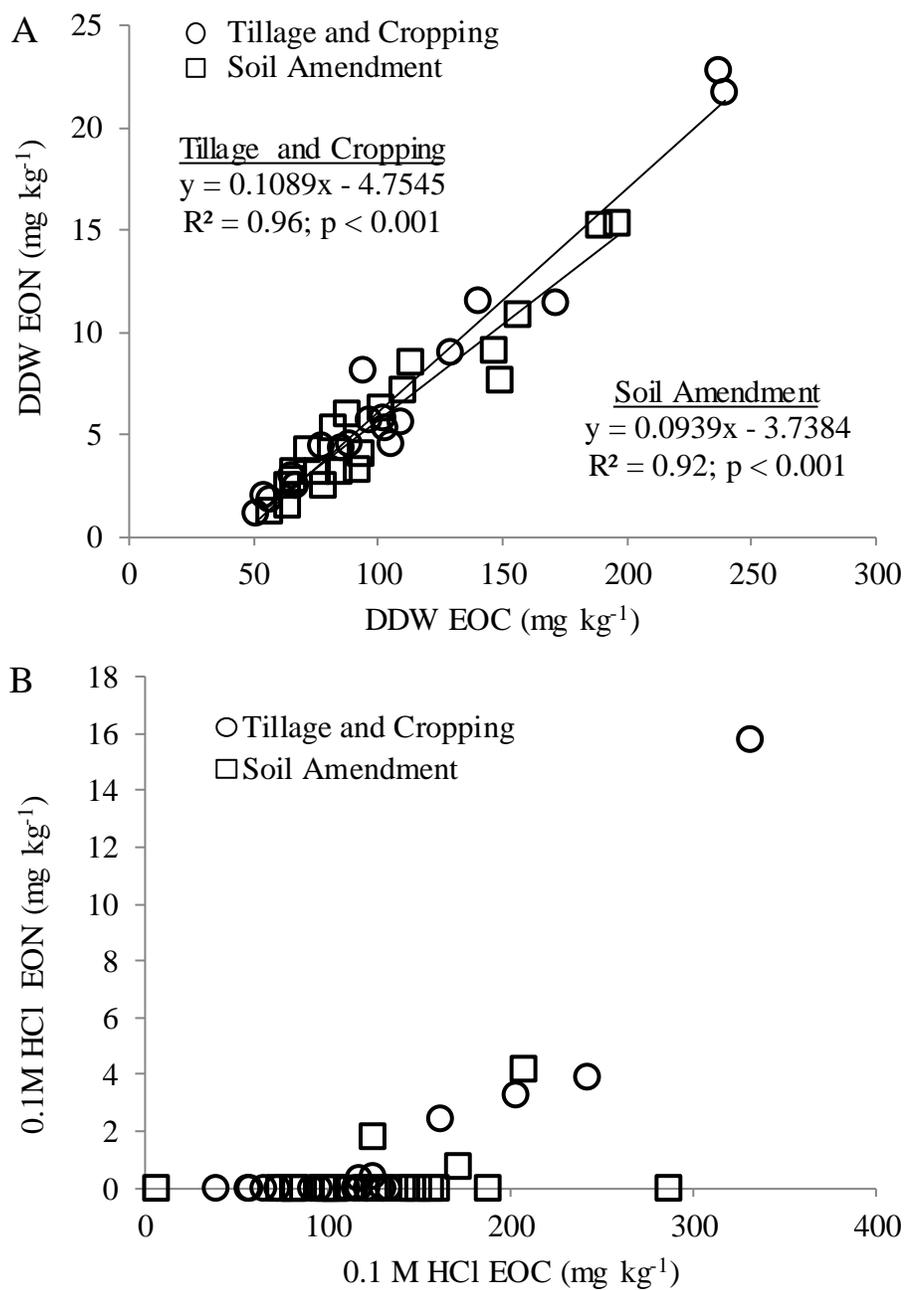


Fig. 35. Relationship between EON and EOC in A) UPW extractable soil and B) 0.1 M HCl extracted soil.

4.5. DISCUSSION

Multiple different extract solutions have been used in the analysis of inorganic-N, organic-N and organic-C from soils over the years (Haney et al., 1999; Gregorich et al., 2003; Haney et al., 2006; Jones and Willett, 2006; Carrillo-Gonzalez et al., 2013; Inselbacher et al., 2014; Davies et al., 2014).

4.5.1. Extraction of N and C from Tropical Agricultural Soils

4.5.1.1 Extraction of inorganic and organic N

Over the last decade there have been several studies examining the performance of different soil extracts on N detection (Bordoloi et al., 2013; Carrillo-Gonzalez et al. 2013; Inselsbacher, 2014). Bordoloi et al. (2013) suggested that in comparing biological and chemical methods for determination of soil nitrogen supply that biological methods tended to be the most reliable as the concentrations are most correlated with plant N-uptake and crop yields. However they further suggested that biological methods are tedious and not recommended for routine analysis. Of the chemical extractants tested, a phosphate borate buffer method (Gianello and Bremner, 1986) displayed the highest correlation with plant nitrogen uptake (Bordoloi et al., 2013).

Davies et al (2014) used a 0.1 M extract on soils collected from the same locations collected year earlier which was two years after the treatments were initiated. The concentration of N species was significantly different when comparing the study of Davies et al. (2014) and the current study. In the 2012 study for tillage and cropping, N species distribution was 36% EON, 41% NO₃-N and 23% NH₄-N but in the 2013 study for tillage and cropping there was a significant loss of EON and a doubling of NH₄-N although the percentage of TEN that was NO₃-N remained constant between the two years (Fig. 8). In the Davies et al. (2014) study of 2012 with a 0.1 M HCl extract on the soil amendment soils, EON was 19% of TEN could be somewhat expected in fertilized soils; NH₄-N had 46% of NH₄-N which would be expected due to the urea addition and NO₃-N was 35% of TEN. Using the same SOP on soils collected in 2013 (current study), There was a 17% loss of EON and an 18% gain of NO₃-N with NH₄-N between the two years remaining relatively consistent (Fig. 36).

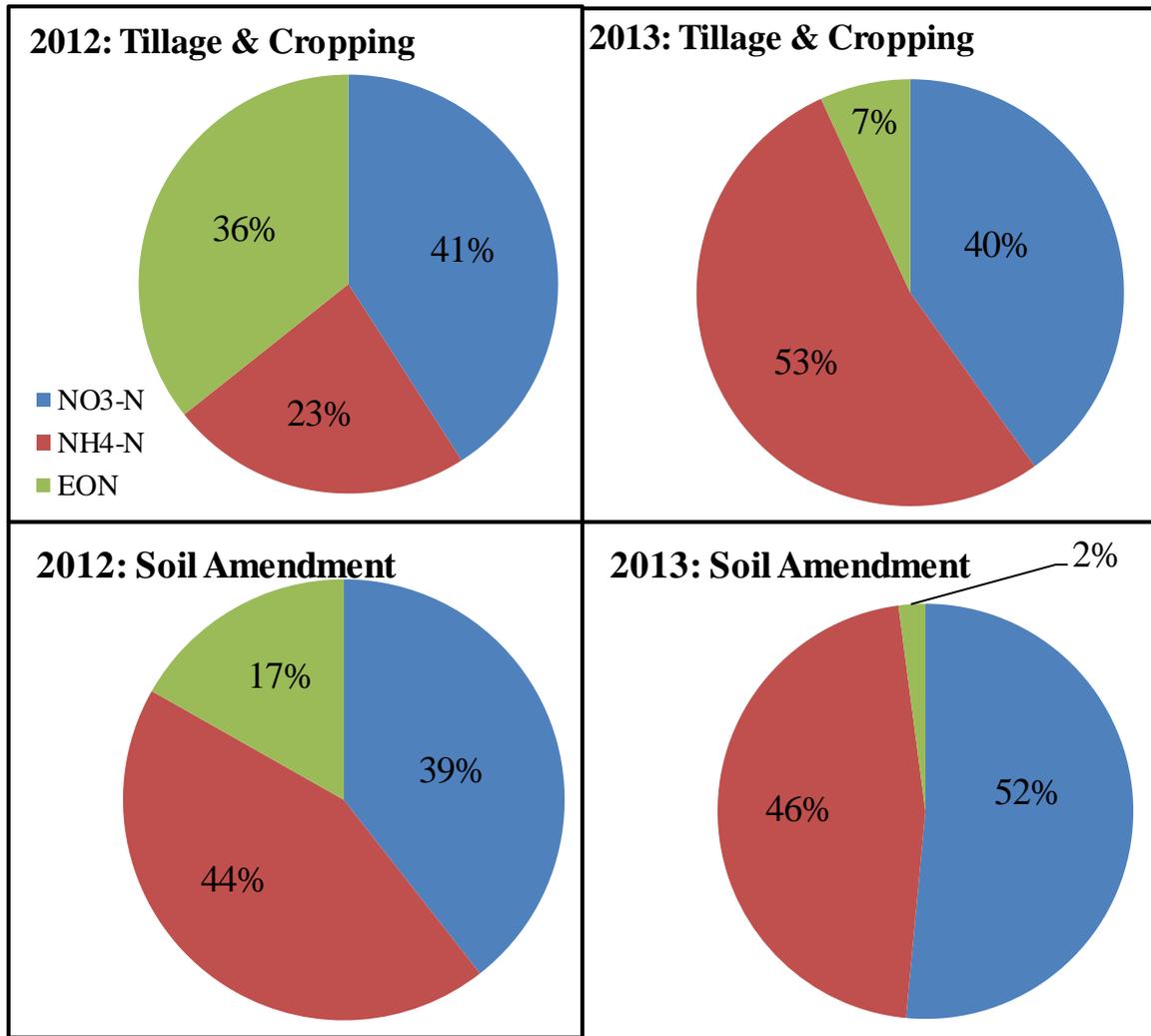


Fig. 36. Percentage of nitrogen species that made up TEN in the 2012 and 2013 soil collections for the 40 soils across all agro-ecosystems that were used in this study. Percentages are for the samples only that were used in the extract comparison.

This discrepancy in inorganic and organic N species between years (2012 and 2013) using the same SOP suggests that either field treatments were starting to have an effect or that there was some deviation in the SOP for the 2013 study.

Extraction of DON in agricultural soils is rarely seen in the literature with most European researchers preferring to report DOM with the exception of a few temperate soil studies by Jones and Willet (2006) and Gregorich et al. (2003) and the tropical soil study by Carrillo-Gonzalez et al. (2013). Gregorich et al. (2003) reported that DON accounted for 61-83% of total nitrogen extracted with cold water and 87-97% of the total N extracted with hot water. The Gregorich et al. (2003) study was performed on soils under manured and non-manured maize cropping in Canada. The effect that 0.1 M HCl had on the extraction of EON in the current study was quite shocking especially since EON concentrations were relatively high in the tillage and cropping experiment during 2012 when it ranged from 5 ± 3 to 12 ± 4 mg kg soil⁻¹ in the coastal savannah, from 15 ± 13 to 27 ± 3 mg kg soil⁻¹ in the forest, from 4 ± 7 to 24 ± 9 mg kg soil⁻¹ in the transition and from 0 ± 0 to 9 ± 1 mg kg soil⁻¹ in the Guinea Savannah (Davies et al. 2014). In the current study EON extracted by 0.1 M HCl was for the most part non-detectable in both the tillage and cropping experiment and the soil amendment studies (Chapters 2 and 3). A number of reasons can be put forth for this anomaly: 1) the preparation of the 0.1 M HCl extract was incorrect in this study, 2) time between collection and receipt of soils by the NaWA laboratory was compromised due to new restrictions by the Ghanaian Government. 3) the field treatments had an effect on the speciation and molecular

weights of EON during year 2013 or 4) extracted samples were not analyzed on the day of extraction or within the 18 h holding time allowing the acidic extract the opportunity to digest and mineralize the EON in solution.

4.5.1.2. Extraction of DOC

Dissolved organic matter (DOM) includes dissolved organic carbon (DOC), organic nitrogen (DON), organic sulfur (DOS) and organic phosphorus (DOP). DON, DOS and DOP can be considered subsets of DOC because once the functional group (i.e. NH_2 , SH, R-P) has been cleaved by extracellular enzymes the molecule will become DOC. Dissolved organic matter (DOM) extracted from forest and agricultural soils has generally been performed using a cold DDW (Linn and Doran, 1984; Cronan et al. 1992; De Luca and Keeney 1993, 1994; Zsolnay and Gorlitz 1994; Boyer and Groffman 1996; Delprat et al. 1997; Erich and Trusty 1997; Jensen et al. 1997; Norman et al. 1997; Gregorich et al. 1998, 2000; Rochelle and Gregorich 1998; Haney et al. 1999; Haynes 2000; Aitkenhead-Peterson et al. 2006; Jones and Willett 2006; Carrillo-Gonzalez et al. 2013). The pH of soil solution, generally obtained by zero tension or tension lysimeters can range in forest soils (Hendershot and Courchesne, 199) suggesting that analysis of DOM using a neutral solution will not reflect equilibrium or exchangeable DOM that is in soils with lower pH values. This has led to the examination of other extractants compared to a water extract for analysis of DOM (DeLuca and Keeney 1993, 1994; Zsolnay and Gorlitz 1994; Rochelle and Gregorich 1998; Haney et al. 1999; Gregorich et al. 2000; Jones and Willett 2006; Carrillo-Gonzalez et al. 2013). Alternative

extractants for DOM detection have been either 4 mM K_2SO_4 or 125 mM and 0.5 M $CaSO_4$ until the work by Jones and Willett (2006) on temperate soils and Carrillo-Gonzalez et al. (2013) on tropical soils. Jones and Willett (2006) examined soils obtained from agricultural pasture and crops as well as forest soils and used multiple extracts, different sieve sizes, soil:extract ratios, different shaking times, different extraction temperatures to answer the call by Chantigny (2003) to develop a standard method for the extraction of DOM. Carrillo-Gonzalez et al. (2013) opted to examine tropical soils under tillage and cropping and kept conditions (sieve size, soil:extract ratio, shaking time and temperature) constant and used cold DDW, hot DDW, 0.5 M K_2SO_4 , 2 M KCl and 10 mM $CaCl_2$ as extractants and reported a significant effect of extract type on the concentrations of DOC. Detection of DOC was $K_2SO_4 > hot DDW > KCl > cold DDW = CaCl_2$ (Carrillo-Gonzalez et al. 2013). Soil leachate pH in the Carrillo-Gonzalez et al. (2013) study ranged from pH 7.8-8.6 depending on tillage or cropping treatment which would suggest that a neutral-basic extract would be appropriate to analyze exchangeable DOC. Haney et al. (1999) examined C concentrations detected using hot water, cold water and 0.5 M K_2SO_4 and reported some interesting results. They found that the maximum residual C was detected at pH 6.5 using either hot or cold water and at pH 8.5 using 0.5 M K_2SO_4 ; furthermore they showed that as soil pH decreased below pH 6.5 or increased above pH 6.5 extracted C decreased using both hot and cold water. The significance of the effect of soil pH on extractant selected cannot be understated. This prior work by Haney et al. (1999)

suggested that a weak acid solution may be an appropriate extract for detection of C in the acid soils of the agro-ecosystems in Ghana. Soils in the current study had pH ranging from pH 4.7-6.3 prior to tillage and cropping and soil amendment treatment initiation in 2011; the higher pH observed in the Coastal Savannah and Forest soils and lower pH in the Transition and Guinea Savannah soils (Davies et al., 2014). Based on this information the decision to use a weak acid (0.1 M HCl) extractant was made for the current study. This decision appeared to be well founded for the tillage and cropping study where no significant difference in EOC amounts was found when comparing DDW and 0.1 M HCl extract. Similarly in the soil amendment study there was no significant difference in extracted EOC with UPW vs 0.1 M HCl in the higher soil pH Coastal Savannah or Forest agro-ecosystems but in the lower pH Transition and Guinea Savannah agro-ecosystems a slightly higher, but nevertheless significant EOC concentrations were observed with 0.1 M HCl extract.

The length of time a soil is shaken during extraction has a significant effect on the concentration of DOC (Jones and Willett, 2006). According to their published paper, soils released their maximum DOC at about 6 hours of shaking at a temperature of 20° C and the concentration thereafter (up to 24 h) remained fairly constant. The soils in the current study were shaken for 2 h for both 0.1 M HCl and UPW at room temperature (20° C) with the expectation of recovering EOC that would be analogous to DOC in soil solution. What was unexpected in this study was how much more EOC was obtained using 0.1 M HCl compared to UPW for some of the agro-ecosystems. For example,

EOC was significantly higher in the transition and Guinea Savannah agro-ecosystems that were undergoing fertilization which suggests, because of their low initial pH that a weak acid is appropriate for recovering DOC from acid soils. Use of HCl on soils primarily to test for carbonates has been speculated to destroy some organic carbon compounds leading to a loss in quantification (Shumacher 2002). Overall, at the molarity of HCl used there was no statistically significant difference in EOC extracted whether using UPW or 0.1 M HCl.

4.5.1.3 Potential causes of radical change in N speciation between years

The salts in extracts can have a significant effect on the detection of N species (Carrillo-Gonzales et al. 2013). Examination of N species and EOC extraction using 0.1 M HCl from the same plots and sites used in this comparison study and collected in 2012 showed some significant difference in C and N amounts but also significant similarities (Table 22), suggesting that the molarity of the extract was accurate. New Ghanaian Government regulations in 2013 meant that all collected soils to be shipped out of the country had to be physically examined by the Ghanaian Geological Survey and passed for shipment before they could be sent. They were responsible for the packaging of soils in 2013 whereas in 2012 NAWA personnel packaged the soils for shipment. Soils from the 2013 sampling trip were not collected from Houston airport until 19th February, 2014 and processing commenced in June, 2014. In contrast, soils from the 2012 sampling trip were delivered as packed by researchers and received on 26th December, 2012 and 2nd January, 2013 and processing commenced in July 2013. Given similar timelines

between soil collection and processing it is expected that time between deliveries and processing would not have an effect on the soils which were air dried in Ghana before shipping.

Table 22. Significant differences in the residual C and N between 2012 and 2013 using a 0.1 M HCl extract.

	NO ₃ -N	NH ₄ -N	EOC	TEN	EON
<u>Tillage & Cropping</u>	Significance (p =)				
Coastal Savannah	0.01	0.01	0.15	0.49	0.04
Forest	0.17	0.86	0.01	0.01	0.00
Transition	0.21	0.85	0.06	0.05	0.03
Guinea Savannah	0.10	0.39	0.00	0.02	0.14
<u>Soil Amendment</u>					
Coastal Savannah	0.11	0.99	0.26	0.00	0.14
Forest	0.01	0.04	0.01	0.29	0.83
Transition	0.46	0.10	0.00	0.06	0.06
Guinea Savannah	0.62	0.04	0.00	0.04	0.05

Inselbachers (2014) work examining the effect of sieving and extraction type on extraction rates of inorganic-N and amino acids from forest and agricultural soils in Sweden suggested that the act of sieving and extraction of soils led to the microbial mineralization of amino acids to inorganic-N which may lead to an over-estimation of inorganic-N and under-estimation of organic-N which is important when comparing concentrations of individual N forms in different soils but less important when considering total N. The same standard operating procedures (SOPs) were performed for soil sieving and extraction and so it is unlikely that sieving caused microbial

mineralization of amino acids in 2013 and not in 2012. Extracts of the 2012 soils were analyzed on the day of extraction with sets of ~40 samples extracted daily and analyzed immediately. For the 2013 soils they were analyzed sporadically between 1-24 hours after extraction which may have initiated digestion and mineralization of amino acids in the tillage and cropping extracts and digestion, mineralization and nitrification in the soil amendment extracts (Fig. 36). This sporadic analysis or delay in analysis of N may have had a significant effect on the results of the N species examined in the current study as well as on all the soils collected in 2013. An effect of treatment on C and N concentrations between the two years could also explain the very different N speciation between 2012 and 2013. A first glance at the data when comparing UPW and 0.1 M HCl would suggest that the extractant used was the cause for the different speciation and that the acid extract indeed mineralized the entire organic N. Examination of the data using univariate analysis of variance suggested that this was the case in the soil amendment study but in the tillage and cropping study where cropping also had a significant effect on EON concentrations. This extreme change in speciation of N between UPW and HCl extracts prompted examination and comparison of soils collected in 2012 from the same plots (Fig. 36).

One of the biggest issues with quantification of EON is that there is no direct measure for it. Researchers typically quantify total N and deduct inorganic-N for a value of organic-N. Because of the reactivity of N species in solution and transformations between mineralization and immobilization, it is recommended that all measurements for

N occur on the same day. In fact the USEPA method 353.3 states a maximum holding time of 18 h for NO₃-N analysis.

4.5.1.4 Selection of an appropriate soil extract

Over the last decade there are still questions posed as to the best method to extract EOC, EON especially if extractable NO₃-N and NH₄-N concentrations are needed as well to avoid multiple extraction procedures (Jones and Willett 2006; Carrillo-Gonzalez et al. 2013). Based on this experiment it is suggested that either cold UPW or 0.1 M HCl extract are suitable for analysis of EOC and TEN from acidic tropical soils. Delay in extract analysis when using 0.1 M HCl may result in mineralization of EON and its subsequent conversion to NH₄-N and NO₃-N suggest that the 0.1 M HCl method should be used in a disciplined manner with a set number of samples extracted daily (40) and immediate analysis for NO₃-N, NH₄-N and TEN.

Soil solution dissolved organic carbon (DOC) and organic nitrogen (DON) is best retrieved using a zero-tension lysimeter as this adequately describes the solution *in situ* as it percolates a soil. However it is not always convenient to instrument a site and rely on staff in a different country to collect samples and ship them in a timely manner. This is why some researchers use a cold water to extract DOC from air dried soil. Nomenclature of these extracts varies in the literature from DOC and DON (Wright et al. 2007; Carrillo-Gonzalez et al. 2013) to WEDOC and WEDON (Aitkenhead et al. 2007). Carrillo-Gonzalez et al. (2013) reported that cold water extractable DOC from a high pH (8.3-8.6) Fluventic Ustochrept soil was not significantly different in concentration when

compared to DOC extracted from the same soil after it had been leached. While the use of cold water to extract soils removes the effect of ionic strength or electrolyte concentration on DOC and DON concentrations there still may be an effect of pH for acidic or alkaline soils.

In studies where hundreds of soil samples from multiple experimental treatments are collected (i.e. Davies et al. 2014; Aitkenhead-Peterson et al. 2015) a decision must be made as to an appropriate extractant to use to quantify multiple soil chemistries such as EOC, EON, NO₃-N and NH₄-N (Davies et al. 2014) or EOC, EON, NO₃-N, NH₄-N, PO₄-P, Na⁺, K⁺, Mg²⁺ and Ca²⁺ (Aitkenhead-Peterson et al. 2015) especially in university research laboratories where generally one graduate student does all of the work. The 1 M HCl is frequently the extractant of choice for the detection of cations and prior work by Haney et al. (1999, 2006) suggests that use of an extractant that is compatible with the pH of the soil is desirable for the analysis of EOC. Because the detection of EOC (and likely EON) is a result of desorption from soil minerals the ionic strength or electrolyte concentration of the extractant also becomes important.

Table 23. Comparisons of soil extracts from Ghana (W. Africa) and Texas (USA) under No-Till (NT) and Rotational Cropping (R) and Traditional-Till (TT) and Rotational Cropping (R). UPW=cold ultrapure water and DDW(h) = hot distilled deionized water.

Location	TMT	Depth (cm)	Condition	Ratio	Extract	Shake (min)	Filter (μ m)	EOC mg kg ⁻¹	EON mg kg ⁻¹	Source
Ghana	NT-R	0-15	Air dry	1:10	UPW	120	0.7	129	10	1
Ghana	NT-R	0-15	Air dry	1:10	0.1 M HCl	120	0.7	100	0	1
Ghana	TT-R	0-15	Air dry	1:10	UPW	120	0.7	77	4	1
Ghana	TT-R	0-15	Air dry	1:10	0.1 M HCl	120	0.7	54	0	1
Ghana	NT-R	0-15	Air dry	1:10	0.1 M HCl	120	0.7	67	10	2
Ghana	TT-R	0-15	Air dry	1:10	0.1 M HCl	120	0.7	60	10	2
Texas	NT-R	0-5	Air dry	1:4	DDW	60	0.45	250	-	3
Texas	TT-R	0-5	Air dry	1:4	DDW	60	0.45	200	-	3
Texas	NT-R	0-5	Moist	1:5	DDW	15	2.5	150	13	4
Texas	NT-R	0-5	Moist	1:5	DDW(h)	15	2.5	260	33	4
Texas	NT-R	0-5	Moist	1:5	10 mM CaCl ₂	15	2.5	130	7	4
Texas	NT-R	0-5	Moist	1:5	2 M KCl	15	2.5	190	4	4
Texas	NT-R	0-5	Moist	1:5	0.5 M K ₂ SO ₄	15	2.5	280	12	4
Texas	TT-R	0-5	Moist	1:5	DDW	15	2.5	90	6	4
Texas	TT-R	0-5	Moist	1:5	DDW(h)	15	2.5	155	17	4
Texas	TT-R	0-5	Moist	1:5	10 mM CaCl ₂	15	2.5	50	4	4
Texas	TT-R	0-5	Moist	1:5	2 M KCl	15	2.5	180	2	4
Texas	TT-R	0-5	Moist	1:5	0.5 M K ₂ SO ₄	15	2.5	175	4	4

Sources: ¹This study, ² Davies et al. 2014, ³ Wright et al. 2007, ⁴ Carrillo-Gonzalez et al. 2013

5. CONCLUSION

Universal adoption of individual tillage practices or crop rotations throughout Ghana is unlikely to enhanced soil nutrients, according to this study. The Coastal Savannah agro-ecosystem tended to display higher concentrations of inorganic-N ($\text{NO}_3\text{-N}$ and $\text{NH}_4\text{-N}$) when compared to all other agro-ecosystems, whereas the forest agro-ecosystem tended to display higher organically bound N and C. Soil texture and prior with in an agro-ecosystem impacted whether N was found in the organic or inorganic form. Multiple studies across West Africa have attempted to find the best agronomic practices to maintain or increase soil fertility for increasing yields (Lal, 1976; Niemeijer et al., 2002; Smaling et al., 2012; Vanlauwe et al., 2006; Giller et al., 1997).

Combinations of fertilizers had a significant effect on the amounts of certain nutrients detected in certain Ghanaian agro-ecosystems but not others. It is important therefore to identify which combination of fertilizer works best for each specific soil nutrient. Soil testing is important to determine which nutrients are needed for agriculture in a broad sense (NPK) and knowledge of fertilizer combinations can help to increase specific N and P needs.

Results after three years of treatments indicated that agro-ecosystem had a significant effect on all the soil nutrients evaluated. The Guinea and Coastal Savannahs had the greatest residual inorganic N, the Forest had the greatest organically bound C and N, and the Coastal Savannah and Transition had the greatest residual P. Within the Coastal Savannah agro-ecosystem maize monocrop was linked with the greatest residual

inorganic N, the application of urea alone led to greatest inorganic N concentration, and that triple superphosphate (TSP) combined with an N input was associated with greater inorganic P concentration. In the Transition zone no-till was associated with high concentrations of inorganic N and P, while the maize-mucuna rotation had high residual inorganic N, and TSP fertilizer was closely linked to greater inorganic P concentrations. Finally, in the Guinea Savannah urea and compost additions are beginning to influence inorganic N soil concentrations and TSP was associated with greater amounts of inorganic P.

One limitation of this investigation was the sampling strategy within a row of crop. As mentioned previously, this method of sampling did not capture well differences between traditional and zonal tillage. Future trial years in this investigation should consider a sampling strategy that can better detect differences between zonal and traditional till, such as random sampling or sampling both between rows and within rows.

Based on the results of this investigation, more years of the current crop management treatments would be very useful before making recommendations to subsistence farmers. Soil collection and analysis in the 5th and final year of this should reveal more differences in nutrient concentrations by treatment within each agro-ecosystem. To the extent possible these results should be linked with crop yield data. A medium-term crop management investigation with soil fertility and yield results is likely to provide highly valuable information to smallholder farmers in these regions.

The most important finding is that a ‘one-size-fits-all’ approach to fertilization in the Ghanaian agro-ecosystems will not produce the same results with respect to increasing soil fertility. Differences in environmental factors such as annual rainfall and soil parent material and texture will affect how nutrient concentrations will respond to fertilizer combinations. Soil scientists have advocated for decades the nutrient requirements based on soil type, cropping system, farm size and the availability of essential inputs (Lal, 1987).

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APPENDIX A

A.1. Tillage x cropping system experiment. Analysis of variance for NO₃N across all zones. Data is log transformed.

Source	Type III Sum of Squares	df	Mean Square	F	Sig.
Corrected Model	8.177 ^a	47	.174	8.486	.000
Intercept	93.637	1	93.637	4567.319	.000
Zone	6.987	3	2.329	113.607	.000
Tillage	.019	2	.010	.472	.625
Cropping	.094	3	.031	1.533	.211
Zone * Tillage	.056	6	.009	.454	.840
Zone * Cropping	.445	9	.049	2.411	.016
Tillage * Cropping	.222	6	.037	1.802	.107
Zone * Tillage * Cropping	.353	18	.020	.957	.514
Error	1.968	96	.021		
Total	103.782	144			
Corrected Total	10.145	143			

a. R Squared = .806 (Adjusted R Squared = .711)

A.2. Tillage x cropping system experiment. Analysis of variance for NH₄N across all zones. Data is log transformed.

Source	Type III Sum of Squares	df	Mean Square	F	Sig.
Corrected Model	4.143 ^a	47	.088	3.460	.000
Intercept	121.043	1	121.043	4751.594	.000
Zone	2.742	3	.914	35.886	.000
Tillage	.014	2	.007	.269	.765
Cropping	.046	3	.015	.596	.619
Zone * Tillage	.132	6	.022	.860	.527
Zone * Cropping	.240	9	.027	1.047	.409
Tillage * Cropping	.150	6	.025	.980	.443
Zone * Tillage * Cropping	.820	18	.046	1.787	.038
Error	2.446	96	.025		
Total	127.631	144			
Corrected Total	6.588	143			

a. R Squared = .629 (Adjusted R Squared = .447)

A.3. Tillage x cropping system experiment. Analysis of variance for DOC across all zones. Data is log transformed.

Source	Type III Sum of Squares	df	Mean Square	F	Sig.
Corrected Model	2.980 ^a	47	.063	6.385	.000
Intercept	600.036	1	600.036	60430.823	.000
Zone	2.581	3	.860	86.634	.000
Tillage	.033	2	.016	1.659	.196
Cropping	.008	3	.003	.266	.850
Zone * Tillage	.014	6	.002	.230	.966
Zone * Cropping	.136	9	.015	1.524	.150
Tillage * Cropping	.061	6	.010	1.030	.411
Zone * Tillage * Cropping	.147	18	.008	.822	.670
Error	.953	96	.010		
Total	603.969	144			
Corrected Total	3.933	143			

a. R Squared = .758 (Adjusted R Squared = .639)

A.4. Tillage x cropping system experiment. Analysis of variance for TEN across all zones. Data is log transformed.

Source	Type III Sum of Squares	df	Mean Square	F	Sig.
Corrected Model	5.124 ^a	47	.109	3.940	.000
Intercept	184.729	1	184.729	6675.448	.000
Zone	3.253	3	1.084	39.180	.000
Tillage	.032	2	.016	.579	.563
Cropping	.078	3	.026	.936	.426
Zone * Tillage	.121	6	.020	.731	.626
Zone * Cropping	.467	9	.052	1.874	.065
Tillage * Cropping	.234	6	.039	1.412	.218
Zone * Tillage * Cropping	.939	18	.052	1.885	.026
Error	2.657	96	.028		
Total	192.510	144			
Corrected Total	7.781	143			

a. R Squared = .659 (Adjusted R Squared = .491)

A.5. Tillage x cropping system experiment. Analysis of variance for EON across all zones. Data is log transformed.

Source	Type III Sum of Squares	df	Mean Square	F	Sig.
Corrected Model	4.785 ^a	16	.299	2.957	.011
Intercept	.562	1	.562	5.558	.028
Zone	2.365	2	1.182	11.692	.000
Tillage	.740	2	.370	3.659	.043
Cropping	.842	3	.281	2.775	.067
Zone * Tillage	.491	2	.246	2.429	.112
Zone * Cropping	0.000	0			
Tillage * Cropping	.484	6	.081	.798	.582
Zone * Tillage * Cropping	0.000	0			
Error	2.124	21	.101		
Total	15.969	38			
Corrected Total	6.909	37			

a. R Squared = .693 (Adjusted R Squared = .458)

A.6. Tillage x cropping system experiment. Analysis of variance for PO₄P across all zones. Data is log transformed.

Source	Type III Sum of Squares	df	Mean Square	F	Sig.
Corrected Model	25.989 ^a	47	.553	4.532	.000
Intercept	.467	1	.467	3.829	.053
Zone	17.210	3	5.737	47.015	.000
Tillage	.934	2	.467	3.828	.025
Cropping	.409	3	.136	1.116	.346
Zone * Tillage	.637	6	.106	.870	.520
Zone * Cropping	2.129	9	.237	1.939	.055
Tillage * Cropping	2.231	6	.372	3.048	.009
Zone * Tillage * Cropping	2.439	18	.135	1.110	.354
Error	11.714	96	.122		
Total	38.170	144			
Corrected Total	37.703	143			

a. R Squared = .689 (Adjusted R Squared = .537)

A.7 Within-zone analysis of variance for NO₃-N in the Coastal Savannah.
Data log transformed

Source	Type III Sum of Squares	df	Mean Square	F	Sig.
Corrected Model	.323 ^a	11	.029	2.150	.057
Intercept	27.792	1	27.792	2032.190	.000
Tillage	.016	2	.008	.595	.559
Cropping	.221	3	.074	5.381	.006
Tillage * Cropping	.086	6	.014	1.053	.417
Error	.328	24	.014		
Total	28.444	36			
Corrected Total	.652	35			

a. R Squared = .496 (Adjusted R Squared = .266)

A.8 Within-zone analysis of variance for NH₄-N in the Coastal Savannah. Data log transformed.

Source	Type III Sum of Squares	df	Mean Square	F	Sig.
Corrected Model	.672 ^a	11	.061	2.131	.059
Intercept	46.375	1	46.375	1618.315	.000
Tillage	.110	2	.055	1.925	.168
Cropping	.101	3	.034	1.180	.338
Tillage * Cropping	.460	6	.077	2.676	.039
Error	.688	24	.029		
Total	47.735	36			
Corrected Total	1.360	35			

a. R Squared = .494 (Adjusted R Squared = .262)

A.9 Within-zone analysis of variance for TDN in the Coastal Savannah.
Data log transformed.

Source	Type III Sum of Squares	df	Mean Square	F	Sig.
Corrected Model	.774 ^a	11	.070	2.270	.045
Intercept	53.273	1	53.273	1718.670	.000
Tillage	.057	2	.028	.918	.413
Cropping	.104	3	.035	1.118	.361
Tillage * Cropping	.613	6	.102	3.297	.016
Error	.744	24	.031		
Total	54.791	36			
Corrected Total	1.518	35			

a. R Squared = .510 (Adjusted R Squared = .285)

A.10 Within-zone analysis of variance for NO₃-N in the Transition. Data log transformed.

Source	Type III Sum of Squares	df	Mean Square	F	Sig.
Corrected Model	.094 ^a	11	.009	1.835	.104
Intercept	10.718	1	10.718	2289.561	.000
Tillage	.025	2	.012	2.656	.091
Cropping	.051	3	.017	3.663	.026
Tillage * Cropping	.018	6	.003	.647	.692
Error	.112	24	.005		
Total	10.925	36			
Corrected Total	.207	35			

a. R Squared = .457 (Adjusted R Squared = .208)

A.11. Within-zone analysis of variance for PO₄P in the Transition. Data log transformed.

Source	Type III Sum of Squares	df	Mean Square	F	Sig.
Corrected Model	2.662 ^a	11	.242	3.353	.006
Intercept	3.747	1	3.747	51.925	.000
Tillage	.710	2	.355	4.919	.016
Cropping	.830	3	.277	3.836	.022
Tillage * Cropping	1.121	6	.187	2.589	.044
Error	1.732	24	.072		
Total	8.141	36			
Corrected Total	4.394	35			

a. R Squared = .606 (Adjusted R Squared = .425)

A.12. Within-zone analysis of variance for NO₃-N in the Guinea Savannah. Data log transformed.

Source	Type III Sum of Squares	df	Mean Square	F	Sig.
Corrected Model	.504 ^a	11	.046	2.244	.047
Intercept	45.782	1	45.782	2243.485	.000
Tillage	.011	2	.006	.277	.760
Cropping	.111	3	.037	1.818	.171
Tillage * Cropping	.381	6	.064	3.113	.021
Error	.490	24	.020		
Total	46.775	36			
Corrected Total	.993	35			

A.13. Within-zone analysis of variance for PO₄P in the Guinea Savannah. Data log transformed.

Source	Type III Sum of Squares	df	Mean Square	F	Sig.
Corrected Model	3.059 ^a	11	.278	3.118	.010
Intercept	7.118	1	7.118	79.809	.000
Tillage	.377	2	.189	2.115	.143
Cropping	.809	3	.270	3.024	.049
Tillage * Cropping	1.873	6	.312	3.500	.012
Error	2.140	24	.089		
Total	12.318	36			
Corrected Total	5.200	35			

a. R Squared = .588 (Adjusted R Squared = .400)

A.14. Fertilizer experiment. Analysis of variance for NO₃-N across all zones. Data is log transformed.

Source	Type III Sum of Squares	df	Mean Square	F	Sig.
Corrected Model	9.626 ^a	59	.163	9.166	.000
Intercept	88.262	1	88.262	4958.438	.000
Zone	6.160	3	2.053	115.352	.000
TSP	.099	2	.050	2.787	.065
Urea	.571	2	.285	16.033	.000
Compost	.354	2	.177	9.931	.000
Zone * TSP	.215	6	.036	2.010	.067
Zone * Urea	.163	6	.027	1.530	.172
Zone * Compost	.189	6	.031	1.768	.109
TSP * Urea	.021	4	.005	.296	.880
TSP * Compost	.099	4	.025	1.393	.239
Urea * Compost	0.000	0			
Zone * TSP * Urea	.171	12	.014	.799	.652
Zone * TSP * Compost	.154	12	.013	.720	.730
Zone * Urea * Compost	0.000	0			
TSP * Urea * Compost	0.000	0			
Zone * TSP * Urea * Compost	0.000	0			
Error	2.777	156	.018		
Total	133.828	216			
Corrected Total	12.403	215			

a. R Squared = .776 (Adjusted R Squared = .691)

A.15. Fertilizer experiment. Analysis of variance for NH₄-N across all zones. Data is log transformed.

Source	Type III Sum of Squares	df	Mean Square	F	Sig.
Corrected Model	2.113 ^a	59.0000	0.0358	2.954	.000
Intercept	106.387	1.0000	106.3873	8776.208	.000
Zone	.703	3.0000	0.2344	19.335	.000
TSP	.007	2.0000	0.0036	.297	.744
Urea	.013	2.0000	0.0067	.553	.576
Compost	.008	2.0000	0.0038	.315	.730
Zone * TSP	.098	6.0000	0.0163	1.348	.239
Zone * Urea	.115	6.0000	0.0192	1.586	.155
Zone * Compost	.037	6.0000	0.0062	.509	.801
TSP * Urea	.027	4.0000	0.0068	.561	.691
TSP * Compost	.065	4.0000	0.0162	1.337	.259
Urea * Compost	0.000	0.0000			
Zone * TSP * Urea	.252	12.0000	0.0210	1.735	.064
Zone * TSP * Compost	.211	12.0000	0.0176	1.450	.149
Zone * Urea * Compost	0.000	0.0000			
TSP * Urea * Compost	0.000	0.0000			
Zone * TSP * Urea * Compost	0.000	0.0000			
Error	1.891	156.0000	0.0121		
Total	162.666	216.0000			
Corrected Total	4.004	215.0000			

a. R Squared = .528 (Adjusted R Squared = .349)

A.16. Fertilizer experiment. Analysis of variance for DOC across all zones. Data is log transformed.

Source	Type III Sum of Squares	df	Mean Square	F	Sig.
Corrected Model	5.540 ^a	59	.094	6.835	.000
Intercept	635.921	1	635.921	46287.204	.000
Zone	2.773	3	.924	67.268	.000
TSP	.075	2	.038	2.748	.067
Urea	.020	2	.010	.721	.488
Compost	.112	2	.056	4.082	.019
Zone * TSP	.079	6	.013	.959	.455
Zone * Urea	.050	6	.008	.604	.727
Zone * Compost	.152	6	.025	1.839	.095
TSP * Urea	.025	4	.006	.461	.765
TSP * Compost	.089	4	.022	1.622	.171
Urea * Compost	0.000	0			
Zone * TSP * Urea	.054	12	.004	.327	.983
Zone * TSP * Compost	.226	12	.019	1.370	.185
Zone * Urea * Compost	0.000	0			
TSP * Urea * Compost	0.000	0			
Zone * TSP * Urea * Compost	0.000	0			
Error	2.143	156	.014		
Total	953.787	216			
Corrected Total	7.684	215			

a. R Squared = .776 (Adjusted R Squared = .691)

A.17. Fertilizer experiment. Analysis of variance for TDN across all zones. Data is log transformed.

Source	Type III Sum of Squares	df	Mean Square	F	Sig.
Corrected Model	3.443 ^a	59	.058	2.985	.000
Intercept	178.623	1	178.623	9136.578	.000
Zone	1.157	3	.386	19.729	.000
TSP	.025	2	.013	.645	.526
Urea	.088	2	.044	2.254	.108
Compost	.260	2	.130	6.641	.002
Zone * TSP	.181	6	.030	1.539	.169
Zone * Urea	.235	6	.039	2.004	.068
Zone * Compost	.132	6	.022	1.127	.349
TSP * Urea	.093	4	.023	1.190	.317
TSP * Compost	.038	4	.009	.481	.750
Urea * Compost	0.000	0			
Zone * TSP * Urea	.296	12	.025	1.263	.246
Zone * TSP * Compost	.209	12	.017	.890	.559
Zone * Urea * Compost	0.000	0			
TSP * Urea * Compost	0.000	0			
Zone * TSP * Urea * Compost	0.000	0			
Error	3.050	156	.020		
Total	264.873	216			
Corrected Total	6.493	215			

a. R Squared = .530 (Adjusted R Squared = .353)

A.18. Fertilizer experiment. Analysis of variance for DON across all zones. Data is log transformed.

Source	Type III Sum of Squares	df	Mean Square	F	Sig.
Corrected Model	17.683 ^a	30	.589	2.817	.002
Intercept	.009	1	.009	.045	.834
Zone	5.858	3	1.953	9.334	.000
TSP	.920	2	.460	2.199	.125
Urea	.320	2	.160	.765	.472
Compost	.765	2	.382	1.827	.175
Zone * TSP	1.530	3	.510	2.438	.080
Zone * Urea	.120	2	.060	.286	.753
Zone * Compost	.176	2	.088	.420	.660
TSP * Urea	.608	4	.152	.727	.579
TSP * Compost	1.079	4	.270	1.290	.292
Urea * Compost	0.000	0			
Zone * TSP * Urea	.007	1	.007	.034	.855
Zone * TSP * Compost	.920	3	.307	1.465	.240
Zone * Urea * Compost	0.000	0			
TSP * Urea * Compost	0.000	0			
Zone * TSP * Urea * Compost	0.000	0			
Error	7.741	37	.209		
Total	34.135	68			
Corrected Total	25.424	67			

a. R Squared = .696 (Adjusted R Squared = .449)

A.19. Fertilizer experiment. Analysis of variance for PO₄P across all zones. Data is log transformed.

Source	Type III Sum of Squares	df	Mean Square	F	Sig.
Corrected Model	41.325 ^a	59	.700	5.344	.000
Intercept	37.459	1	37.459	285.801	.000
Zone	17.248	3	5.749	43.865	.000
TSP	1.363	2	.681	5.199	.007
Urea	.188	2	.094	.718	.489
Compost	2.653	2	1.326	10.121	.000
Zone * TSP	.908	6	.151	1.154	.334
Zone * Urea	1.414	6	.236	1.798	.103
Zone * Compost	1.816	6	.303	2.309	.037
TSP * Urea	.127	4	.032	.242	.914
TSP * Compost	.727	4	.182	1.387	.241
Urea * Compost	0.000	0			
Zone * TSP * Urea	1.127	12	.094	.716	.734
Zone * TSP * Compost	1.120	12	.093	.712	.738
Zone * Urea * Compost	0.000	0			
TSP * Urea * Compost	0.000	0			
Zone * TSP * Urea * Compost	0.000	0			
Error	20.446	156	.131		
Total	112.344	216			
Corrected Total	61.771	215			

a. R Squared = .669 (Adjusted R Squared = .544)

A.20. Fertilizer experiment. Within-zone analysis of variance for NO₃-N in the Coastal Savannah. Data log transformed.

Source	Type III Sum of Squares	df	Mean Square	F	Sig.
Corrected Model	.568 ^a	14	.041	1.465	.171
Intercept	35.859	1	35.859	1294.773	.000
TSP	.194	2	.097	3.497	.040
Urea	.269	2	.134	4.853	.013
Compost	.139	2	.070	2.513	.094
TSP * Urea	.040	4	.010	.358	.837
TSP * Compost	.108	4	.027	.975	.432
Urea * Compost	0.000	0			
TSP * Urea * Compost	0.000	0			
Error	1.080	39	.028		
Total	50.634	54			
Corrected Total	1.648	53			

a. R Squared = .345 (Adjusted R Squared = .109)

A.21. Fertilizer experiment. Within-zone analysis of variance for NH₄-N in the Coastal Savannah. Data log transformed.

Source	Type III Sum of Squares	df	Mean Square	F	Sig.
Corrected Model	.195 ^a	14	.014	1.085	.400
Intercept	23.852	1	23.852	1857.790	.000
TSP	.048	2	.024	1.859	.169
Urea	.069	2	.034	2.679	.081
Compost	.007	2	.004	.284	.755
TSP * Urea	.081	4	.020	1.571	.201
TSP * Compost	.010	4	.002	.190	.942
Urea * Compost	0.000	0			
TSP * Urea * Compost	0.000	0			
Error	.501	39	.013		
Total	34.857	54			
Corrected Total	.696	53			

a. R Squared = .280 (Adjusted R Squared = .022)

A.22. Fertilizer experiment. Within-zone analysis of variance for DOC in the Coastal Savannah. Data log transformed.

Source	Type III Sum of Squares	df	Mean Square	F	Sig.
Corrected Model	.132 ^a	14	.009	1.199	.315
Intercept	156.880	1	156.880	19873.189	.000
TSP	.018	2	.009	1.126	.335
Urea	.009	2	.004	.552	.580
Compost	.080	2	.040	5.038	.011
TSP * Urea	.015	4	.004	.471	.757
TSP * Compost	.013	4	.003	.396	.810
Urea * Compost	0.000	0			
TSP * Urea * Compost	0.000	0			
Error	.308	39	.008		
Total	231.590	54			
Corrected Total	.440	53			

a. R Squared = .301 (Adjusted R Squared = .050)

A.23. Fertilizer experiment. Within-zone analysis of variance for TDN in the Coastal Savannah. Data log transformed.

Source	Type III Sum of Squares	df	Mean Square	F	Sig.
Corrected Model	.435 ^a	14	.031	1.427	.187
Intercept	47.074	1	47.074	2163.611	.000
TSP	.088	2	.044	2.020	.146
Urea	.200	2	.100	4.596	.016
Compost	.156	2	.078	3.577	.037
TSP * Urea	.086	4	.021	.985	.427
TSP * Compost	.048	4	.012	.555	.696
Urea * Compost	0.000	0			
TSP * Urea * Compost	0.000	0			
Error	.849	39	.022		
Total	66.735	54			
Corrected Total	1.283	53			

a. R Squared = .339 (Adjusted R Squared = .101)

A.23. Fertilizer experiment. Within-zone analysis of variance for PO₄P in the Coastal Savannah. Data log transformed.

Source	Type III Sum of Squares	df	Mean Square	F	Sig.
Corrected Model	5.705 ^a	14	.408	2.939	.004
Intercept	3.605	1	3.605	25.997	.000
TSP	1.057	2	.529	3.813	.031
Urea	.379	2	.189	1.366	.267
Compost	2.968	2	1.484	10.703	.000
TSP * Urea	.203	4	.051	.366	.831
TSP * Compost	.405	4	.101	.729	.577
Urea * Compost	0.000	0			
TSP * Urea * Compost	0.000	0			
Error	5.408	39	.139		
Total	13.606	54			
Corrected Total	11.113	53			

a. R Squared = .513 (Adjusted R Squared = .339)

A.24. Fertilizer experiment. Within-zone analysis of variance for DOC in the Forest. Data log transformed.

Source	Type III Sum of Squares	df	Mean Square	F	Sig.
Corrected Model	.166 ^a	14	.012	1.555	.137
Intercept	195.784	1	195.784	25615.232	.000
TSP	.049	2	.025	3.225	.051
Urea	.034	2	.017	2.215	.123
Compost	.005	2	.002	.322	.727
TSP * Urea	.032	4	.008	1.043	.398
TSP * Compost	.039	4	.010	1.290	.291
Urea * Compost	0.000	0			
TSP * Urea * Compost	0.000	0			
Error	.298	39	.008		
Total	296.186	54			
Corrected Total	.464	53			

a. R Squared = .358 (Adjusted R Squared = .128)

A.25. Fertilizer experiment. Within-zone analysis of variance for NO₃-N in the Transition. Data log transformed.

Source	Type III Sum of Squares	df	Mean Square	F	Sig.
Corrected Model	.146 ^a	14	.010	1.223	.299
Intercept	10.428	1	10.428	1221.659	.000
TSP	.012	2	.006	.678	.513
Urea	.070	2	.035	4.097	.024
Compost	.048	2	.024	2.832	.071
TSP * Urea	.018	4	.004	.525	.718
TSP * Compost	.019	4	.005	.566	.689
Urea * Compost	0.000	0			
TSP * Urea * Compost	0.000	0			
Error	.333	39	.009		
Total	14.883	54			
Corrected Total	.479	53			

a. R Squared = .305 (Adjusted R Squared = .056)

A.26. Fertilizer experiment. Within-zone analysis of variance for DOC in the Transition. Data log transformed.

Source	Type III Sum of Squares	df	Mean Square	F	Sig.
Corrected Model	.132 ^a	14	.009	1.993	.045
Intercept	144.364	1	144.364	30568.351	.000
TSP	.014	2	.007	1.451	.247
Urea	.015	2	.008	1.605	.214
Compost	.081	2	.041	8.582	.001
TSP * Urea	.006	4	.001	.295	.880
TSP * Compost	.012	4	.003	.656	.626
Urea * Compost	0.000	0			
TSP * Urea * Compost	0.000	0			
Error	.184	39	.005		
Total	212.056	54			
Corrected Total	.316	53			

a. R Squared = .417 (Adjusted R Squared = .208)

A.27. Fertilizer experiment. Within-zone analysis of variance for PO₄P in the Transition. Data log transformed.

Source	Type III Sum of Squares	df	Mean Square	F	Sig.
Corrected Model	3.554 ^a	14	.254	2.322	.019
Intercept	16.305	1	16.305	149.104	.000
TSP	.906	2	.453	4.141	.023
Urea	.328	2	.164	1.498	.236
Compost	.641	2	.321	2.932	.065
TSP * Urea	.493	4	.123	1.128	.358
TSP * Compost	.929	4	.232	2.124	.096
Urea * Compost	0.000	0			
TSP * Urea * Compost	0.000	0			
Error	4.265	39	.109		
Total	28.361	54			
Corrected Total	7.819	53			

a. R Squared = .455 (Adjusted R Squared = .259)

A.28. Fertilizer experiment. Within-zone analysis of variance for NO₃-N in the Guinea Savannah. Data log transformed.

Source	Type III Sum of Squares	df	Mean Square	F	Sig.
Corrected Model	.568 ^a	14	.041	1.465	.171
Intercept	35.859	1	35.859	1294.773	.000
TSP	.194	2	.097	3.497	.040
Urea	.269	2	.134	4.853	.013
Compost	.139	2	.070	2.513	.094
TSP * Urea	.040	4	.010	.358	.837
TSP * Compost	.108	4	.027	.975	.432
Urea * Compost	0.000	0			
TSP * Urea * Compost	0.000	0			
Error	1.080	39	.028		
Total	50.634	54			
Corrected Total	1.648	53			

a. R Squared = .345 (Adjusted R Squared = .109)

A.29. Fertilizer experiment. Within-zone analysis of variance for DOC in the Guinea Savannah. Data log transformed.

Source	Type III Sum of Squares	df	Mean Square	F	Sig.
Corrected Model	.132 ^a	14	.009	1.199	.315
Intercept	156.880	1	156.880	19873.189	.000
TSP	.018	2	.009	1.126	.335
Urea	.009	2	.004	.552	.580
Compost	.080	2	.040	5.038	.011
TSP * Urea	.015	4	.004	.471	.757
TSP * Compost	.013	4	.003	.396	.810
Urea * Compost	0.000	0			
TSP * Urea * Compost	0.000	0			
Error	.308	39	.008		
Total	231.590	54			
Corrected Total	.440	53			

a. R Squared = .301 (Adjusted R Squared = .050)

A.29. Fertilizer experiment. Within-zone analysis of variance for PO₄P in the Guinea Savannah. Data log transformed.

Source	Type III Sum of Squares	df	Mean Square	F	Sig.
Corrected Model	5.705 ^a	14	.408	2.939	.004
Intercept	3.605	1	3.605	25.997	.000
TSP	1.057	2	.529	3.813	.031
Urea	.379	2	.189	1.366	.267
Compost	2.968	2	1.484	10.703	.000
TSP * Urea	.203	4	.051	.366	.831
TSP * Compost	.405	4	.101	.729	.577
Urea * Compost	0.000	0			
TSP * Urea * Compost	0.000	0			
Error	5.408	39	.139		
Total	13.606	54			
Corrected Total	11.113	53			

a. R Squared = .513 (Adjusted R Squared = .339)

APPENDIX B

RAW DATA FOR TILLAGE AND CROPPING SYSTEM EXPERIMENT

Soil ID	Sample Code	Main Plot		Rep	Sub Plot	NO ₃ -N	NH ₄ -N	DOC	TDN	DON	PO ₄ -P
		Plot	Zone								
S05828	1-I-122	122	1	1	1	5.87	7.43	116.79	11.48	0.00	1.36
S05690	1-I-221	221	1	2	1	8.58	19.07	84.95	19.39	0.00	3.45
S05723	1-I-323	323	1	3	1	11.09	6.42	78.95	9.24	0.00	11.92
S05675	1-I-124	124	1	1	1	6.74	11.97	121.40	15.81	0.00	0.65
S05855	1-I-223	223	1	2	1	7.06	11.12	95.34	15.47	0.00	0.71
S05781	1-I-322	322	1	3	1	5.40	13.18	113.31	21.63	3.04	18.40
S05800	1-I-123	123	1	1	1	16.26	12.92	95.33	23.74	0.00	0.97
S05865	1-I-222	222	1	2	1	5.49	22.24	89.01	23.41	0.00	9.03
S05801	1-I-324	324	1	3	1	5.69	19.59	57.72	15.09	0.00	2.23
S05778	1-I-121	121	1	1	1	6.78	8.68	102.03	14.11	0.00	7.30
S05688	1-I-224	224	1	2	1	6.37	10.74	77.40	12.68	0.00	0.90
S05794	1-I-321	321	1	3	1	5.27	6.65	113.43	9.24	0.00	1.44
S05795	1-I-134	134	1	1	2	9.50	27.71	96.10	33.47	0.00	11.65
S05726	1-I-234	234	1	2	2	11.54	26.57	83.77	27.57	0.00	1.11
S05850	1-I-314	314	1	3	2	13.32	40.60	109.73	50.13	0.00	1.63
S05864	1-I-131	131	1	1	2	5.97	10.14	146.65	13.87	0.00	0.48
S05830	1-I-233	233	1	2	2	6.65	9.03	62.82	7.35	0.00	1.27
S05842	1-I-312	312	1	3	2	6.26	16.45	65.39	13.59	0.00	0.53
S05861	1-I-132	132	1	1	2	4.80	9.76	38.67	4.98	0.00	3.30
S05717	1-I-232	232	1	2	2	6.85	13.45	73.78	11.66	0.00	0.36
S05856	1-I-311	311	1	3	2	8.71	12.02	79.68	14.20	0.00	1.35
S05847	1-I-133	133	1	1	2	7.59	27.55	103.40	35.33	0.20	1.40

S05858	1-I-231	231	1	2	2	4	7.12	8.84	77.43	9.92	0.00	1.28
S05784	1-I-313	313	1	3	2	4	6.54	13.14	75.04	12.08	0.00	8.88
S05846	1-I-114	114	1	1	3	1	13.21	9.72	104.52	21.45	0.00	3.29
S05708	1-I-212	212	1	2	3	1	10.47	19.74	81.07	28.03	0.00	0.65
S05834	1-I-331	331	1	3	3	1	10.80	12.56	51.46	10.66	0.00	1.09
S05845	1-I-112	112	1	1	3	2	7.91	13.23	83.27	17.89	0.00	3.90
S05831	1-I-213	213	1	2	3	2	8.31	21.90	86.97	27.91	0.00	0.39
S05770	1-I-333	333	1	3	3	2	4.18	7.39	144.60	12.01	0.44	0.94
S05769	1-I-113	113	1	1	3	3	5.80	17.74	67.29	19.33	0.00	4.75
S05757	1-I-214	214	1	2	3	3	7.25	25.64	96.92	30.63	0.00	0.74
S05853	1-I-334	334	1	3	3	3	5.61	8.78	85.28	12.27	0.00	7.31
S05697	1-I-111	111	1	1	3	4	11.30	10.28	111.22	16.24	0.00	1.31
S05793	1-I-211	211	1	2	3	4	8.24	18.86	92.23	23.52	0.00	0.67
S05837	1-I-332	332	1	3	3	4	7.76	14.30	110.94	17.26	0.00	0.91
S05829	2-I-122	122	2	1	1	1	3.10	11.02	138.52	16.08	1.96	0.24
S05849	2-I-221	221	2	2	1	1	4.98	8.88	237.79	21.17	7.32	0.48
S06024	2-I-323	323	2	3	1	1	10.89	6.07	190.65	19.27	2.31	0.34
S05737	2-I-124	124	2	1	1	2	4.90	8.78	185.23	17.74	4.06	0.49
S05857	2-I-223	223	2	2	1	2	5.89	9.11	166.27	23.82	8.82	0.34
S05851	2-I-322	322	2	3	1	2	5.15	7.38	173.19	14.81	2.29	0.29
S05673	2-I-123	123	2	1	1	3	7.09	16.40	330.99	39.28	15.80	0.72
S05703	2-I-222	222	2	2	1	3	8.50	9.71	251.05	26.19	7.97	0.54
S05838	2-I-324	324	2	3	1	3	4.19	9.94	166.34	16.59	2.46	3.47
S05689	2-I-121	121	2	1	1	4	3.42	8.68	200.62	17.35	5.25	0.27
S05852	2-I-224	224	2	2	1	4	3.37	6.15	130.12	9.11	0.00	0.21
S05844	2-I-321	321	2	3	1	4	3.59	8.18	211.58	17.45	5.68	0.27
S05840	2-I-134	134	2	1	2	1	4.32	7.31	178.13	12.92	1.30	0.23
S05771	2-I-234	234	2	2	2	1	3.13	7.28	172.57	12.85	2.44	0.32

S05802	2-I-314	314	2	3	2	1	3.98	8.48	117.11	12.78	0.32	0.45
S05839	2-I-131	131	2	1	2	2	5.63	5.78	134.49	11.33	0.00	0.21
S05747	2-I-233	233	2	2	2	2	8.69	12.71	366.91	35.05	13.66	1.73
S05685	2-I-312	312	2	3	2	2	3.85	7.17	176.70	14.08	3.06	0.47
S05734	2-I-132	132	2	1	2	3	5.80	7.52	195.24	16.82	3.49	0.26
S05862	2-I-232	232	2	2	2	3	6.60	7.47	82.14	10.02	0.00	0.49
S05750	2-I-311	311	2	3	2	3	3.15	7.76	175.96	14.80	3.90	0.36
S05863	2-I-133	133	2	1	2	4	3.37	8.69	198.18	15.86	3.80	2.51
S05693	2-I-231	231	2	2	2	4	4.69	21.36	247.83	39.53	13.48	0.64
S05835	2-I-313	313	2	3	2	4	2.51	7.06	186.87	12.46	2.89	0.23
S05823	2-I-114	114	2	1	3	1	2.57	6.74	182.31	13.05	3.75	0.22
S05751	2-I-212	212	2	2	3	1	3.71	8.15	186.37	15.66	3.80	0.60
S05843	2-I-331	331	2	3	3	1	8.38	8.68	166.32	18.95	1.90	0.65
S05664	2-I-112	112	2	1	3	2	3.99	7.61	188.88	16.68	5.08	0.42
S05824	2-I-213	213	2	2	3	2	20.15	5.42	242.15	29.45	3.89	0.25
S05789	2-I-333	333	2	3	3	2	2.49	7.60	219.69	15.17	5.08	0.19
S05709	2-I-113	113	2	1	3	3	3.65	8.39	215.66	18.80	6.76	0.53
S05826	2-I-214	214	2	2	3	3	4.94	10.39	203.27	18.62	3.29	0.36
S05691	2-I-334	334	2	3	3	3	5.05	9.98	157.37	18.01	2.99	2.01
S06009	2-I-111	111	2	1	3	4	4.62	14.00	188.19	26.30	7.69	0.27
S05859	2-I-211	211	2	2	3	4	3.09	7.89	161.94	13.42	2.43	0.32
S05780	2-I-332	332	2	3	3	4	6.78	10.27	123.65	13.62	0.00	0.66
S05651	3-I-122	122	3	1	1	1	3.10	5.60	87.62	5.88	0.00	5.35
S05872	3-I-221	221	3	2	1	1	3.13	4.67	84.83	5.67	0.00	1.89
S05934	3-I-323	323	3	3	1	1	3.19	5.84	79.66	6.97	0.00	6.10
S05920	3-I-124	124	3	1	1	2	4.09	7.70	90.41	10.63	0.00	6.28
S05993	3-I-223	223	3	2	1	2	3.36	5.81	88.98	7.14	0.00	19.19
S05991	3-I-322	322	3	3	1	2	3.43	4.40	64.06	3.73	0.00	6.89

S05912	3-I-123	123	3	1	1	3	3.21	4.52	80.89	4.75	0.00	1.14
S05974	3-I-222	222	3	2	1	3	5.73	5.93	114.66	9.92	0.00	0.92
S05890	3-I-324	324	3	3	1	3	4.42	5.44	79.55	6.29	0.00	0.68
S05983	3-I-121	121	3	1	1	4	6.75	4.82	117.32	9.20	0.00	6.80
S05977	3-I-224	224	3	2	1	4	3.34	7.63	93.95	10.44	0.00	5.59
S05882	3-I-321	321	3	3	1	4	3.63	73.53	77.29	74.63	0.00	1.20
S05595	3-I-134	134	3	1	2	1	3.24	6.80	106.36	8.19	0.00	1.60
S05908	3-I-234	234	3	2	2	1	3.18	7.02	84.43	9.87	0.00	2.32
S05874	3-I-314	314	3	3	2	1	3.10	5.96	76.31	6.73	0.00	1.39
S05957	3-I-131	131	3	1	2	2	3.14	5.14	54.67	3.06	0.00	2.44
S06021	3-I-233	233	3	2	2	2	3.08	5.28	97.68	5.86	0.00	3.60
S05899	3-I-312	312	3	3	2	2	3.32	16.20	56.75	12.52	0.00	1.12
S05913	3-I-132	132	3	1	2	3	4.31	5.44	95.00	8.79	0.00	1.32
S05959	3-I-232	232	3	2	2	3	3.83	5.36	80.17	5.37	0.00	0.77
S05922	3-I-311	311	3	3	2	3	3.33	6.29	72.88	6.99	0.00	0.83
S05621	3-I-133	133	3	1	2	4	3.29	4.89	87.94	5.95	0.00	3.37
S05867	3-I-231	231	3	2	2	4	3.05	7.23	85.35	9.90	0.00	1.08
S05643	3-I-313	313	3	3	2	4	3.25	6.01	92.20	7.72	0.00	1.40
S05925	3-I-114	114	3	1	3	1	3.18	6.58	109.59	8.65	0.00	3.95
S05602	3-I-212	212	3	2	3	1	3.35	5.65	85.11	6.23	0.00	1.21
S06008	3-I-331	331	3	3	3	1	3.25	4.53	76.95	4.08	0.00	1.61
S05900	3-I-112	112	3	1	3	2	3.05	4.50	81.78	4.54	0.00	0.79
S05958	3-I-213	213	3	2	3	2	3.40	7.26	63.33	5.30	0.00	5.09
S05984	3-I-333	333	3	3	3	2	3.06	5.06	97.90	5.95	0.00	1.48
S05929	3-I-113	113	3	1	3	3	4.20	7.82	93.11	9.91	0.00	5.30
S05926	3-I-214	214	3	2	3	3	3.42	5.72	97.86	7.65	0.00	1.05
S05893	3-I-334	334	3	3	3	3	4.09	6.31	105.06	9.54	0.00	2.40
S05597	3-I-111	111	3	1	3	4	3.39	7.89	92.10	10.02	0.00	1.05

S05881	3-I-211	211	3	2	3	4	3.81	5.18	89.41	6.24	0.00	1.49
S05871	3-I-332	332	3	3	3	4	2.94	5.73	62.66	7.55	0.00	1.16
S05964	4-I-122	122	4	1	1	1	10.80	6.21	113.14	14.49	0.00	5.30
S05605	4-I-221	221	4	2	1	1	11.03	5.80	110.65	15.00	0.00	7.37
S05971	4-I-323	323	4	3	1	1	11.32	5.90	82.14	12.22	0.00	0.29
S05989	4-I-124	124	4	1	1	2	13.99	5.24	146.03	16.30	0.00	0.46
S05909	4-I-223	223	4	2	1	2	8.88	6.10	124.75	12.49	0.00	0.36
S05927	4-I-322	322	4	3	1	2	7.62	13.28	83.38	17.96	0.00	0.31
S05905	4-I-123	123	4	1	1	3	13.63	5.62	107.99	14.32	0.00	0.26
S05894	4-I-222	222	4	2	1	3	34.25	7.30	126.23	36.42	0.00	0.20
S05661	4-I-324	324	4	3	1	3	30.08	6.01	113.94	20.19	0.00	0.25
S06004	4-I-121	121	4	1	1	4	9.53	4.43	110.79	10.29	0.00	0.23
S05623	4-I-224	224	4	2	1	4	10.24	6.71	89.55	13.84	0.00	0.17
S05654	4-I-321	321	4	3	1	4	12.81	6.90	91.03	14.57	0.00	0.21
S05877	4-I-134	134	4	1	2	1	11.20	5.54	133.70	14.34	0.00	0.43
S05644	4-I-234	234	4	2	2	1	8.55	7.61	92.73	12.65	0.00	0.43
S05653	4-I-314	314	4	3	2	1	8.01	4.88	92.27	12.16	0.00	0.41
S05970	4-I-131	131	4	1	2	2	10.57	4.38	107.82	12.13	0.00	0.27
S05896	4-I-233	233	4	2	2	2	15.58	6.74	91.67	16.02	0.00	0.27
S05901	4-I-312	312	4	3	2	2	11.36	5.72	119.72	13.31	0.00	0.25
S05599	4-I-132	132	4	1	2	3	17.83	6.54	125.20	24.76	0.40	0.28
S05897	4-I-232	232	4	2	2	3	12.88	7.56	100.27	16.19	0.00	0.25
S05988	4-I-311	311	4	3	2	3	19.84	7.80	96.90	19.14	0.00	0.43
S05961	4-I-133	133	4	1	2	4	12.17	5.38	120.67	14.48	0.00	0.39
S05886	4-I-231	231	4	2	2	4	30.89	7.45	109.70	34.14	0.00	0.29
S05603	4-I-313	313	4	3	2	4	10.59	5.39	93.58	11.10	0.00	3.73
S05911	4-I-114	114	4	1	3	1	13.99	10.50	110.42	17.62	0.00	0.34
S05987	4-I-212	212	4	2	3	1	31.16	5.56	115.05	19.37	0.00	0.29

S05892	4-I-331	331	4	3	3	1	13.97	6.04	85.57	13.58	0.00	0.22
S05657	4-I-112	112	4	1	3	2	11.89	5.59	110.30	15.45	0.00	0.17
S05625	4-I-213	213	4	2	3	2	17.34	6.71	92.60	13.39	0.00	0.24
S05620	4-I-333	333	4	3	3	2	17.35	5.13	100.46	18.80	0.00	0.15
S05976	4-I-113	113	4	1	3	3	8.02	4.13	114.61	17.29	5.14	0.54
S05914	4-I-214	214	4	2	3	3	16.13	5.75	109.47	17.04	0.00	0.51
S05876	4-I-334	334	4	3	3	3	12.23	4.32	126.42	17.45	0.91	0.28
S05891	4-I-111	111	4	1	3	4	9.39	11.65	124.73	20.27	0.00	0.28
S05626	4-I-211	211	4	2	3	4	16.28	4.92	124.98	15.98	0.00	0.14
S05622	4-I-332	332	4	3	3	4	12.87	5.41	90.07	12.27	0.00	0.23

APPENDIX C

RAW DATA FOR FERTILITY EXPERIMENT

Soil ID	Sample Name	Plot	Zone	Main Plot	Sub Plot	NO ₃ -N	NH ₄ -N	DOC	TDN	DON	PO ₄ -P	
				Phosphorus	Nitrogen							Compost
Soil extractable mass (mg/kg)												
S06023	ExII 1-136	136	1	0	0	0	6.45	11.23	69.85	13.39	0.00	11.54
S05731	ExII 1-232	232	1	0	0	0	6.98	14.96	97.10	19.37	0.00	3.91
S05674	ExII 1-316	316	1	0	0	0	10.00	9.03	101.94	14.93	0.00	23.87
S05732	ExII 1-131	131	1	0	0	0	6.86	11.59	101.76	13.93	0.00	22.21
S05785	ExII 1-235	235	1	0	0	0	8.10	9.23	103.90	12.76	0.00	4.81
S05808	ExII 1-311	311	1	0	0	0	7.46	12.61	89.93	14.64	0.00	9.50
S05804	ExII 1-135	135	1	0	70	0	8.10	5.20	69.25	5.72	0.00	13.47
S05812	ExII 1-233	233	1	0	70	0	20.59	10.61	75.35	20.12	0.00	6.98
S05699	ExII 1-312	312	1	0	70	0	10.24	5.82	144.94	17.23	0.00	17.01
S05803	ExII 1-133	133	1	0	140	0	12.16	6.97	99.19	13.91	0.00	17.46
S05813	ExII 1-231	231	1	0	140	0	4.34	4.78	116.74	6.06	0.00	7.53
S05819	ExII 1-313	313	1	0	140	0	10.20	7.42	74.33	9.84	0.00	3.17
S05817	ExII 1-134	134	1	0	0	3	9.09	9.67	94.29	14.87	0.00	7.77
S05764	ExII 1-236	236	1	0	0	3	8.02	9.97	105.60	14.23	0.00	4.02
S05676	ExII 1-315	315	1	0	0	3	8.24	11.41	93.21	11.15	0.00	13.87
S05779	ExII 1-132	132	1	0	0	6	14.87	10.27	118.48	20.83	0.00	15.21
S05810	ExII 1-314	314	1	0	0	6	14.14	11.71	89.00	15.43	0.00	10.74
S05702	ExII 1-234	234	1	0	0	6	15.98	11.54	109.61	19.83	0.00	21.41
S05822	ExII 1-116	116	1	20	0	0	6.36	7.81	89.71	8.97	0.00	5.73
S05721	ExII 1-225	225	1	20	0	0	5.37	6.33	105.26	10.63	0.00	11.44
S05694	ExII 1-332	332	1	20	0	0	4.75	7.68	113.51	11.16	0.00	4.83
S05807	ExII 1-115	115	1	20	0	0	6.86	5.40	108.40	8.75	0.00	6.79

S05782	ExII 1-224	224	1	20	0	0	6.19	6.28	117.88	10.16	0.00	21.57
S05790	ExII 1-334	334	1	20	0	0	5.87	6.39	79.29	7.78	0.00	7.08
S05733	ExII 1-113	113	1	20	70	0	9.29	9.56	121.89	16.73	0.00	11.53
S05730	ExII 1-222	222	1	20	70	0	8.78	6.96	96.09	13.61	0.00	17.47
S05679	ExII 1-336	336	1	20	70	0	18.87	10.71	88.26	22.04	0.00	3.09
S05724	ExII 1-114	114	1	20	140	0	8.93	8.55	86.58	10.49	0.00	6.18
S05787	ExII 1-223	223	1	20	140	0	11.82	6.76	109.08	15.81	0.00	12.48
S05696	ExII 1-331	331	1	20	140	0	7.13	8.43	97.34	12.94	0.00	2.15
S05738	ExII 1-112	112	1	20	0	3	7.50	7.78	116.25	13.59	0.00	13.31
S05820	ExII 1-221	221	1	20	0	3	5.74	7.65	6.69	7.79	0.00	12.44
S05833	ExII 1-335	335	1	20	0	3	4.20	4.76	118.28	9.05	0.09	4.61
S05742	ExII 1-111	111	1	20	0	6	13.13	23.46	120.13	32.88	0.00	16.09
S05739	ExII 1-226	226	1	20	0	6	7.84	7.10	118.25	10.82	0.00	9.49
S05765	ExII 1-333	333	1	20	0	6	14.06	8.95	106.35	17.51	0.00	52.20
S05761	ExII 1-121	121	1	40	0	0	8.66	14.17	80.94	15.53	0.00	15.88
S05774	ExII 1-211	211	1	40	0	0	8.11	6.40	121.75	11.28	0.00	17.58
S05743	ExII 1-324	324	1	40	0	0	6.16	8.30	77.59	9.79	0.00	4.77
S05695	ExII 1-126	126	1	40	0	0	5.26	6.78	119.02	11.57	0.00	40.40
S05706	ExII 1-215	215	1	40	0	0	6.08	11.95	129.29	16.52	0.00	12.67
S05821	ExII 1-323	323	1	40	0	0	6.12	11.55	50.15	9.29	0.00	47.19
S05672	ExII 1-122	122	1	40	70	0	9.52	17.09	92.28	22.97	0.00	11.27
S05716	ExII 1-214	214	1	40	70	0	10.34	7.97	87.68	11.88	0.00	15.40
S05815	ExII 1-321	321	1	40	70	0	7.81	8.87	82.06	12.48	0.00	2.04
S05736	ExII 1-124	124	1	40	140	0	9.52	8.36	123.82	18.24	0.36	6.55
S05713	ExII 1-213	213	1	40	140	0	14.45	8.81	137.37	21.70	0.00	13.20
S05811	ExII 1-325	325	1	40	140	0	8.50	7.24	103.88	11.96	0.00	5.72
S05680	EXII 1-123	123	1	40	0	3	10.20	28.65	96.17	33.58	0.00	11.52
S05714	ExII 1-212	212	1	40	0	3	4.65	6.93	160.10	11.17	0.00	10.15

S05683	ExII 1-322	322	1	40	0	3	9.12	13.76	98.55	18.25	0.00	8.09
S05809	ExII 1-125	125	1	40	0	6	8.75	6.32	127.83	13.50	0.00	14.31
S05798	ExII 1-216	216	1	40	0	6	5.26	7.25	111.35	11.78	0.00	7.97
S05725	ExII 1-326	326	1	40	0	6	15.16	10.34	111.11	17.79	0.00	16.79
S05763	ExII 2-136	136	2	1	1	4	3.73	9.32	251.17	17.21	4.16	0.54
S05799	ExII 2-232	232	2	1	1	4	5.10	8.73	211.28	16.68	2.85	0.43
S05754	ExII 2-316	316	2	1	1	4	3.86	7.02	273.15	14.79	3.91	1.05
S05727	ExII 2-135	135	2	1	2	4	4.12	8.95	216.47	20.05	6.97	0.55
S05719	ExII 2-233	233	2	1	2	4	4.62	10.01	226.11	25.39	10.77	0.84
S05744	ExII 2-312	312	2	1	2	4	3.90	6.02	126.52	7.44	0.00	0.54
S05692	ExII 2-133	133	2	1	3	4	3.72	9.92	201.25	21.03	7.40	0.61
S05797	ExII 2-231	231	2	1	3	4	6.57	7.70	208.06	18.43	4.15	0.72
S05753	ExII 2-313	313	2	1	3	4	5.05	9.49	146.25	8.52	0.00	0.52
S05663	ExII 2-131	131	2	1	1	4	3.04	8.39	196.88	17.51	6.08	0.53
S05786	ExII 2-235	235	2	1	1	4	3.46	10.61	225.86	17.47	3.39	0.46
S05832	ExII 2-311	311	2	1	1	4	3.42	9.20	241.83	19.48	6.86	4.63
S05666	ExII 2-134	134	2	1	1	5	3.98	9.20	200.88	13.49	0.31	0.99
S05735	ExII 2-236	236	2	1	1	5	4.47	8.23	188.34	12.23	0.00	0.56
S05684	ExII 2-315	315	2	1	1	5	3.44	6.78	194.26	13.38	3.17	1.52
S05816	ExII 2-132	132	2	1	1	6	4.14	5.62	232.10	12.61	2.86	4.50
S05745	ExII 2-234	234	2	1	1	6	5.08	8.98	262.07	23.21	9.15	2.55
S05701	ExII 2-314	314	2	1	1	6	3.78	6.80	144.38	9.23	0.00	0.97
S05748	ExII 2-116	116	2	2	1	4	5.85	9.63	208.95	12.78	0.00	1.26
S05705	ExII 2-225	225	2	2	1	4	3.99	8.48	322.22	22.53	10.06	0.67
S05698	ExII 2-332	332	2	2	1	4	4.13	12.82	209.38	21.32	4.37	0.99
S05681	ExII 2-113	113	2	2	2	4	2.49	7.85	222.17	18.33	7.99	0.70
S05814	ExII 2-222	222	2	2	2	4	4.37	7.87	200.48	15.42	3.19	0.63
S05729	ExII 2-336	336	2	2	2	4	2.69	8.22	206.38	17.80	6.88	1.07

S05762	ExII 2-114	114	2	2	3	4	7.31	10.00	287.11	15.44	0.00	1.65
S05788	ExII 2-223	223	2	2	3	4	3.81	7.05	214.74	16.39	5.52	0.74
S05773	ExII 2-331	331	2	2	3	4	3.64	7.22	204.92	13.99	3.12	0.77
S05669	ExII 2-115	115	2	2	1	4	2.24	7.22	213.60	16.97	7.51	1.80
S05805	ExII 2-224	224	2	2	1	4	6.37	7.87	199.97	15.87	1.63	1.07
S05686	ExII 2-334	334	2	2	1	4	3.92	8.62	141.27	10.05	0.00	1.98
S05670	ExII 2-112	112	2	2	1	5	2.56	8.63	233.24	21.35	10.17	0.74
S05752	ExII 2-221	221	2	2	1	5	3.52	7.10	209.38	12.07	1.45	0.76
S05796	ExII 2-335	335	2	2	1	5	4.12	6.92	223.60	15.63	4.59	0.93
S05776	ExII 2-111	111	2	20	0	6	2.42	7.07	198.44	15.75	6.26	2.37
S05841	ExII 2-226	226	2	2	1	6	3.81	15.54	248.72	24.90	5.55	6.48
S05777	ExII 2-333	333	2	2	1	6	4.65	8.21	206.07	15.07	2.21	0.83
S05682	ExII 2-121	121	2	3	1	4	7.19	9.11	251.59	22.85	6.55	8.22
S05707	ExII 2-211	211	2	40	0	0	3.99	10.78	416.20	33.95	19.17	214.43
S05712	ExII 2-324	324	2	3	1	4	2.27	7.27	211.35	13.77	4.23	0.94
S05758	ExII 2-122	122	2	3	2	4	5.72	8.31	196.70	15.21	1.18	0.74
S05700	ExII 2-214	214	2	3	2	4	3.62	6.32	163.73	9.39	0.00	0.62
S05668	ExII 2-321	321	2	3	2	4	4.23	6.60	187.58	10.44	0.00	1.21
S05772	ExII 2-124	124	2	3	3	4	4.64	7.31	216.26	13.98	2.02	1.56
S05783	ExII 2-213	213	2	3	3	4	8.73	8.88	220.88	19.97	2.37	0.58
S05792	ExII 2-325	325	2	3	3	4	3.74	6.71	217.60	13.54	3.09	1.12
S05741	ExII 2-126	126	2	3	1	4	3.57	6.18	205.94	11.39	1.65	0.52
S05678	ExII 2-215	215	2	3	1	4	3.02	8.75	222.19	22.03	10.26	1.17
S05720	ExII 2-323	323	2	3	1	4	2.46	7.44	193.53	15.55	5.65	0.59
S05827	ExII 2-123	123	2	3	1	5	6.74	7.74	310.05	18.58	4.11	1.73
S05746	ExII 2-212	212	2	3	1	5	4.70	8.26	395.57	24.81	11.85	2.22
S05791	ExII 2-322	322	2	3	1	5	3.62	6.38	255.60	14.11	4.12	2.76
S05704	ExII 2-125	125	2	3	1	6	3.53	8.15	274.81	19.20	7.52	4.08

S05677	ExII 2-216	216	2	3	1	6	5.68	8.76	199.10	14.32	0.00	2.19
S05718	ExII 2-326	326	2	3	1	6	2.77	10.37	264.36	25.20	12.07	3.86
S05982	ExII 3-136	136	3	1	1	4	3.29	4.57	66.24	5.44	0.00	0.77
S05879	ExII 3-232	232	3	0	0	0	2.62	7.25	99.71	9.27	0.00	1.83
S05972	ExII 3-316	316	3	1	1	4	4.50	5.16	73.28	5.82	0.00	2.66
S06016	ExII 3-135	135	3	1	2	4	3.46	4.47	78.53	6.56	0.00	3.73
S06011	ExII 3-233	233	3	1	2	4	3.20	5.72	98.95	8.37	0.00	1.55
S05998	ExII 3-312	312	3	1	2	4	4.98	7.91	85.92	6.22	0.00	3.44
S05921	ExII 3-133	133	3	1	3	4	3.33	5.15	89.19	7.47	0.00	1.61
S05907	ExII 3-231	231	3	1	3	4	3.83	5.49	94.02	8.57	0.00	0.97
S05999	ExII 3-313	313	3	1	3	4	3.66	7.81	88.56	10.94	0.00	5.84
S06003	ExII 3-131	131	3	1	1	4	3.05	6.32	88.61	7.74	0.00	0.80
S06015	ExII 3-235	235	3	1	1	4	2.67	5.65	76.98	8.19	0.00	2.02
S05996	ExII 3-311	311	3	1	1	4	2.29	5.71	68.57	5.92	0.00	1.77
S05969	ExII 3-134	134	3	1	1	5	2.46	5.50	126.48	7.72	0.00	6.41
S05869	ExII 3-236	236	3	1	1	5	2.96	5.07	103.89	8.84	0.81	7.22
S05917	ExII 3-315	315	3	1	1	5	2.51	7.06	88.03	9.49	-0.07	1.39
S06005	ExII 3-132	132	3	1	1	6	3.48	6.54	98.72	10.58	0.57	3.37
S05875	ExII 3-234	234	3	1	1	6	4.00	5.33	104.42	9.16	0.00	6.57
S05887	ExII 3-314	314	3	1	1	6	3.45	6.07	85.28	9.12	0.00	3.04
S06006	ExII 3-116	116	3	2	1	4	2.60	3.79	71.32	5.52	0.00	7.04
S05978	ExII 3-225	225	3	20	0	0	5.87	5.78	98.45	8.83	0.00	5.99
S05650	ExII 3-332	332	3	2	1	4	2.62	4.88	94.38	7.28	0.00	1.28
S06020	ExII 3-113	113	3	2	2	4	3.49	6.50	86.64	9.34	0.00	11.52
S06000	ExII 3-222	222	3	20	2	0	3.34	7.26	149.58	12.56	1.96	4.84
S05975	ExII 3-336	336	3	2	2	4	4.21	5.75	88.07	6.33	0.00	2.43
S05968	ExII 3-114	114	3	2	3	4	2.92	5.33	82.53	7.21	0.00	3.94
S05870	ExII 3-223	223	3	2	3	4	3.72	6.69	104.49	16.81	6.39	1.30

S05915	ExII 3-331	331	3	2	3	4	3.60	6.18	118.46	11.14	1.37	3.69
S05659	ExII 3-115	115	3	2	1	4	2.27	5.85	92.81	7.79	0.00	6.31
S05924	ExII 3-224	224	3	2	1	4	3.37	11.68	114.67	21.12	6.07	40.22
S05906	ExII 3-334	334	3	2	1	4	2.33	4.90	77.71	5.53	0.00	5.46
S06014	ExII 3-112	112	3	2	1	5	3.16	7.47	92.45	10.68	0.05	3.92
S05981	ExII 3-221	221	3	2	1	5	2.63	6.67	100.23	10.01	0.71	3.64
S05933	ExII 3-335	335	3	2	1	5	3.25	6.04	116.32	11.92	2.63	3.15
S06018	ExII 3-111	111	3	2	1	6	2.91	4.95	124.99	9.65	1.78	7.93
S05660	ExII 3-226	226	3	20	0	6	3.55	8.09	107.13	12.04	0.39	5.38
S05916	ExII 3-333	333	3	2	1	6	3.42	5.40	102.02	8.37	0.00	12.89
S06002	ExII 3-121	121	3	3	1	4	2.62	4.76	82.04	5.93	0.00	8.32
S06007	ExII 3-211	211	3	3	1	4	2.80	9.73	85.73	17.07	4.54	4.73
S05930	ExII-3-324	324	3	40	0	0	2.58	8.34	90.77	9.33	0.00	6.38
S05967	ExII 3-122	122	3	3	2	4	4.22	4.73	87.08	8.56	0.00	23.22
S05935	ExII 3-214	214	3	3	2	4	3.21	6.61	84.51	9.06	0.00	2.05
S06010	ExII 3-321	321	3	3	2	4	2.60	7.40	99.39	9.69	0.00	15.60
S05889	ExII 3-124	124	3	3	3	4	3.94	4.91	84.81	7.39	0.00	2.80
S05910	ExII 3-213	213	3	3	3	4	4.25	5.06	103.47	5.84	0.00	5.78
S05931	ExII 3-325	325	3	3	3	4	4.22	6.17	99.99	10.48	0.09	3.97
S06012	ExII 3-126	126	3	3	1	4	2.97	5.09	97.02	8.08	0.01	0.69
S06013	ExII 3-215	215	3	3	1	4	3.74	5.11	84.14	4.26	0.00	2.84
S05923	ExII 3-323	323	3	3	1	4	3.02	4.95	106.72	8.25	0.29	3.03
S06017	ExII 3-123	123	3	3	1	5	3.09	4.71	87.17	7.25	0.00	19.17
S05902	ExII 3-212	212	3	3	1	5	2.73	4.92	95.63	8.01	0.35	13.49
S05965	ExII 3-322	322	3	3	1	5	4.42	6.07	137.98	10.80	0.30	11.19
S05903	ExII 3-125	125	3	3	1	6	3.03	5.10	101.82	9.02	0.90	3.27
S05919	ExII 3-216	216	3	3	1	6	5.83	6.14	149.80	11.72	0.00	5.71
S05918	ExII 3-326	326	3	3	1	6	3.50	5.68	126.47	10.70	1.51	13.17

S05960	ExII 4-136	136	4	1	1	4	6.21	6.24	90.50	7.97	0.00	0.22
S05630	ExII 4-232	232	4	1	1	4	6.83	4.65	93.42	7.28	0.00	2.85
S05656	ExII 4-316	316	4	1	1	4	6.96	5.40	131.04	10.40	0.00	2.47
S05868	ExII 4-135	135	4	1	2	4	18.10	31.63	70.22	45.15	0.00	0.29
S05652	ExII 4-233	233	4	1	2	4	7.65	5.92	112.86	10.82	0.00	0.73
S05619	ExII 4-312	312	4	1	2	4	9.99	5.62	124.55	13.83	0.00	0.39
S05596	ExII 4-133	133	4	1	3	4	16.73	7.44	81.22	12.08	0.00	0.77
S05631	ExII 4-231	231	4	1	3	4	9.63	6.48	110.83	13.95	0.00	1.03
S05628	ExII 4-313	313	4	1	3	4	15.87	6.01	133.37	18.98	0.00	0.84
S05635	ExII 4-131	131	4	1	1	4	10.20	5.60	100.62	11.28	0.00	0.49
S05618	ExII 4-235	235	4	1	1	4	5.32	6.00	83.06	7.64	0.00	0.49
S05593	ExII 4-311	311	4	1	1	4	6.91	7.33	159.15	17.15	2.91	0.65
S05624	ExII 4-134	134	4	1	1	5	12.87	9.20	95.28	15.56	0.00	0.70
S05992	ExII 4-236	236	4	1	1	5	6.75	5.84	103.41	8.79	0.00	1.38
S05648	ExII 4-315	315	4	1	1	5	10.82	5.23	142.19	12.65	0.00	2.95
S05594	ExII 4-132	132	4	1	1	6	27.43	6.51	135.76	26.35	0.00	2.69
S05617	ExII 4-234	234	4	1	1	6	9.57	6.60	120.29	14.13	0.00	2.71
S05885	ExII 4-314	314	4	1	1	6	13.03	5.67	173.87	18.49	0.00	7.30
S05607	ExII 4-116	116	4	2	1	4	8.71	5.21	133.85	11.44	0.00	0.87
S05598	ExII 4-225	225	4	2	1	4	5.52	6.04	78.88	7.50	0.00	0.99
S05612	ExII 4-332	332	4	2	1	4	7.04	5.77	119.36	13.54	0.73	0.48
S05629	ExII 4-113	113	4	2	2	4	12.56	5.75	112.85	14.54	0.00	0.60
S05606	ExII 4-222	222	4	2	2	4	13.56	7.32	104.21	16.64	0.00	0.54
S05895	ExII 4-336	336	4	2	2	4	7.84	7.02	127.75	12.62	0.00	11.65
S05658	ExII 4-114	114	4	2	3	4	29.81	6.30	146.37	31.73	0.00	2.14
S05627	ExII 4-223	223	4	2	3	4	7.82	6.17	128.90	12.52	0.00	14.49
S05615	ExII 4-331	331	4	2	3	4	7.41	6.73	121.77	12.59	0.00	0.56
S05604	ExII 4-115	115	4	2	1	4	11.50	5.00	101.56	12.33	0.00	1.49

S05646	ExII 4-224	224	4	2	1	4	10.93	6.23	109.32	13.66	0.00	1.92
S05932	ExII 4-334	334	4	2	1	4	5.94	5.49	90.69	8.43	0.00	1.77
S05616	ExII 4-112	112	4	2	1	5	7.85	5.65	130.90	12.77	0.00	1.37
S05609	ExII 4-221	221	4	2	1	5	12.41	8.36	148.10	15.38	0.00	4.03
S05997	ExII 4-335	335	4	2	1	5	5.96	5.56	105.11	8.20	0.00	1.57
S05601	ExII 4-111	111	4	2	1	6	13.26	7.82	159.98	18.96	0.00	2.66
S05610	ExII 4-226	226	4	2	1	6	8.82	6.42	145.73	14.53	0.00	7.21
S05639	ExII 4-333	333	4	2	1	6	6.00	5.32	107.90	9.17	0.00	6.86
S05878	ExII 4-121	121	4	3	1	4	10.03	5.82	105.83	10.05	0.00	1.32
S05655	ExII 4-211	211	4	3	1	4	9.06	5.61	104.08	12.45	0.00	0.65
S05633	ExII 4-324	324	4	3	1	4	6.38	5.70	97.47	8.73	0.00	3.66
S05662	ExII 4-122	122	4	3	2	4	10.74	5.73	137.71	14.20	0.00	2.45
S05637	ExII 4-214	214	4	3	2	4	6.97	5.46	89.49	8.83	0.00	0.67
S05883	ExII 4-321	321	4	3	2	4	8.39	5.66	118.45	9.41	0.00	0.48
S05973	ExII 4-124	124	4	3	3	4	11.70	7.33	151.23	14.92	0.00	0.42
S05641	ExII 4-213	213	4	3	3	4	11.24	6.66	106.18	14.53	0.00	4.85
S06022	ExII 4-325	325	4	3	3	4	5.33	6.89	94.67	10.69	0.00	3.30
S05873	ExII 4-126	126	4	3	1	4	8.44	5.26	159.33	13.62	0.00	0.99
S05642	ExII 4-215	215	4	3	1	4	4.85	6.37	104.38	8.63	0.00	0.77
S05611	ExII 4-323	323	4	3	1	4	4.80	5.95	111.87	10.21	0.00	1.37
S05632	ExII 4-123	123	4	3	1	5	5.87	4.52	152.33	8.64	0.00	9.75
S05636	ExII 4-212	212	4	3	1	5	9.69	7.02	156.77	16.40	0.00	6.65
S05608	ExII 4-322	322	4	3	1	5	5.77	5.37	123.27	10.55	0.00	6.03
S05962	ExII 4-125	125	4	3	1	6	6.80	5.74	119.92	11.49	0.00	6.91
S05600	ExII 4-216	216	4	3	1	6	14.03	5.90	171.49	20.70	0.77	7.56
S05613	ExII 4-326	326	4	3	1	6	5.77	5.18	126.34	9.45	0.00	3.56