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Carbon, Nitrogen and Phosphorus Sequestration With Soil Aggregates of Maize and Tree Based Agroforestry Systems

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# CARBON, NITROGEN AND PHOSPHORUS SEQUESTRATION WITHIN SOIL AGGREGATES OF MAIZE AND TREE-BASED AGROFORESTRY SYSTEMS

Ву

James M. Kinyangi

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#### **ABSTRACT**

CARBON, NITROGEN AND PHOSPHORUS SEQUESTRATION WITHIN SOIL AGGREGATES OF MAIZE AND TREE-BASED AGROFORESTRY SYSTEMS

By

# James M Kinyangi

Direct contributions by tree crops to the nutrient capital and nutrient cycling on small farm holdings in Sub Saharan Africa have been demonstrated for several soil types. In many ecosystems, carbon appears to be the most limiting factor controlling microbial biomass-mediated nutrient turnover. Soil aggregate sequestration of carbon (C), nitrogen (N) and phosphorus (P) was investigated on a P deficient Oxisol in Western Kenya, during four crop cycles of maize and tree rotations (1997-1999) in an agroforestry management system. Concentric layers of soil aggregrates (4-8 mm across) were removed using stainless steel meso-SAE chambers, developed for separating concentric layers from interior regions by gentle erosion of aggregate surfaces. Anion exchange resins (Cl'), converted to bicarbonate form, were used to evaluate plant available phosphorus supplied by layers removed from soil aggregates. Carbon contributions by tree  $(C_3, \delta^{13}C - 26\%)$  and maize  $(C_4, \delta^{13}C - 10\%)$  crops to specific concentric layers within soil aggregates sampled at the 0-15 cm depth were estimated by changes in their delta <sup>13</sup>C signatures. Total C and N were analyzed by the dry combustion technique. Exterior to interior ratios (EIRs), used to calculate relative locations of plant available P within soil aggregates, showed maize crop removal of plant

available P from the exterior surface layers of soil aggregates when applied fertilizer P was insufficient to meet plant demand. A positive linear relationship  $(r^2 = 0.66)$  was observed between a 9-month change in EIR with July 1999 maize grain yield, suggesting that greater P depletion resulted in increases in maize grain yield of the tree-crop rotation system. Poor plant growth in the continuous maize cropping system resulted in 48% more accumulations of plant available P to surface layers of soil aggregates. Shifting plant inputs of C between C<sub>3</sub> and C<sub>4</sub> sources revealed a pronounced seasonal influx of C<sub>4</sub> carbon accumulations onto surface layers of soil aggregates. The contemporary maize crop contributed as much as 50% of the current carbon onto surface layers in July 1998 (field study) and August 1999 (pot experiment). However, this influx was not apparent when whole aggregate measurements were taken. During the same time period, C contents in surface layers of soil aggregates, 4-8 mm across, declined nearly 23% when fertilizer P was applied to planted tree crops. These results suggest that uniform separation and analyses of soil aggregate layers should be integrated into current research methods to further enhance our understanding of the rapid and short term changes in C and P associated with management modifications in low input crop production systems of sustainable tropical agriculture.

Copyright by James M. Kinyangi 2000 This work is dedicated to my wife Millicent, our children, Sandra, Valessa, Barbara, my parents, brothers and sisters.

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#### INTRODUCTION

Improved tree fallow systems where maize is interplanted with fast growing trees such as *Sesbania*, *Tephrosia*, *Crotalaria* or *Tithonia* promote biomass production and preserve soil resources. Tree fallows are usually untilled for periods from 8-10 months before the woody material is taken off the field while the leaf litterfall is incorporated into the plow layer during land preparation before the next food crop is planted. Tree-crop production systems include:

- a) sequential systems in which trees and crops are either grown in rotation, or maximum growth rates of the tree and crop components occur at different points in time, where competition for growth resources is absent or minimal (Rao et al., 1997), and
- b) simultaneous systems in which trees and crops are grown together.
   These are fast becoming an important part of cropping systems in western Kenya

Direct contributions by tree crops to soil nutrient deposition and availability warrant more attention as nutrients from tree crops are added to nutrients already in the soil. For example, the return of maize stover to cropland and the transfer of nutrients from trees to the soil through pruning, leaf litter or decomposition of roots in agroforestry systems, contribute to the nutrient capitals of nitrogen (N), phosphorus (P) and other essential elements in the soil. These nutrients become available to successive crops during a time scale ranging from 1-10yrs.

Soil fertility benefits by tree fallows include;

- a) maintenance or increased levels of soil organic matter (SOM),
- b) uptake of nutrients below the food crop rooting zone,
- c) increased water storage and infiltration resulting in lower nutrient losses,
- d) reduced losses of soils and associated nutrients by erosion,
- e) improved soil aeration
- f) reduced soil acidity and
- g) improved soil biological activity.

Most of these soil processes have been researched under simultaneous agroforestry systems such as hedgerow intercropping (Juo et al., 1995; Kang et al., 1997) than for sequential systems such as planted tree fallows (Sanchez, 1995).

This report summarizes the carbon, nitrogen and phosphorus dynamics associated with soil aggregates sampled from three maize-tree-cropping systems. In the first study, carbon and nitrogen sequestration by *Crotalaria grahamiana*, (nitrogen fixing) and *Tithonia diversifolia* (non-nitrogen fixing) trees in rotation with maize in a low input cropping system was investigated.

Objectives of this first study included:

Determination of carbon and nitrogen deposits within aggregate size fractions
of 8-10 months planted tree fallow and maize crop rotations.

Hypothesis: Carbon accumulations in soil are a function of tree type and rotation.

2) An evaluation of C and N dynamics within aggregates, when fertilizer P is applied to maize and planted tree crops rather than maize crops only.

Hypothesis: Biomass production and carbon deposition within aggregates is enhanced when fertilizer P is applied to planted maize and tree crops.

Short duration fallows (<14 months), in which crop production is omitted for one season may have some potential in areas with high population where cropping is often continuous, such as western Kenya (Franzel., 1998; Buresh and Tian., 1998). The potential is greatest when returns to cropping are low, the opportunity cost of labor is high and the tree provides valuable products such as fuel wood (ICRAF, 1997; Swinkels et al., 1997; Buresh and Tian., 1998). It is financially more attractive to apply phosphorus inputs during the cropping phase of tree fallow systems as was shown with sesbania in western Kenya (ICRAF, 1997; Jama et al., 1998). Little is known, however, about the effects of short-term fallows on soil P and supply of plant available P to subsequent crops.

Accumulation of carbon, nitrogen, phosphorus and sulfur in SOM is dependent on P content of the soil parent material (Walker., 1965) but today few studies indicate that higher P results in more carbon and nitrogen accumulation. In Australia and New Zealand, fertilizer P increased the fertility of managed tillage systems (Paul and Clark). Soil organisms contribute to the renewal of the P pool both by their solubilizing effects on labile phosphorus (Nziguheba et al,

1998) and by their mineralization of organic phosphorus. Soil organisms and plant roots participate in solubilization of soil P through the production of CO<sub>2</sub> and organic acids (Smithson et al, 1999; Jones et al, 1999). The low level of P in soil solution and necessity for its frequent renewal suggests that the transfer of SOM P to inorganic P is primarily, but not exclusively, microbially mediated.

Mycorrhiza fungi play an important role in P transfers within soil root interactions. Three mechanisms have been proposed for this; I) mycorrhizal hyphae contribute to solubilization of mineral phosphate by their production of CO<sub>2</sub> and organic acids, ii) mycorrhizal hyphae extend soil exploration over and beyond the soil exploration by the plant roots. Hyphae penetrate SOM particles and macro aggregates more thoroughly than do roots, iii) hyphae may take-up P at lower levels of concentration in solution than roots.

In a second study, supply of plant available phosphorus was determined in concentric layers of soil aggregates, 4-8mm across.

The Objective was:

To evaluate the physical locations of recently applied plant available phosphorus within concentric layers of soil macro-aggregates and its relationship to maize yields.

Hypothesis: Soil aggregate P sequestration affects short-term plant available phosphorus.

The application of a stable carbon isotope as an intrinsic marker for tracer studies has recently provided a new impetus for root and soil C studies. During

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photosynthesis, C<sub>4</sub> plants discriminate less against <sup>13</sup>CO<sub>2</sub> than C<sub>3</sub> plants. The result of this fractionation of stable carbon isotopes during photosynthesis is a characteristic carbon isotope ratio of delta <sup>13</sup>C signature in the plant tissue. These isotopic signatures serve as effective markers for the occurrence of C<sub>3</sub> and C<sub>4</sub> photosynthesis (Dzurec et al, 1985). Crotalaria, a N-fixing fallow crop, has a C<sub>3</sub> metabolism, which generates delta <sup>13</sup>C signatures of approximately –26 ‰. Maize has a delta <sup>13</sup>C signature of –10 ‰. The trend would be to observe increased deposits of recent carbon at the surface layers of soil aggregates decreasing towards the interior. While using stable carbon isotopes to study two tillage systems, Smucker et al (1997) observed that twice as much carbon, derived from the contemporary crop, was deposited in the outer layers than the interior regions of soil aggregates sampled from 0-0.05m soil depth.

A third study was designed to investigate crop C deposits within aggregates derived from rotations of 8-10 months which had been planted to tree fallow (C<sub>3</sub> metabolism) and continuous maize cropping (C<sub>4</sub> metabolism) systems. The Objective was:

To determine *Crotalaria grahamiana* (C<sub>3</sub>) and maize (*Zea mays* L.) (C<sub>4</sub>) carbon contributions to external surfaces and interior regions of soil aggregates.

Hypothesis: Stable Carbon isotope signatures following a switch from continuous maize cropping to planted tree fallows in an agroforestry management system would demonstrate that recent organic inputs have a higher proportion of. carbon derived from the contemporary crop.

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#### Chapter 1

MANAGEMENT MODIFICATIONS OF CARBON AND NITROGEN DEPOSITS, IN SOIL AGGREGATES OF TREE AND MAIZE BASED SYSTEMS IN WESTERN KENYA

#### INTRODUCTION

Carbon (C) sequestration in soil appears to occur primarily in associations with the structural configurations of soil aggregates. Nearly 90% of soil organic matter has been reported to be located within soil aggregates (Angers and Chenu, 1997, Jastrow et al, 1996). Exudates and rhizosphere deposits of growing roots and hyphae are rapidly turned over by microbial populations, resulting in the deposition of polysaccharides in rhizosphere soils. As roots and hyphae die, organic substrates are deposited throughout the micro aggregate structures of the soil where the microbial activity reduces these substrates to particulate organic matter, resulting in further deposition of polysaccharides (Jastrow and Miller, 1997). The concept of an aggregate hierarchy in soils, for which organic matter is the major binding agent, is widely believed to occur through the organization of primary particles, cemented together to form micro aggregates up to 0.25mm in diameter. These micro aggregates are then bonded together into macro aggregates greater than 0.25mm (Tisdall and Oades, 1982; Oades, 1984).

In a recent review, Angers and Chenu, (1997) propose that soil organic matter (SOM) content in aggregate fractions results from, 1) the incorporation of SOM into different fractions, 2) specific turnover rates of carbon (C) which

depend upon chemical and physical configurations within aggregates, and 3) the turnover rate of the fractions. These mechanisms by which SOM becomes incorporated into soil aggregates are poorly understood. Perhaps more research is necessary to quantify the physical dissolution times of natural soil aggregates, their stability and associated processes by which SOM is incorporated into aggregates.

Loss of soil organic carbon resulting from long term continuous cultivation comes chiefly from inter-aggregate organic material that binds micro aggregates into macro aggregates (Elliot, 1986). This fraction of organic matter is characterized by Paul, (1984), and Voroney et al (1981) as less highly processed, aliphatic, stabilized slow or decomposable fractions.

In agricultural ecosystems, the portion of total organic carbon in the micro aggregate fraction is highest under non-cultivated systems and will decline with time under arable crops (Christensen, 1992). Agroforestry practices have an important potential to increase the soil content of sequestered carbon in tropical regions (Greenland, 1995). However, the magnitude of increase that can be attained depends on several factors, including nitrogen (N) and phosphorus (P) availability, and the sulfur (S) status of the soil. Management systems integrating shifting tree fallow and food crop rotations with high C inputs and reduced tillage should therefore favor carbon storage directly by reducing aggregate breakdown and indirectly by enhancing the SOM mediated stable aggregation. Carbon and N deposition within aggregates would be a function of total biomass added by the cropping system. Fallow tree crops produce large quantities of leaf litter and root

litter that is incorporated back into the soil as inputs in the cropping system. Non-N fixing trees of genus *Senna* and *Tithonia* accumulate as much N in their leaves as N-fixing legumes (Szott et al, 1991; Gachengo, 1996; ICRAF, 1998) and may compete as species alternatives on P limiting sites where N fertilizers can be added.

Little is known however, about the effects of short-term fallows on soil aggregation and C and N sequestration. We hypothesized that sequestration of C and N occurs in soil aggregates when tree-crop rotation with leguminous N fixing trees rather than maize monoculture is practiced. Additionally, non-N fixing tree fallow crops, growing on phosphorus depleted soil may posses equal C and N sequestration abilities within the same tree to maize crop rotation season, thus expanding farmer options for using tree fallow species to regenerate soil fertility. This study was conducted to a) determine carbon and nitrogen deposits within aggregate size fractions of 8-10 months planted tree fallow and maize crop rotations, b) assess nitrogen fixing Crotalaria grahmiana and non-nitrogen fixing Tithonia diversifolia (Hemsley A) Gray, as suitable tree species for rotation. These species have been proposed for adoption by farmers in a continuous maize cropping system with limited C and N inputs (ICRAF, 1998), and c) to compare C and N dynamics within aggregates, when fertilizer P is applied to maize and planted tree crops rather than sole maize crops.

# **MATERIALS AND METHODS**

# Site description

The study site is located at Osita farm in Vihiga District, Western Kenya (0°06' N, 34°34' E) at an elevation of 1420m. Mean annual rainfall is 1800mm (split between 2 cropping seasons, February-August and September to January). Often, the first cropping season will receive 1000-1200mm of the annual rainfall. The soil type is a very fine Isohyperthermic Kandiudalfic Eutrudox (USDA, 1996). It has few chemical or physical barriers to rooting in the top 4m. Air dried soil for the top 15cm had a pH of 5.1 (1:2.5 soil:water suspension), organic carbon 17g C kg<sup>-1</sup>, ex. acidity 0.4 cmolc kg<sup>-1</sup>, calcium 3.4 cmolc kg<sup>-1</sup>, (1.0 M KCL extractable), ex magnesium cmolc kg<sup>-1</sup> 1.4, ex potassium cmolc kg<sup>-1</sup> 0.12, phosphorus 2 mg kg<sup>-1</sup>, (bicarbonate-EDTA extractable). Clay, silt and sand textural contents were 46%, 28% and 26%, respectively.

# **Experimental design**

The experiment consisted of a factorial combination of three cropping systems (maize-maize, *Crotalaria*-maize, and *Tithonia*-maize) and two rates of P application (no P and 50kg P ha<sup>-1</sup>). Phosphorus was applied in March 1997as broadcast and incorporated into the top 0-15cm depth at 50kg P ha<sup>-1</sup> (as Triple Super Phosphate, 46% P<sub>2</sub>O<sub>5</sub>) The tree crop was relay planted into maize during March 1997 and April 1998. Maize (HB-512, Kenya Seed Co, Kitale, Kenya), was planted at 2 seeds per hill, at a spacing of 0.75m between rows and 0.25m between plants, and later thinned to 1 plant per hill to achieve a plant population

of 53 000 plants ha<sup>-1</sup>. One row of tree crop was planted for every row of maize resulting in the same tree plant population. Maize crop was harvested for grain yield in August 1998 and July 1999 in the tree-crop rotation plots. Two additional maize crops were harvested in January 1998 and 1999 (continuous maize plots). All maize crops were assessed for grain yields. The tree crop was harvested in February 1998 and 1999. All residue and litter fall was weighed and returned to the plots, while the woody components were exported from the plots for fuel wood. All above ground maize crop biomass was removed since it was common practice for farmers to harvest the stovers and utilize them as livestock feed. We only considered the residue returned as it had a direct effect on the fertility status of the soil. Leaf litter and pods were harvested and oven dried at 60 ° C for dry matter and plant N, P and K determination (Allen and Parkinson, 1975).

# Soil sampling

Soils were sampled for C and N determination from an ongoing experiment initiated in March 1997 to investigate phosphorus (P) replenishment with fallow tree crops in western Kenya. Soil samples were collected from different experimental plots on 13 Feb. 1998, which was designated to be the end of the tree fallow and the beginning of the food-cropping season. A second soil sample was collected on 7 Jul 1998, at the physiological maturity stage for the maize crop. Soils were collected (0-15cm depth) with cores (10cm diameter, 15cm height) gently driven into the ground to minimize aggregate disruption. Each soil sample was a composite of 9 locations per plot. The soil was loaded

onto trays secured with rubber strapping and transported to the station, where they were air-dried and stored in a cool, dry and dark room.

# Aggregate dry sieving

During both sampling dates, equal amounts (1 kg) of initial air-dried soil (moisture content 20a H<sub>2</sub>O kg<sup>-1</sup> soil) were prepared for dry sieving by removing rock, root and other debris. Samples were manually shaken through a nest of sieves (20-30cm diameter), separating them into eight size fractions (>8mm, 4-8mm, 2-4mm, 1-2mm, 0.5-1mm, 0.25-0.5mm, 0.053-0.25mm, 0.038-0.053mm and <0.038mm). The fraction retained on each sieve was weighed to estimate the mass of each size fraction of soil aggregates. These values were summed to determine each aggregate size fraction as a percentage of the total soil mass. The <0.038mm fraction was considered small (<0.5% of the fractions by mass) and in some cases was missing from the sieving process. Any sample collected was retained, but no further analysis was conducted for this fraction. Subsamples were obtained from these fractions for fine grinding and weighing at 5 decimal places for total carbon and nitrogen determinations. Angers and Giroux. (1996) evaluated C and N contents among wet and dry sieved aggregate fractions and found trends in C and N distribution among wet but not dry sieved aggregates. Therefore, we explored the differences in C and N across time for dry sieved aggregate fractions between February 1998 (undisturbed soil sample at the end of tree crop season) and July 1998 (end of 1st residual maize crop). Seasonal differences were a function of physical and chemical disruptions of

aggregates due to tillage, rainfall, maize crop activity and applied fertilizer materials. This provided an *in situ* opportunity to observe changes in dry aggregate mean weight diameter (MWD) as proposed by Kemper and Rosenau, (1986). MWD was calculated using the equation;

$$MWD = \sum_{i=1}^{n} XiWi$$

Where:

Xi = mean diameter of size fraction/size class midpoint;

Wi = proportion of total sample retained on the sieve.

## Carbon and nitrogen analyses

Carbon and nitrogen were analyzed by the dry combustion technique using a CHN analyzer (Carlo Erba, Milan, Italy). After fine grinding, a soil sample (20-40mg) is weighed and loaded into pressed tin cups (8mm x 5mm). It is then introduced to the CHN analyzer via an auto sampler. The sample was combusted at 1000° C in a stream of O<sub>2</sub> gas, oxidizing all forms of C to CO<sub>2</sub>. Nitrogen oxides are reduced to N<sub>2</sub> gas. These analyses provided carbon and nitrogen concentrations in the soil sample. We converted these percentages into actual amounts of C and N by mass for each aggregate size fraction and sub-sample in order to estimate the quantities of C and N sequestered by whole aggregates for each size fraction. Amounts of C or N in each size fraction were calculated by the equations (1) and (2).

C mass (mg) = [(%C \* aggregate size fraction mass (mg))]. (1)

N mass (mg) = [%N \* aggregate size fraction mass (mg)]. (2)

## Statistical analysis

Statistical analyses were performed with Statistical Analysis System (SAS Institute, 1999). Analysis of variance using the Proc GLM option was run for 3 crop factors at 2 phosphorus levels within 8 aggregate size fractions for % carbon, % nitrogen, mass of carbon, mass of nitrogen and C: N ratio. Seasonal changes associated with these components were also examined. In addition, hypotheses were decomposed into single degree of freedom contrast questions in order to investigate the pair wise cropping system effect on biomass and nutrient yields. The slice option was used to compare significant interaction data for the effects of crop at P levels and P levels for cropping factors. The objective is to gain insight into the interaction structure without physically splitting the data and yet retaining the power of the overall analysis. In the absence of a P interaction, data were pooled in order to increase the power of the *F*-tests.

## **RESULTS AND DISCUSSION**

Carbon and nitrogen distribution in soil aggregates sampled from 0-15cm soil depths followed a seasonal pattern. Initial P fertilization in March 1997 did not significantly affect C and N weight or concentrations of aggregate size

fractions in February and July 1998. There were no significant effects of cropping system on % C, % N, mass of C and mass of N in February 1998 (Table 1). During this time, lack of significant differences may be influenced by the dry season (December to February). In July 1998, significant differences in % C, % N, mass of C and mass of N were observed both across aggregate size fractions and among crop rotations. Changes in aggregate mean weight diameter by dry sieving showed a breakdown of large macro aggregates (>4mm) into smaller macro aggregates (<4mm) (Fig 1.). During the same time period, there was little change associated with aggregates <2mm. Soil aggregates are sensitive to management practices (Six et al., 1998). Upon cultivation, macro aggregates are expected to break down (Tisdall and Oades., 1982) into micro aggregates < 0.053mm (Six-et al, 1998). Large macro aggregates are less stable. They are probably bound together by less highly processed aliphatic and more stabilized or slowly decomposable fractions of OM (van Veen and Paul., 1981). However, SOM may not be the principal binding agent in Oxisols as iron and aluminium oxides contribute to the aggregation process (Oades and Waters., 1991). Kay et al (1988) suggested a model to propose a rapid decline of aggregate stability during conversion of a native old meadow to cultivation. Angers, (1992) reported little change in a single plow pass but recorded further declines in aggregation with repeated cropping Baldock et al. (1987) found that changes in aggregation following modifications in the cropping system were not always associated with changes in total organic carbon. In this study, changes in aggregate stability may have resulted from seasonal influences determined by tillage, crop root activity and wetting and drying cycles in the field.

# Aggregate fraction sequestration of carbon

Combined values of aggregates 4mm and larger accounted for 72% of the total C by mass in February 1998 but declined to 31% in July 1998 (Fig 2). The proportion of C and N sequestered in aggregates 0.5-4mm was highest in July. Perhaps physical disruption of aggregates across the season had resulted in reallocation of C among size fractions. Further, an increase in the mass of C was observed in aggregates <4mm (Table 2). In contrast, carbon concentration increased in the 4-8mm fraction but decreased in the 0.038-0.053mm fractions. No change was observed for all other fractions. In the same period, there was no significant change in the total C of the bulk soil. The highest decline in the mass of C was observed in large macro aggregates >8mm (Table 2). Redistribution among smaller fractions resulted in gains ranging between 700-3000 mg C kg<sup>-1</sup>.

Numerous variations in the carbon and soil organic matter associations among aggregate sizes have been reported. (Angers and Giroux., 1996). Elliot (1986), and Cambardella and Elliot (1993) reported decreasing contents of C and N with decreasing aggregate size. Earlier, Baldock et al., (1987) had observed elevations in both C and carbohydrate content with decreasing aggregate size while Gupta and Germida (1988) and Beare et al (1994) found no conclusive trends in C concentrations among size fractions. It is probable that some of the uncertainty associated with locations in SOM, C and N is related to the seasonal

migration of C and N (Fig 2). Among other factors, these seasonal trends would likely depend on climate, soil type and management.

# Aggregate fraction sequestration of nitrogen

Significant trends in nitrogen location (Table 3) and distributions (Fig 3) between February to July 1998, were similar to those observed with carbon. However, macro aggregates larger than 4mm lost 644mg N while aggregates 0.053mm-4mm gained 639mg N. Therefore, any changes that occurred in the total nitrogen pool were insufficient to cause significant losses of N within aggregate size fractions.

# Influence of tree-crop rotation on carbon and nitrogen distributions

Significant differences of % C and % N among crop rotations before maize planting in February 1998 appear too close to be of practical importance. These differences did not reflect alterations in the mass of C and N. Crop rotations significantly affected C and N deposits (Table 4 and 5) and the subsequent changes in C and N, after maize planting in March 1998. During the growing season, *Crotalaria* and *Tithonia* tree fallow rotation with maize increased the mass of N in contrast to maize-maize which, caused a depletion (p < 0.10, data not reported). Seasonal changes showed that, only tree-crop rotation increased the mass of C and N in July (p=0.08, data not shown)

# Carbon to nitrogen ratios

Significant differences among aggregate size fractions and crop rotations (Table 6 and 7) suggest that an aggregate by crop rotation interaction was the most important process influencing decomposition of recent organic inputs. We hypothesize that recent inputs associated with and responsible for binding large macro aggregates are transferred to smaller fraction sizes during decomposition. This process narrows the C: N ratio and precipitates an enrichment in N rather than C of aggregate 0.5-2mm across. Six et al., (1998) observed but failed to explain a N enrichment of the coarse Inter aggregate particulate organic matter (iPOM) in slaked aggregates.

#### Yields

Maize crop productivity for the first post-fallow season determined by grain yield was related to nitrogen availability (Fig 4). The larger quantities of crop residues that were returned to the fallow plots (Table 8) might have supplied sufficient plant nutrients to support yield levels 2-3 times higher than the maize control (Fig 5). Phosphorus addition to tree crops significantly increased P in tissue and the yield of P in biomass but not N or K (data not shown).

#### CONCLUSIONS

There seems to exist a time by management interaction, which controls the allocation of carbon and nitrogen within soil aggregate size fractions. Early

evidence has been presented to propose concentric sequestration of surface applied material (e.g. crop residue C and fertilizer N), with the most recent input being located on external regions of aggregates (Smucker et al., 1999; Santos et al 1997). Development of gradients of C would be expected to diminish with decreasing aggregate size as more highly processed, aliphatic or stabilized SOM (Paul, 1984) becomes incorporated into the humic fractions of soil. Past models do not seem to be adequate in explaining some of the differences occurring when SOM is incorporated into soil aggregates. This study suggests a migratory pattern of carbon and nitrogen that appears to be controlled by soil aggregate stability. Perhaps this pattern is a controlling mechanism in the breakdown and reformation of soil aggregates. Fallow cropping with tree-crop rotations rather than only maize does not seem to deplete C and N stored in soil aggregates, but maintains high productivity. Further research is needed to separate these control mechanisms in order to understand management modifications of C and N sequestration in low input agriculture.

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Table 1.1. Analysis of variance for the effect of cropping system and aggregate size on carbon and nitrogen dynamics of a Kenyan Oxisol (0-15cm depth), 1998. n=96.

			February					July		
					C:N					C:N
Source DF	ပ	Z	ပ	z	ratio	ပ	z	ပ	z	ratio
	I	(%)	<b>E</b>	(gm)		(%)	(9	<b>E)</b>	(Bш)	
Crop 2	8	<b>8</b>	2	8	‡	ŧ	#	*	‡	ŧ
Aggsize 7	ŧ	**	ŧ	#	ŧ	#	ŧ	ŧ	ŧ	ŧ
Crop*aggsize 14	#	***	ŧ	ŧ	ŧ	Ş	2	2	2	2
CV (%)	6.9	9.3	29.3	28.7	5.3	9.1	8.6	23.3	22.6	3.9

\*\*\*, \*\* and \* denotes significance at <0.0001, 0.01 and 0.05 level according to Fishers protected LSD<sub>0.08</sub> ns - not significant

Table 1.2. Soil carbon distribution within aggregate size fractions of a Kenyan Oxisol after 10 months of tree-crop rotation, 0-15cm depth, 1998.

•			Son carbon		
Aggregate size	ı		•	•	
Traction	<b>TeD</b>	rebruary		July	△ (change)
(mm)	(%)	(mg)	(%)	(mg)	(mg)
<b>&amp;</b>	1.98d	9468a	1.99de	3200b	-6268f
4-8	2.47b	4520b	2.64a	3064bc	-1456e
24	1.98d	2060c	2.09dc	2853c	794c
1-2	2.11c	2470c	2.17c	4389a	1919b
0.5-1	2.15c	1122d	2.15c	4290a	3169a
0.25-0.5	1.84e	547e	1.92e	2083d	1536b
0.053-0.25	1.96d	614e	1.92e	9520	339cd
0.038-0.053	2.64a	101f	2.46b	74f	-26d

Means in a column followed by the same letter are not significantly different according to Fishers protected LSD<sub>0.06</sub>

Table 1.3. Soil nitrogen deposits within soil aggregates of a Kenyan Oxisol following 10 months of tree fallow-crop rotation, 0-15cm depth, 1998.

			Soil nitrogen	J.	
Aggregate size					
fraction	Feb	February	J	July	
(mm)	(%)	(Bm)	(%)	(mg)	∆ mass (mg)
<b>&amp;</b>	0.17c	795a	0.17c	273b	-521g
8-4	0.21a	390b	0.23a	267bc	-123f
2-4	0.15d	155c	0.18c	240c	84d
1-2	0.12f	142c	0.18c	358a	215b
0.5-1	0.14e	74d	0.18c	347a	272a
0.25-0.5	0.13e	39ef	0.16d	173d	133c
0.053-0.25	0.14de	43ed	0.16d	78e	350
0.038-0.053	0.19b	<b>8</b> £	0.19b	<b>2</b> {	-26

Means in a column followed by the same letter are not significantly different according to Fishers protected LSD<sub>0.06</sub>

Table 1.4. Cropping system influence on sequestration of soil carbon of a Kenyan Oxisol, 0-15cm depth, 1998.

		Soll carbon	Irbon	
Crop rotation	Feb	February	Ĺ	July
·	(%)	(bw)	(%)	(mg)
Maize-Maize	2.11	2659	2.05	2451
<i>Crotalaria</i> -Maize	2.17	2575	2.28	2722
<i>Tithonia</i> -Maize	2.15	2602	2.18	2667
LSD 0.08	0.05	267	0.07	213

Table 1.5. Cropping system influence on sequestration of soil nitrogen of a Kenyan Oxisol, 0-15cm depth, 1998.

		Soil nitrogen	rogen	
Crop rotation	Feb	February	7	July
	(%)	(gm)	(%)	(mg)
Maize-Maize	0.155	206	0.17	202
Crotalaria -Maize	0.159	207	0.19	231
<i>Tithonia</i> -Maize	0.154	205	0.18	220
LSD <sub>0.06</sub>	0.005	20	0.005	11

Table 1.6. Changes in the C:N ratio of aggregate size fractions, 0-15cm depth, 1998.

•		N.	
Aggregate size fraction	February	ylnt	∆ (change)
(mm)			
<b>8</b>	11.90	11.7de	-0.22a
4-8	11.6e	11.50	-0.12a
2-4	13.4d	11.8cd	-1.60c
1-2	17.6a	12.2b	-5.40e
0.5-1	15.7b	12.3b	-3.4d
0.25-0.5	13.8cd	12.1bc	-1.70c
0.053-0.25	14.0c	12.3b	-1.77c
0.038-0.053	13.9c	13.0a	-0.90b

Change in the C and N deposit is expressed as the difference across the maize cropping season (Jul-Feb).

Means in a column followed by the same letter are not significantly different according to Fishers protected LSD<sub>0.06</sub>

Table 1.7. Maize cropping season and the change in the ratio of carbon to nitrogen, 0-15cm depth, 1998.

	N.	N.	V:S ∨
Crop rotation	February	July	
Maize-Maize	13.8b	12.2a	-1.61a
Crotalaria-Maize	13.8b	11.9b	-1.93b
Tithonia-Malze	14.3a	12.1a	-2.13b

Means in a column followed by the same letter are not significantly different according to Fishers protected LSD<sub>0.06</sub>

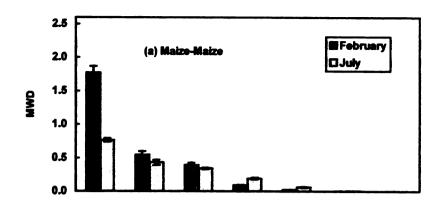
Table 1.8. Single degree of freedom contrasts for differences in means of yields, 1998.

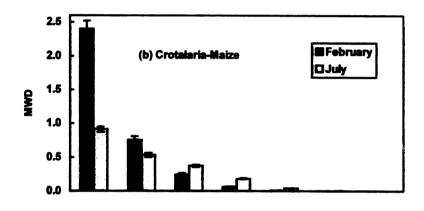
	Crop	Crop rotation	
	Crotalaria -		
Crop rotation	Maize	Tithonia -Maize	Significance level
	) <del>\</del>	kg ha <sup>-1</sup>	
Total tree biomass yield	9836	9364	ns
Crop residue returned	5702	1060	44 44
Plant N in residue returned	143	36	44 44
Plant P in residue returned	10	4	* *
Plant K in residue returned	29	36	æ
Maize yield (August 1998)**	3775	2561	*

\*\*\*, \*\* and \* denotes significance at <0.0001 0.01 and 0.05 level, according to Fishers protected LSD0.06

ns - not significant

\*\*Comparison of maize yields in Crotalaria and Tithonia plots excludes continuous maize.





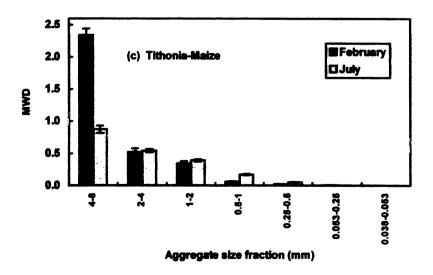


Fig. 1.1. Mean weight diameter changes of dry sieved soil aggregates as affected by maize-tree crop rotation on a Kenyan Oxisol (0-15cm), 1998.

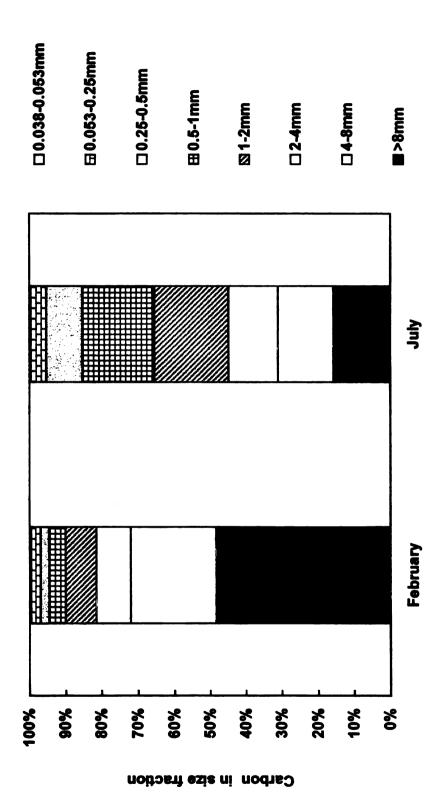


Fig. 1.2. Carbon mass in soil aggregate size fractions of a Kenyan Oxisol, 0-15cm depth, 1998.

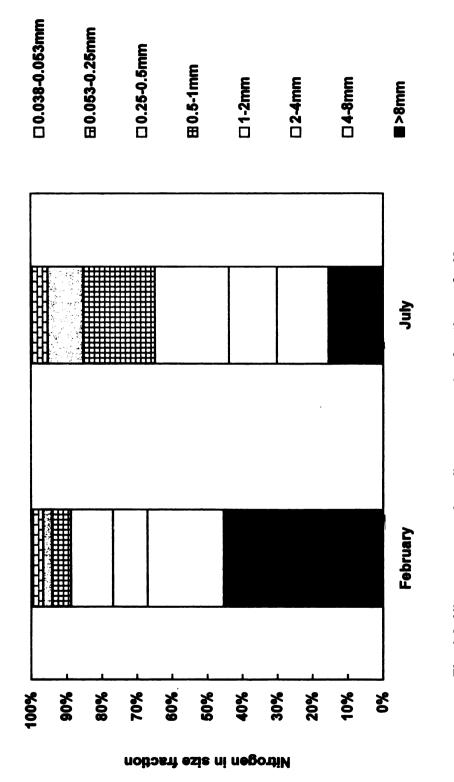


Fig. 1.3. Nitrogen mass in soil aggregate size fractions of a Kenyan Oxisol, 0-15cm depth, 1998.

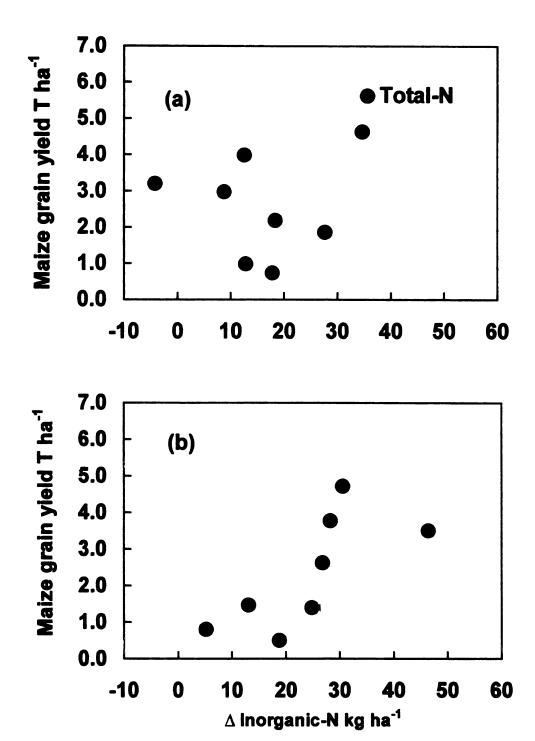


Fig. 1.4. Pre-season changes in the inorganic-N pool, with (a) or without (b) phosphorus fertilization and their relationship to August 1998 maize grain yield. Data are for continuous maize and plots converted to rotation with Crotalaria grahamiana, a leguminous tree. Changes in N are measured at the onset of the maize cropping season in February-March 1998.

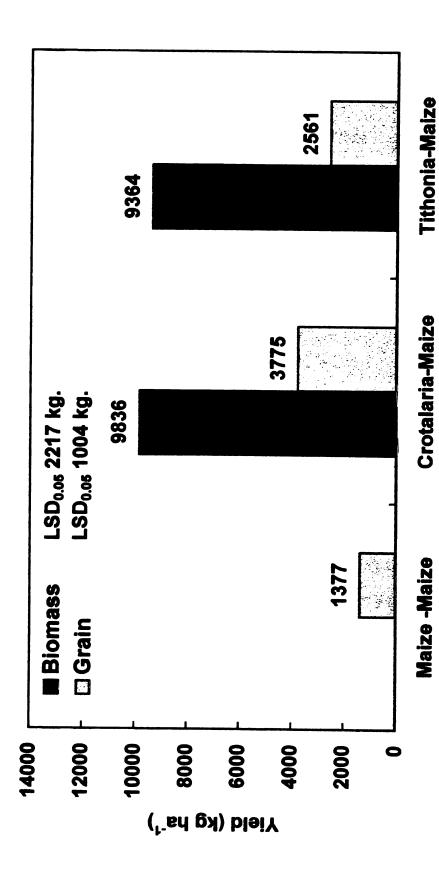


Fig. 1.5. Maize grain and tree biomass yield of three cropping systems at Osita Farm, Western Kenya, 1998.

## Chapter 2

# CONCENTRIC SEQUESTRATION OF PHOSPHORUS IN SOIL AGGREGATES OF MAIZE AND TREE BASED SYSTEMS IN WESTERN KENYA

#### INTRODUCTION

Soil phosphorus stocks in Sub Saharan Africa have decreased as increasing population has led to replacement of traditional systems of shifting cultivation with shorter duration fallow systems and sedentary agriculture (Buresh, et al. 1997). As traditional fallow systems are replaced by continuous cropping, soil organic matter (SOM) declines leading to reduced supplies of plant available P. Reduced SOM and lower P mineralization of organic phosphorus (P<sub>o</sub>) leads to greater reliance upon external P inputs. Elimination of P deficiency can enhance N<sub>2</sub> fixation by legumes (Giller et al, 1997) and integrated use of P fertilizers with organic materials to supply K and N can potentially enhance P availability (Palm et al 1997). In many ecosystems, including small farm holders in Africa, carbon (C) is the most limiting factor controlling microbial biomass and nutrient turnover. Huffman et al. (1996) suggested a close linkage between cycling of P through plants and soil biota is closely linked with C cycling. Others, (Gressel et al. 1996) have considered P as a component of organic material 'quality' and a factor in determining decomposition rates. Gressel et al, (1996) expressed concern that extraction based indices and phosphatase activity failed to explain P availability. They further proposed that chemical linkages with Cstructure might be an important factor controlling P release; in agreement with

Tiessen et al, (1992) who found that NaOHP<sub>o</sub> declined at about the same rate as C in a Brazilian soil. It is most probable that the availability of P is largely determined by organic equilibria but that current methods for studying these equilibria are poorly developed.

During the formation of soil aggregates, many components of the soil are redistributed within the structural unit. Equilibration of developing aggregates with the various biogeochemical activities result in the development of gradients within individual soil aggregates (Horn, et al., 1994; Santos et al., 1997). Aggregate erosion of concentric layers using stainless steel meso soil aggregate erosion (SAE) chambers (Smucker et al, 1999), is a new method being developed for separating multiple surface layers from soil aggregates, by gentle erosion, provide samples for further analysis. Quantification of these separate components could enhance our understanding of the fate of surface applied materials such as crop residues, fertilizer granules and pesticides that are controlled by the formation, breakdown and reformation processes of soil aggregation. Standard P-sorption isotherm methods, which grind and vigorously shake soil (Fox and Kamprath, 1970), appear to expose diffusion limited Psorption sites, overestimating the buffering capacity of field soil. Based on sorption isotherms of an aggregated soil, Cassman et al. (1981) estimated and recommended >500kg P ha<sup>-1</sup> for optimum yields but Cassman. (1993) reported that 100% of maximum yield could be achieved by applying 100kg P ha<sup>-1</sup>. Other procedures that extract labile P from throughout the aggregate may give potentially erroneous values of available P since some of this P may be

unavailable to plants due to its physical sequestration within aggregates (Linquist et al, 1997).

Despite extensive research on soil P reactions, little is known about the effect of soil aggregation processes on these reactions. Nutrients commonly occurring at low concentrations in soil solution, such as P are transferred to root surfaces at insufficient rates to meet uptake requirements of plants. Root uptake of P results in a decrease in their concentration in the soil solution surrounding plant roots. This depletion generates a concentration gradient at the soil-root interface. Linguist et al. (1997) demonstrated that P sorption is related to aggregate size, soil buffering capacity and soil P release rates. They observed P retention on surface regions of soil aggregates 0.188mm across, which they described as mean reactive mass. Therefore, P flux between the surface layer and interior regions of aggregates appears to be a time dependent process controlled primarily by diffusion. These concepts led to the hypothesis that P sequestration within interior regions of soil aggregates adversely affects shortterm plant available phosphorus. The objectives of this study were to determine, i) the physical location of recently applied phosphorus within soil aggregates and ii) the effect of planted tree-crop rotations on short term plant available phosphorus and its relationship to yields.

## **MATERIALS AND METHODS**

### Site description

The study site of this project is located at Osita Farm in Vihiga District, Western Kenya (0°06' N, 34°34' E) at an elevation of 1420m. Mean annual rainfall is 1800mm (split between 2 cropping seasons, February-August and September to January). Often, the first cropping season will receive 1000-1200mm of the annual rainfall. The soil type is a very fine Isohyperthermic Kandiudalfic Eutrudox (USDA, 1996). It has few chemical or physical barriers to rooting in the top 4m. Air dried soil for the top 15cm had a pH of 5.1 (1:2.5 soil:water suspension), organic carbon 17g C kg<sup>-1</sup>, ex. acidity 0.4 cmolc kg<sup>-1</sup>, calcium 3.4 cmolc kg<sup>-1</sup>, (1.0 M KCL extractable), Ex magnesium 1.4 cmolc kg<sup>-1</sup>. ex potassium cmolc kg<sup>-1</sup> 0.12, phosphorus 2 mg kg<sup>-1</sup>, (bicarbonate-EDTA extractable). Soil textural distributions were 46%, 28% and 26% for clay, silt and sand respectively.

The experiment consisted of a factorial combination of three cropping systems (maize-maize, *Crotalaria*-maize, and *Tithonia*-maize) and two rates of P application (no P and 50kg P ha<sup>-1</sup>). Phosphorus was applied in March 1997as broadcast and incorporated into the top 0-15cm depth at 50kg P ha<sup>-1</sup> (as Triple Super Phosphate, 46% P<sub>2</sub>O<sub>5</sub>). Maize (HB-512, Kenya Seed Co, Kitale, Kenya), was planted at 2 seeds per hill, at a spacing of 0.75m between rows and 0.25m between plants, and later thinned to 1 plant per hill to achieve a plant population of 53,000 plants ha<sup>-1</sup>. The tree crop was relay planted into maize during March

1997 and April 1998. One row of tree crop was planted for every row of maize with the same intra-row spacing, resulting in the same tree plant population.

Maize crop was harvested for grain yield in August 1998 and July 1999 in the tree-crop rotation plots. Two additional maize crops were harvested in January 1998 and 1999 (continuous maize plots). All maize crops were assessed for grain yields. All above ground maize crop biomass was removed since it was common practice for farmers to harvest the stovers for livestock feed. Therefore, only root C, N, and P were considered for maize trials. The tree crop was harvested in February 1998 and 1999. All residue and litter fall was weighed and returned to the plots (Kinyangi et al., unpublished data), while the woody components were exported from the plots for fuel wood. Only the residues returned were considered as directly affecting the fertility status of the soil. Leaf litter and pods were harvested and oven dried at 60 ° C for dry matter and plant N, P and K determination (Allen and Parkinson, 1975).

### Soil sampling

Soil from the field trial was sampled on the following dates; 13 February 1998 (end tree crop season but before tillage), 7 July 1998 (post tree crop season and maturity of residual maize), 19 September 1998, (initiation of 2<sup>nd</sup> tree crop season), 13 February 1999, (end of tree crop season 2 and return to maize cropping) and 1 July 1999 (post tree crop 2 and maturity of maize). Soil samples were collected (0-15cm depth) with sharpened cores (10cm diameter by 15cm height) which were gently driven into the soil surface to minimize disruption of

aggregates. The soil was loaded onto trays secured with rubber strapping and transported to the laboratory. Air-dried samples (1 kg) were manually shaken through a nest of sieves, separating them into eight size fractions (>8mm, 4-8mm, 2-4mm, 1-2mm, 0.5-1mm, 0.25-0.5mm, 0.053-0.25mm and 0.038-0.053mm). We followed P dynamics during the tree crop season in the 4-8mm aggregate size fraction. This size fraction contained 40% of the total carbon and nitrogen, by mass, for all aggregates <8mm, (Kinyangi et al, unpublished data). This fraction of aggregates contains the least depth of the reactive mass layer as defined by Linquist et al, (1997).

## Aggregate erosion

Soil aggregates (4-8mm across) collected from the experiment were further processed by eroding individual aggregates (n = 15) into external, transitional, transitional and internal regions of equal mass (0.15 g) using stainless steel meso soil aggregate erosion (SAE) chambers (Santos et al., 1997). Bases of the SAE chambers were fitted with screens with 356 $\mu$ m openings. Individual aggregates were placed into a SAE chamber (2.5cm diameter) with finely knurled walls, the top was sealed with a rubber stopper secured with para film to prevent dust contamination and secured into glass beaker assembly (100ml) using foam packing. The glass beaker assembly was rotated on a variable speed rotary shaker (Innova 2300, New Brunswick, Canada). As the aggregate rolled around the inside of the knurled SAE chamber the fine (<356 $\mu$ m) fraction fell through the sieve and was deposited in the lower

base (Fig 1). Additional protocols for uniformly removing equal quantities of 33% from each concentric layer are described by Smucker et al, (1999). In short, samples were weighed periodically to determine the time required too remove each concentric layer/region from each soil aggregate fraction processed from each treatment (Fig 2). The final eroded samples obtained were passed through a 0.053mm sieve to exclude the sand fraction as most of the resin extractable phosphorus (REP) varied with sand content ( (Fig 3).

## Modified resin extractable phosphorus.

Plant-available P is a functional concept rather than a measurable quantity (Tiessen and Moir, 1993). Further, it may be described as solution inorganic P (P<sub>i</sub>) which is continuously recharged by that P<sub>i</sub> which enters the solution during the period which defines availability during the growing season. Exchange resins have recently gained importance in identifying plant-available P<sub>i</sub>. The resin offsets the equilibrium between dissolved and soluble P<sub>i</sub> allowing more precipitated forms to enter the P depleted solution. The P sorbed by the resin is extracted by a weak acid and quantified spectrophotometrically. This pool has often been equated to the isotopically exchangeable P since the resin does not chemically modify the soil solution. Linquist et al, (1996) as well as Guo and Yost (1998) have shown that extraction by Fe-impregnated paper can also characterize this pool. However we chose to use the exchange resin approach because it provided the best opportunity to determine available P in the presence of a P sink similar to the root activity of growing plants.

A variety of methods have been developed and adopted using different anionic forms, shaking times, ratios of soil:water:resin for enclosed bags or beads in free suspension (Sibbesen, 1977; Sibbesen 1978; Tiessen, 1991, Tiessen, 1993). Enclosed bags are commonly used because they offer the advantage to regenerate the resin by washing through HCL baths. It seems however that the decision regarding ratios of a resin-P to soil mass should be determined by the objective of the research. A resin membrane (BDH No. 55164, 9\*62mm) extraction of 0.5g soil in 30ml water was described by Tiessen and Moir, (1993). Maroko et al, (1999) used mesh bags (Dowex 1-X8, 1g converted to bicarbonate form) to extract 2.5g soil in 40ml water. In a widely adopted sequential extraction, Hedley et al, (1982) used resin enclosed in nylon bags (Dowex 1-X8-50, 0.4g converted to bicarbonate form) to extract 0.5g soil in 30ml of de-ionized water (DIW)., Agbenin et al, (1998) have also used the Hedley (1982) fractionation to characterize available P.

We modified the resin in mesh bags approach in order to: a) establish a minimum soil sample weight required to extract a detectable P level which could be removed by the resin. This approach was based on the fact that soil eroded from concentric layers of aggregates were small with a mass <200mg; b) vary the resin:soil ratio to achieve maximum sensitivity of extraction while reducing insample variability and c) determine if the method could adequately characterize available P in concentric layers of soil aggregates. We selected soils with a range of resin extractable P and varied the ratio of resin: soil (1, 2, 5 and 10) at 3 soil

weights (0.5g, 0.25g and 0.125g). The soil was collected from the experiment on 7 July 1998 in the following plots;

Maize-maize, no P applied, (Soil 1)

Maize-maize, 50kg P ha<sup>-1</sup> as initial application in March 1997, (Soil 2).

Maize- maize, 50kg P ha<sup>-1</sup> with a repeated P application in March 1998, (Soil 3).

P was extracted by shaking soil samples in 60ml bottles for 16 hrs at 175 reciprocations min<sup>-1</sup> with a Dowex resin (1-X8,Cl<sup>-</sup>, Sigma Chemicals, St. Louis, MO) enclosed in mesh bags (5mm\*4mm, 83µm opening) in 40ml of de-ionized water (DIW). The resin was supersaturated with sodium bicarbonate (0.5M). Phosphate ions adsorbed on resin were extracted by shaking the P-resin, retained within the mesh bag, in 30ml bottles with 15ml of 0.1M HCl extractant for 1hr to establish equilibrium. The extract is filtered through Whatman paper (no. 1). Phosphorus was determined spectrophotometrically (Murphy and Riley, 1962), on an automatic Lachat (Quickchem Systems, Milwaukee, WI). Ammonium molybdate and potassium tartarate reacted with dilute solutions of phosphorus to form antimony phospho molybdate complex. This complex was reduced to an intensely blue colored complex by ascorbic acid. The color, determined at 900nm, was proportional to the phosphorus concentration in the sample. Minimum detectable limits of P were 0.01ppm.

Varying the resin:soil ratio extracted different amounts of P (Fig 4). An increase in resin load relative to the soil weight resulted in more REP. The extraction seemed to increase with the amount of available phosphorus. This

suggests that there is need for higher resin loading in P fertilized soils, as well as soils undergoing rapid organic matter transformation that releases phosphorus. In contrast, a lower resin load, as is common in most present extractions seems to under-estimate available phosphorus. Significant differences in available P with decreasing sample weight (Table 1), may be due to greater concentrations of P in solution, since the volume of DIW we used remained constant at 40ml. We concluded that the best P extraction occurred with 0.125g of soil sample and 0.625g of resin at a ratio of 5:1 (resin:soil). Although within sample variability was comparable across extraction ratios, 5:1 offered the best opportunity to maximize P extraction from soils with both low and high P concentrations.

## Exterior to interior ratio (EIR).

Ratios of the measured REP (ie., interior to exterior ratio – EIR) were used to estimate the relative allocation of one or more P pools within concentric layers of soil aggregates (Kinyangi et al, unpublished data). This provided an evaluation of the physical location of P from recently applied fertilizer or P from plant residue materials. These evaluations were used to compare the EIR relationship to crop productivity for the crop rotations reported in this study. Additionally, changes in the EIR were monitored over time to observe shifts within pools. Studying these dynamics was intended to yield knowledge of residual P benefits with the intent of predicting the need for fertilizer P replenishment, using estimates of a "critical ratio". The EIR was calculated by the following equations:

$$EIRt_1 = [layer (Xt_1)/layer (Yt_1)]$$
 (Eq. 1)

$$EIRt_2 = [layer (Xt_2)/layer (Yt_2)]$$
 (Eq. 2)

Change in EIR = 
$$[EIRt_2-EIRt_1]$$
 (Eq. 3)

## Where:

EIRt<sub>1</sub> is the ratio at time 1

EIRt<sub>2</sub> is the ratio at time 2

Xt<sub>1</sub> is the exterior value of P (mg kg<sup>-1</sup>) at time 1

Yt<sub>1</sub> is the interior value of P (mg kg<sup>-1</sup>) at time 1

Xt<sub>2</sub> is the exterior value of P (mg kg<sup>-1</sup>) at time 2

Yt<sub>2</sub> is the interior value of P (mg kg<sup>-1</sup>) at time 2

# Statistical analysis

Analysis of variance was performed using the Proc GLM option of the Statistical Analysis System (SAS Institute, 1999). All significant tests reported, refer to  $\alpha = 0.05$  unless otherwise stated. Individual hypotheses were decomposed into single degree of freedom contrast questions in order to investigate the individual cropping system or combined fallow effect on available phosphorus. In the absence of a significant P interaction, data were pooled in order to increase the power of the F tests.

#### **RESULTS AND DISCUSSION**

## Plant available phosphorus in whole aggregates

Seasonal patterns in resin extractable P of whole aggregates between March 1997 and February 1999 were influenced by the presence or absence of maize and tree crops (Fig 5). Initial application of 50kg P ha<sup>-1</sup> to planted maize and trees in March 1997 increased REP of whole aggregates in February 1998 (data not shown). A modest addition of 50kg P ha<sup>-1</sup> as triple super phosphate in the 5<sup>th</sup> season during March of 1999 increased resin extractable P (REP) within all three cropping systems (data not shown). However, these increases of available P in whole aggregates across 5 cropping seasons were not significant probably due to high variability associated with heterogeneity among the different soil aggregates. More replications of samples, up to 3 times, in the laboratory did not seem to effectively address this problem. Extractable P peaked at 12mg P kg<sup>-1</sup> <sup>1</sup> in March 1998 but declined over the maize cropping season (March 1998-July 1998). A build-up in this P pool was observed in September 1998 at the initiation of the 2<sup>nd</sup> fallow season. The absorption of P by growing maize and tree crops was observed in February to July 1998 and February to July 1999.

It is known that the interaction between biological (organic) and chemical (inorganic) transformation determines P availability. Considerable research (Hedley et al, 1982; Tiessen et al, 1984; Stewart & Tiessen, 1987; Beck & Sanchez, 1994) has revealed the existence of P fractions of differing extractability based on their functional status. Phosphorus can be partitioned into

readily available, reversibly available and sparingly available pools based on a) accessibility to plant roots, b) reversible conversion into available P and c) pools that undergo long term transformations which however may still contribute to P cycling in the short term. In this study, the appearance of a trend, which lacked significant differences among management systems, may be an indication that REP interacts with additional pools that we did not measure. Tiessen and Moir (1993) described this concept as an activity where less soluble or slower pools of P replenish available P at varying rates.

Previously, (Kinyangi et al, unpublished data), we reported changes of C and N in aggregate fractions. We then correlated the fraction with the largest shift in the C: N ratio (1-2mm) and larger macro-aggregates (>8mm) with changes in REP (Fig 6). There is a strong relationship between changes in C concentrations of aggregates 1-2mm across and P turnover during the post fallow season. This observation suggested that P availability was influenced by a negative change in C concentrations. It is speculated that this negative change was probably a result of C-mineralization. This would support the proposal by Tiessen et al (1992) who reported declines in the NaOHP<sub>0</sub> organic pool, at about the same rate as C contents declined in a Brazilian soil.

# Comparisons of EIR P to gradients in plant available P

EIRs of resin-extracted P from concentric layers of soil aggregates, reported in this study, appeared to be a sensitive and early indicator of plant responses to cropping and fertilizer management modifications. Wan and El

Swaify (1998) reported an enrichment ratio (ER) and changes in the ER to study mechanisms responsible for enrichment of organic carbon and extractable P in fine aggregates of an Oxisol. Elsewhere, carbon to nitrogen (C: N) and carbon to phosphorus (C: P) ratios have been used to demonstrate mineralization-immobilization phases of carbon, nitrogen and phosphorus cycling. Smucker et al (1997) used clay mineralization ratio (CMR) to show changes associated with accelerated mineral weathering in the external layers of soil aggregates from multiple locations and ecosystems.

During this two year study, the EIR of available P in aggregates of the Crotalaria-maize rotation declined steadily (Fig 7) suggesting that plant available P at the surface of these aggregates was constantly being utilized. This absorption further resulted in 2 to 3 fold increases in maize yields of the Crotalaria rotation in comparison to maize monoculture (Fig 8). In contrast, available P at the surface of aggregates in the continuous maize management system appeared to accumulate. Nitrate accumulations in bulk soils of P deficient sites in western Kenya has been reported where maize growth is severely limited by P deficiency (Hatermink et al, 1996; Jama et al, 1997; Mekonen et al, 1997). Phosphorus application on these sites would therefore be expected to increase maize production. The accumulation of P and subsequent poor yield of maize (Fig 8) may indicate the need for both N and P fertilization where non-N fixing fallow crops, such as Tithonia, are integrated into the cropping system.

It has been recognized that plant available P is a functional concept rather than a measureable quantity. Total plant available P includes all P taken up

during a specific cropping season or growth cycle period (Tiessen and Moir, 1993). Phosphorus fertilization appears to initially deposit P on aggregate surfaces where it becomes accessible to plant roots exploring the soil matrix. Linquist et al (1997) showed fertilizer P retention on aggregate surfaces and used a *mean reactive mass* to characterize this region of retention in aggregates. They linked declining productivity from residual crops to aggregate sequestration of P where a time dependent diffusion process limits P availability. We observed an equilibration in the EIR of available P in aggregates when P application was sufficient to overcome plant extraction (Fig 9b). The reverse also occurred when P application was insufficient to meet extraction needs (Fig 9a) resulting in higher P concentrations in the interior regions than the exterior layers of soil aggregates under *Crotalaria*-maize rotation.

#### CONCLUSIONS

Phosphorus fertilization of soils deposits much of the P on surfaces of soil aggregates, which slowly diffuses into the interiors of aggregates. The mean-free diffusion pathway and physical chemistry of each aggregate appears to control this diffusion of available P to surrounding plant root systems. Additional research should reveal how these spatial gradients at multiple locations within soil aggregates will temporally influence plant available P and how consistent these aggregate layered P gradients are with other P pools previously described by Hedley et al., (1982).

Whether similar P gradients exist in smaller aggregate sizes remains to be discovered. The high correlation between yield and EIR of soil aggregates (4-8 mm across), representing 23.7% of the total bulk soil, suggests that a cascading effect of P availability may be occurring from interior regions of smaller aggregates, followed by the interiors of larger aggregates. In the process, plant available P is removed from surface layers of large aggregates. Soil tests that crush bulk soil samples suggest these current measurements of labile P fractions may be poor indicators of P availability to plant roots since these tests destroy the original aggregate structure. The absence of detectable differences among different management systems and P extracted from crushed whole aggregates. in this study, further suggests the need for establishing a concentric layer-based index of soil aggregates before more accurate identification of P availability to plants can be achieved. Accumulations of plant available P at surfaces of larger aggregates in *Tithonia*-maize and continuous maize rotations would not have been apparent, using contemporary methods, as these gradients would have been diluted during the extraction of labile soil P from macerated whole soil samples. Separation of uniform concentric layers by meso SAE chambers provides an integrated non-destructive technique that more accurately projects plant available P-pools and gradients experienced by roots in situ. Therefore, a ratio relating extractable P at the surfaces of aggregates to interior P can be useful for determining requirements for P fertilization. Using this knowledge of P processes, it is hypothesized that, P fertilization will be required when a decrease in the EIR coincides with a decline in residual plant yield, where temporal

demands for other essential nutrients (N and K) have been satisfied. Further research is needed such as rate response experiments, to evaluate the accuracy of EIR values for predicting plant and fertilizer interactions for multiple soil types for maximizing the efficiency of P fertilization in low input agricultural systems.

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Quality 27:132-136.

Table 2.1. Increasing resin:soil ratio and changes in extraction of plant available phosphorus of 3 soils.

	Soil 3		Soil 2		Soil 1		
Resin:Soil ratio	0.25g	0.5g	0.25g	0.125g	0.25g	0.125g	
	(mg kg <sup>-1</sup> )						
1	5.9	5.2	9.6	13.1	24.8	25.6	
2	5.9	7.3	10.5	15.8	27.4	26.2	
5	nd	nd	12.1	17.8	31.5	33.8	
10	nd	nd	nd	nd	29.0	32.8	
Soil mean ext P (mg kg <sup>-1</sup> )	6.1		1	13.1		28.9	
ANOVA							
Soil weight	ns		***		**		
Ratio	*		***		***		
Ratio*Soil weight	*		**		**		

<sup>\*\*\*, \*\*</sup> and \* denotes significance at 0.001, 0.01 and 0.05 level tests according to Fishers protected LSD<sub>0.05</sub>

ns - not significant.

nd - not determined.

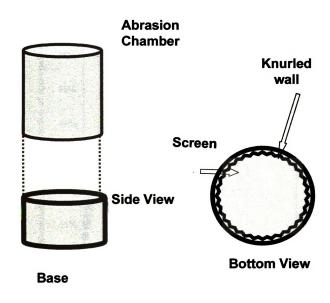


Fig 2.1 Diagramatic illustration of meso- Soil Aggregate Erosion (SAE) chamber

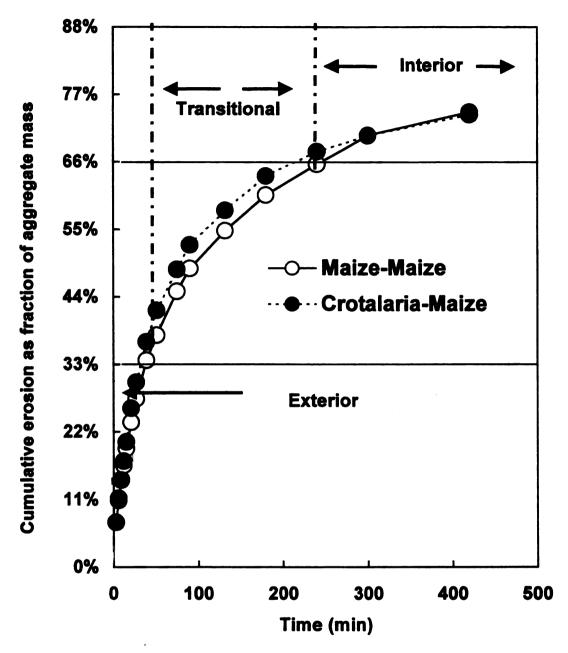


Fig. 2.2 Time versus progressive removal of concentric layers of aggregates, 4-8mm across (mean=6mm), from continuous maize and *Crotalaria* rotation plots of a Kenyan Oxisol, 1999. Each region/layer is one third of the original mass of the aggregate. n = 12.

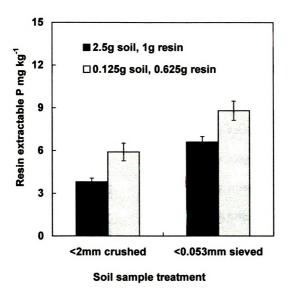


Fig. 2.3. Effect of sand removal on resin extractable phosphorus of a Kenyan Oxisol. The two extraction regimes are representative of the conventional resin P method (2.5g soil, 1g resin) and a more recent method (0.125g soil, 0.625g resin) developed to extract P from layers of soil aggregates. Bars are standard error. n=24.

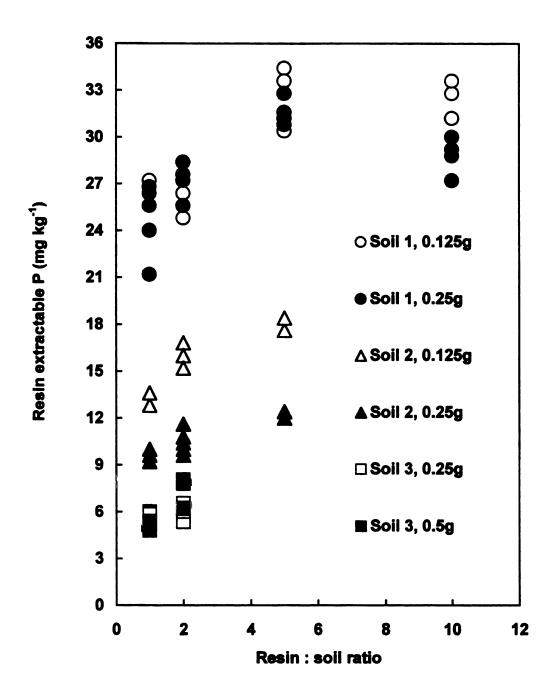


Fig. 2.4. Influence of resin load and soil weight on resin extractable phosphorus. The resin is chloride base converted to bicarbonate form. The soil is shaken for 16 hrs at 175 reciprocations min-1.

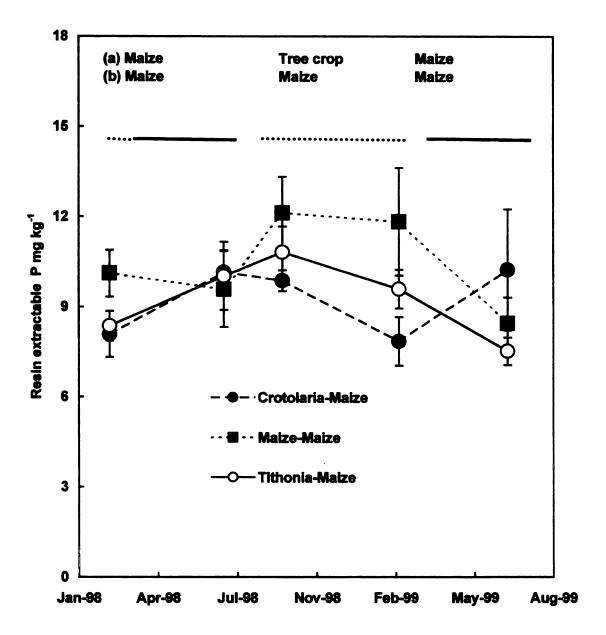


Fig. 2.5. Seasonal flux of resin extractable phosphorus in whole aggregates of 3 management systems without P fertilization on a Kenyan Oxisol. Crop sequence represents (a) Tree-crop rotation and (b) continuous maize plots. Error bars are standard errors for the difference in means.

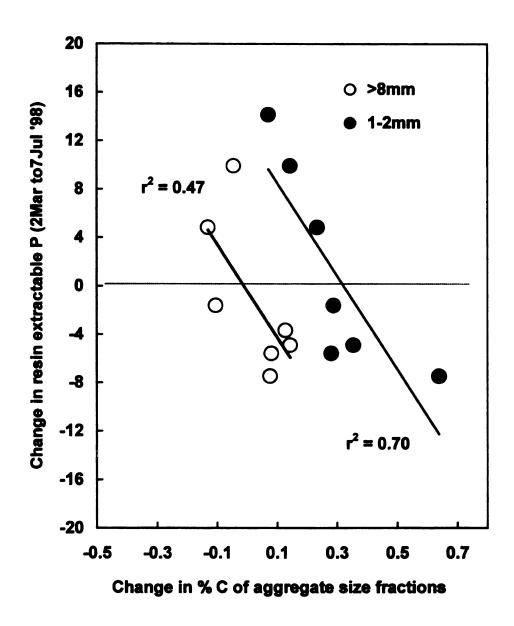


Fig. 2.6. Influence of post fallow C concentrations of aggregates on subsequent pre-season changes in resin extractable phosphorus. Changes of resin P in continuous maize plots (control) is subtracted from fallow rotation plots. n=4

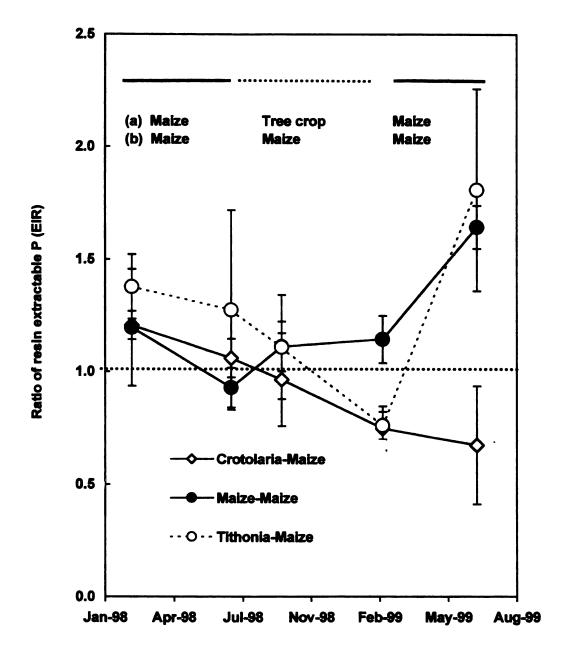


Fig. 2.7. Cropping system effect on seasonal location of plant available phosphorus in concentric layers of soil aggregates (4-8mm across), of a Kenyan Oxisol, 0-15cm depth. Crop sequence represents (a) Tree-crop rotation and (b) continuous maize plots. Error bars are standard errors for the difference in means. n= 4.

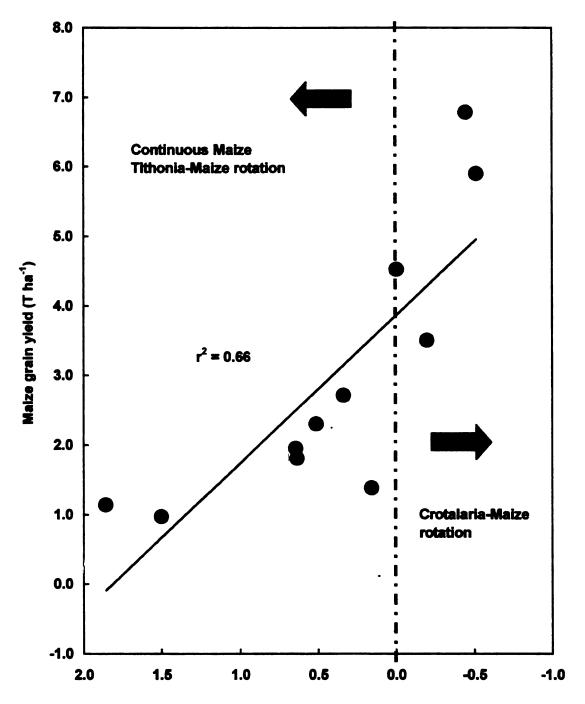


Fig. 2.8. Relationship between change in the ratio (Jul 99-Sep 98) of exterior to interior resin extractable phosphorus and July 1999 grain yield. n = 4.

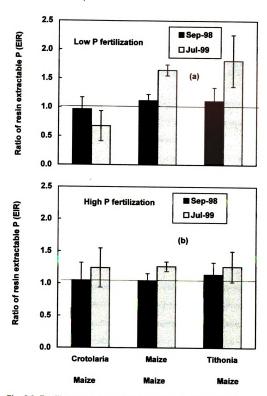


Fig. 2.9. Fertilizer management and gradients in P location in aggregates, 4-8mm across (0-15cm) of a Kenyan Oxisol, 1999. n =4.

#### Chapter 3

# ESTIMATING RECENT CROP CARBON INPUT INTO CONCENTRIC LAYERS OF SOIL AGGREGATES FROM MAIZE AND TREE BASED SYSTEMS

#### INTRODUCTION

Decline in soil organic carbon (SOC) appears to occur when farming practices do not return crop residues to the soil and increase soil disturbance. thereby enhancing carbon (C) mineralization and CO<sub>2</sub> losses to the atmosphere (Lal, 1989; Bajracharya et al, 1996). The rate and quality of carbon transfer below ground, together with microbial activity are recognized as important processes governing SOC dynamics (Jastrow and Miller, 1997). A frequent C loss during conversion of land to agriculture, within small holder farms in Africa. has recently been characterized as a three-step process (Woomer et al, 1997). First, nutrients and C stocks held in plant biomass of natural vegetation are mobilized during land conversion. Second, the soil resource is exploited for several years of productive cropping, during which nutrient rich root residues decompose and organic matter (OM) is mineralized. At this point, reduced plant yields will often be associated with a declining level of soil organic matter (SOM). Third, a lower level equilibrium of SOM is established, resulting from mineralization of the more labile SOM fractions. Continued productivity then becomes dependent on external inputs, largely consisting of organic materials and/or inorganic fertilizer.

Current research approaches for studying C mineralization have been limited to estimating long-term C balances based on whole soil tests and above or below ground productivity of crop biomass. Woomer et al, (1997) summarized data (2-30yrs in 0-40cm soil depth) on C loss due to cultivation in Eastern and Southern Africa. The authors reported system losses ranging between 0.9 T of C ha<sup>-1</sup> vr<sup>-1</sup> to 10 T of C ha<sup>-1</sup> vr<sup>-1</sup>. These losses are similar to those observed over 6 years (10 T of C ha<sup>-1</sup>) by Tiessen et al., (1992) during thorn bush conversion to agriculture in Brazil. Current short-term estimations of soil C dynamics have been limited to soil emissions of CO<sub>2</sub>. However, it has been difficult to partition and interpret some of the results when the presence of growing plants contributes to rhizosphere respiration (Rochette et al., 1992; Reicosky and Lindstrom, 1993; Rochette et al., 1999). Some of these difficulties may be resolved by integrating different methods for studying C such as root exclusion by fine mesh screens, isotopic labeling and more recently minirhizotron evaluation of root activities (Aiken and Smucker, 1996). Smucker et al. (1997) identified C gradients within soil aggregates and these C gradients changed rapidly after 20 months of plant root growth in the field and as short as a few weeks in the greenhouse. suggesting that total C analyses of bulk soil samples has concealed very important short term changes of soil C.

The application of a stable carbon isotope as an intrinsic marker for tracer studies has recently provided new and exciting opportunities for soil C studies.

During photosynthesis, C<sub>4</sub> plants discriminate less against <sup>13</sup>CO<sub>2</sub> than C<sub>3</sub> plants.

The result of this isotopic discrimination, is a characteristic carbon isotope ratio in

the plant tissue, which serves as an effective marker for the occurrence of C<sub>3</sub> and C<sub>4</sub> photosynthesis (Dzurec et al, 1985). The carbon isotopic composition of SOM is comparable to that of the source plant material prior to humification (Schwartz et al, 1986). The natural abundance of <sup>13</sup>C has been used to evaluate vegetation shifts, (Desjardins et al., 1996; Boutton et al., 1998), community composition, (Mariotti et al., 1994; Boutton et al, 1999), plant and soil respiration, (Hogberg et al., 1996; Nyberg et al., 1999; Rochette et al., 1999), SOC (Arrouays et al., 1995) and SOM turnover, (Balesdent et al., 1993; Bernoux et al., 1998) and net primary production (Jenkinson et al., 1999).

The location of carbon substrates within the aggregate structure appears to be affected by source-sink relationships. Priesack and Priesack (1992) observed that substrate supply at the aggregate surface induced higher microbial activity and concluded that outer surfaces of aggregates are probably the regions of highest microbial activity and biomass. Jones and Farrar, (1999) provided estimates for the rate of C loss by maize (86,400 µg C g<sup>-1</sup> root dry weight d<sup>-1</sup>). They showed that lactic acid and sugars account for the largest proportion of this loss. Greater carbon or nitrogen deposition at aggregate surfaces appears to support increased microbial activity in surface regions of aggregates. Augmented microbial activities associated with the mineralization of C should produce more essential nutrients for plant roots exploring aggregate surface layers. (Kinyangi et al., Chapter 3) identified concentric layering and sequestration of soil phosphorus (P) within soil aggregates and showed maize crop removal of plant available P from the exterior surface layers of soil aggregates (4-8mm across) when applied

P was insufficient to meet plant demand. Ladd et al (1994) reported net losses in SOM of an Australian Alfisol when P was added, during nine years of continuous cultivation.

#### Objectives of this study were:

- 1) To determine *Crotalaria grahamiana* (C<sub>3</sub>) and maize (*Zea mays* L.) (C<sub>4</sub>) carbon contributions to external surfaces and interior regions of soil aggregates.
- To determine the effect of crop rotation and seasonal variations on recent C deposits.
- 3) To assess the effect of P fertilization on shifts of C inputs into aggregate layers in the presence or absence of a maize crop.

#### **MATERIALS AND METHODS**

Two experiments were setup, both in the field and in pots. Soil samples were collected from 0 to 15cm depths of an ongoing field trial initiated in March 1997 to investigate P replenishment with tree crops. The study site is located at Osita farm in Vihiga District, Western Kenya (0°06' N, 34°34' E) at an elevation of 1420m. Mean annual rainfall is 1800mm (split between 2 cropping seasons, February-August and September to January). Often, the first cropping season will receive 1000-1200mm of the annual rainfall. The soil type is a very fine Isohyperthermic Kandiudalfic Eutrudox (USDA, 1996). It has few chemical or

physical barriers to rooting in the top 4m. Air dried soil for the top 15cm had a pH of 5.1 (1:2.5 soil:water suspension), organic carbon 17g C kg<sup>-1</sup>, ex. acidity 0.4 cmolc kg<sup>-1</sup>, calcium 3.4 cmolc kg<sup>-1</sup>, (KCL extractable), Ex magnesium 1.4 cmolc kg<sup>-1</sup>, Ex potassium 0.12 cmolc kg<sup>-1</sup>, phosphorus 2 mg kg<sup>-1</sup>, (bicarbonate-EDTA extractable). Clay, silt and sand contents were 45%, 28% and 45%, respectively.

#### **Experimental design**

The field experiment consisted of two cropping sequences of a maize monoculture and maize crop in rotation with a tree crop (Crotalaria). The tree crop was relay planted into maize during March 1997 and April 1998. Maize (HB-512, Kenya Seed Co, Kitale, Kenya), was planted at 2 seeds per hill, at a spacing of 0.75m between rows and 0.25m between plants, and later thinned to 1 plant per hill to achieve a plant population of 53,000 plants ha<sup>-1</sup>. One row of tree crop was planted for every row of maize, at the same intra row spacing, resulting in the same tree plant population. Maize crops were harvested for grain yields in August 1998 and July 1999 in the tree-crop rotation plots. Two additional maize crops were harvested in January 1998 and 1999 (continuous maize plots). All maize crops were assessed for grain yields. The tree crops were harvested in February 1998 and 1999. All fine residues and litter fall were weighed and returned to the plots, while the woody components were exported from the plots for fuel wood. All above ground maize crop biomass was removed since it was common practice for farmers to harvest the corn stover for livestock feed. We only considered the residue returned as it had a direct effect on the fertility status

of the soil. Leaf litter and pods were harvested and oven dried at 60 ° C for dry matter yield determinations.

#### Soil sampling

Soil samples were taken from the field trials on the following dates; 13

February 1998 (end tree crop season, before tillage), 7 July 1998 (post tree crop season and maturity of residual maize), 19 September 1998 (initiation of 2<sup>nd</sup> tree crop season), 13 February 1999 (end of tree crop season 2 and return to maize cropping) and 1 July 1999 (post tree crop 2 and maturity of maize). Soil samples were collected at depths of 0-15cm by gently driving cores (10cm diameter x 15cm height) into the soil surface to minimize aggregate disruption. Soil cores were loaded onto trays, secured with rubber bands and transported to the laboratory. Air-dried samples (1 kg) were sieved by manually shaking through a nest of sieves, separating them into eight size fractions (>8mm, 4-8mm, 2-4mm, 1-2mm, 0.5-1mm, 0.25-0.5mm, 0.053-0.25mm and 0.038-0.053mm). Total and delta <sup>13</sup>C signatures of C were measured in the 4-8mm aggregate fraction which contained 40% of the total carbon and nitrogen, by mass of all soil aggregates <8mm, during the tree crop season, (Kinyangi et al, unpublished data).

In addition, soil from an adjacent bush fallow site was sampled in July 1999 to provide background reference data which was compared to changes in C inputs between C<sub>3</sub> and C<sub>4</sub> cropping sequences. Smestad et al., (unpublished data) characterized this reference site in July 1998, estimating the age of the bush fallow to be 9-12 years, following its conversion from food crop agriculture.

Trees (Sesbania sesban L. Merr.), accounted for 97% (18.3 t ha<sup>-1</sup>) of the overstorey standing biomass and 94% of the understorey standing biomass (6.8 t ha<sup>-1</sup>) was contributed by *Lantana camara* L. and *Justicia flava*. Leaf and litter fall was 2.7t ha<sup>-1</sup>. In total, the above ground biomass sequestered 275kg N ha<sup>-1</sup> and 16kg P ha<sup>-1</sup>. Previously, the site had undergone a mixed cropping pattern of maize, maize-bean, cassava (*Manihot esculenta* Crantz) or sweet potato (*Ipomea batatus*) rotations, dispersed among mixtures of C<sub>3</sub> and C<sub>4</sub> weed fallows.

#### Pot experiment

The pot experiment was conducted for 5 weeks at the Maseno Research Station under a shelter housing of translucent roofing sheet to allow light penetration, with open sides provided to permit free flow of ambient air. The experiment consisted of a factorial combination of 2 crop factors (Maize, No Maize) at 2 fertilization levels (NPK and No NPK). Soil was sampled from *Crotalaria* plots in February 1998, after leaf and litterfall had been incorporated into the soil. These plant residues provided a substantial background of C<sub>3</sub> organic input. However, since there was a previous history of C<sub>4</sub> input, the initial <sup>13</sup>C signature of this soil in the pots represented a mixture of C<sub>3</sub> and C<sub>4</sub> sources. Bulked soil samples were sieved through 8mm to remove large rocks and plant debris, was air dried, weighed and NPK fertilizer was thoroughly mixed with one-half the soil treatments. Plastic pots (7I) were each filled with 4kg soil on a dry weight basis. An equal weight of gravel (<2cm) was added to the bottom of each pot to facilitate free drainage.

Fertilizer was applied as Triple Super Phosphate (46% P<sub>2</sub>O<sub>5</sub>), Urea (46%N) and Muriate of Potash (60% K<sub>2</sub>O) at 300kg P ha<sup>-1</sup>, 200kg N ha<sup>-1</sup> and 100kg K ha<sup>-1</sup>. Five seeds of maize (HB-513, Kenya Seed Co, Kitale, Kenya), were planted on 26 June 1999, in each pot receiving maize treatment. The pots were watered and brought to field capacity every 3 days for the entire duration of the experiment (5 weeks). Maize was thinned to 3 plants per pot on 11 July 1999. An initial soil sample was collected from the bulk soil to represent conditions at the start of the experiment. Additionally, soil samples for C analyses were collected from 0-5cm depth at 2 weeks (11 July 1999) and the whole pot soil, during maize harvest at 5 weeks (2 August 1999).

#### Aggregate erosion

Soil aggregates (4-8mm across), collected from the field and pot experiments, were separated into concentric layers of external, transitional and internal groupings of equal mass as described by Santos et al., (1997) by separately eroding 15 individual aggregates in stainless steel soil aggregate erosion (SAE) chambers. Individual aggregates were placed into a stainless steel chamber (2.5cm diameter), with knurled walls and secured into glass beakers (100ml) using foam packing, before mounting them onto a variable speed rotary shaker (Innova 2300, New Brunswick, Canada). The top was sealed with a rubber stopper secured with para film to prevent losses. The erosion chamber was fitted with a 356*u*m screen at the base. As the aggregate rolled around the internal knurled surface, the fine (<356*µ*m) fraction fell through the sieve and was

deposited in the lower base (Fig. 1) which could be removed and the fine eroded portion removed. Samples were weighed periodically to determine the time of removal for each third of the three concentric layer/region of the aggregate fraction sampled from each treatment. Each layer was eroded to yield 0.15g soil using the meso soil aggregate erosion (SAE) chambers according to the protocol developed by Smucker et al, (1999). The final eroded samples obtained, including the crushed interior region were passed through a 0.053mm sieve to exclude the sand fraction. Further details of this protocol were discussed by Smucker et al, (unpublished data). We concluded that exclusion of the sand portion would provide a more accurate assessment of humic C fractions of SOM, which are associated with the finer silt and clay mineral fractions of the soil. While studying SOM dynamics, Balesdent et al., (1986) reported most of the SOC is retained by the silt and clay particle sizes of soil measuring 0-50 $\mu$ m for silty clay and sandy loam soils. An equivalent number of whole aggregates were crushed and sieved through 0.053mm to compare extracted parameters from peeled layers and whole aggregates. These samples were analyzed for total C and the ratio of  $^{13}\text{C}/^{12}\text{C}$  ( $\delta$   $^{13}\text{C}$ ) of soil, in concentric layers and whole aggregates.

#### Carbon analyses

Samples were further prepared by weighing 20-40mg into pressed tin cups (8mm x 5mm). Total C content was determined by automatic dry combustion on a Carlo Erba NA 1500 elemental analyzer (Carlo Erba, Milan, ltaly), gas chromatograph. The ratio of  $^{13}$ C/ $^{12}$ C ( $\delta$   $^{13}$ C) of soil in concentric layers

of aggregates (4-8mm) was determined on a Europa Model 2020 mass spectrometer (Europa Scientific, Crewe, UK). A finely ground and precisely weighed sample was introduced to the ratio mass spectrometer via an auto sampler. Carbon is converted to CO<sub>2</sub> and carried to the triple collector, isotope ratio mass spectrometer. Isotopic compositions are expressed using the delta notation:

$$\delta^{13}C\%_0 = [(^{13}C/^{12}C_{sample})^{13}C/^{12}C_{standard}) - 1]*1000$$
 [Eq. 1]

The laboratory reference was calibrated against the international standard (Vienna-Pee Dee Belemnite). Multiple analyses of the reference samples, included after every 12 soil samples yielded a standard deviation of <0.05‰. We calculated the change in the fraction of C in concentric layers of soil aggregates resulting from conversion to C<sub>3</sub> or C<sub>4</sub> using the equation:

% C derived from maize = 
$$[(\delta^{13}\text{C layer}_{\text{final}} - \delta^{13}\text{C layer}_{\text{initial}}) / (\delta^{13}\text{C Crop}_{\text{maize}} - \delta^{13}\text{C layer}_{\text{initial}})]$$
 [Eq. 2] 
$$\text{or } \textit{Crotalaria} = \\ [(\delta^{13}\text{C layer}_{\text{final}} - \delta^{13}\text{C layer}_{\text{initial}}) / (\delta^{13}\text{C Crop}_{\text{crotalaria}} - \delta^{13}\text{C layer}_{\text{initial}})]$$
 [Eq. 3]

#### Statistical analyses

Analysis of variance was performed using the Proc GLM option of the Statistical Analysis System (SAS Institute, 1999) to investigate crop and fertilization effects on total C and shift in  $\delta^{13}$ C of whole aggregates and concentric layers of soil aggregates. All significant tests refer to  $\alpha$  = 0.05 unless otherwise stated.

#### **RESULTS AND DISCUSSION**

### δ <sup>13</sup>C of plant material

The difference in  $\delta^{13}$ C values of *Crotalaria* (pods, -25.5 ‰ ± 0.1, n =3: leaves -26.7‰ ± 0.1, n =3) and maize (roots, -10.6 ‰ ± 0.1, n =5, shoots, -10.8‰ ± 0.2, n =4) was -15.4‰. The  $\delta^{13}$ C of roots was used to calculate (Eq. 2 and 3) the contribution of C<sub>4</sub> carbon since all above ground biomass was harvested and removed from the maize plots.

#### Total carbon

Total carbon in the cultivated maize plots (19.6 $\pm$ 0.31g C kg-1, n =16) was nearly 60% lower than the non-cultivated bush fallow (33 $\pm$ 0.04g C kg<sup>-1</sup>, n =9). There was no significant effect of an initial application of 50kg P ha<sup>-1</sup>, fertilizer phosphorus to planted trees and maize on bulk soil carbon. Further, there was no significant difference for total carbon in February 1998 among crop rotations at the experiment site (mean = 19.8 $\pm$ 0.5g C kg<sup>-1</sup>, n =8). In the field experiment, we

investigated changes in total carbon of whole aggregates and concentric layers following two rotations of tree-crops with maize under moderate P fertilization (annual 50kg P ha<sup>-1</sup>, 1997-1999). A background analysis of whole aggregate C showed a significant increase (24.8g C kg<sup>-1</sup> to 26.6g C kg<sup>-1</sup>, p = 0.05) for both maize and tree-crop rotation in plots that did not receive P fertilizer (February 1998 to July 1999). At this point, the exterior to interior ratio (EIR) of C deposited in concentric layers was positively correlated ( $r^2$ =0.95, p = 0.0002) with the first season of residual maize yield in August 1998 (Fig 2). During this period, we also observed a significant rapid decline in total C of interior regions (23.8g C kg<sup>-1</sup> to 18.5g C kg<sup>-1</sup>, p<0.0001), and surface layers (25.4g C kg<sup>-1</sup> to 19.5g C kg<sup>-1</sup>, p<0.0001) under P fertilization. In the pot experiment, significant differences (p=0.03) in total C of interior regions of aggregates were detected after only 2 weeks (Fig. 3), among fertilizer levels of maize and no maize (bare soil) treatments. However, there was a highly significant trend of carbon accumulation at 2 weeks within the interior region (p<0.0001) and exterior surface layer (p<0.0001) (Fig. 3).

#### Sources of crop derived carbon

The delta  $^{13}$ C signature of SOM in the bush fallow, considered, as the reference was -21‰. This value is more negative when compared to the  $\delta$   $^{13}$ C signature recorded in the maize mono-culture plots (-17.5‰) of the field experiment site in February 1998. This suggests that a 9-12 year conversion of the cultivated site to bush fallow has resulted in an accumulation of  $C_3$  derived

carbon of the SOM of the non-cultivated fallow. Nyberg et al, 2000 studied the decomposition of *Sesbania* green manure and  $C_3$  sugar on a  $C_4$  plant dominated Alfisol (soil  $\delta^{13}$ C, ranged from -15.9% to -16.7%), in eastern Kenya. Sixteen samples of *Sesbania* leaves yielded a mean  $\delta^{13}$ C of -27.4%. Using this value and reference from the continuos maize plots to infer, we can calculate (Eq. 2) the  $C_3$  contribution to restoration of SOM at the non-cultivated bush fallow site since Smestad et al., (unpublished data) determined that *Sesbania* was the dominant tree at this site. During this period,  $C_3$  crop C contribution to SOM was 35.4% of total C.

Shifting C inputs between C<sub>3</sub> and C<sub>4</sub> sources revealed a pronounced seasonal, flux of C<sub>4</sub> carbon deposits to surface layers of soil aggregates (Fig 4). However, this flux was not apparent when whole aggregate measurements were taken, as is common in soil analysis methods (Fig. 5). This distinct shift was observed for both crop rotations in the presence of the residual maize crop (July 1998).

Seasonal variation in  $\delta^{13}$ C signatures of maize and tree crops was also investigated. Since there were no significant differences in the  $\delta^{13}$ C signatures across the tree crop season, any temporary maize contributions of C to the soil appear to have been respired. At the end of each crop season the,  $\delta^{13}$ C signature was observed to return to within variability of the initial value by February 1998. Estimates of the sources and proportions of C inputs into surface layers and interior regions of soil aggregates are presented in Table 1. In the pot experiment, we confirmed maize root depositions of C (p<0.0001) to surface

layers of aggregates but not interior regions of soil aggregates 4-8 mm across (Table 2). In addition, we recorded a more negative shift in the  $\delta$  <sup>13</sup>C signatures in fertilized and non-fertilized pots that had no maize growth (Fig 6). Maize biomass yield did not explain this difference (Table 3). This observation seemed unusual since there was no maize input. However, Balesdent, et al. (1987) explained this phenomena to be the result of a normal isotope effect as they reported respired CO<sub>2</sub> is nearly void of <sup>13</sup>C resulting in the enrichment of <sup>13</sup>C in microbial products relative to their substrate. This enrichment is thought to be in the order of 2‰. Rochette et al. (1999) separated soil respiration into soil and plant components using <sup>13</sup>C of the CO<sub>2</sub> evolved on an historically C<sub>3</sub> soil converted to maize. They maintained control plots with no maize growth and attributed the difference to the effect of maize. Maize carbon contribution to respiration was observed 40 days and peaked 65-105 days after planting. In this study, samples in the field trial were collected 98 days after planting, to coincide with the period of maximum nutrient absorption. It appears that 100 days may be close to the maximum period for seasonal carbon deposition by maize, whereas 35-40 days (as we observed in the pots) may be the minimum time. Based on the observations of Rochette et al. (1999), we propose that the maize "effect" may not last beyond 100-105 days after planting. Sanchez et al. (2000) evaluated and found that. maize roots enhanced N mineralization in the presence of a large short-term pool of nitrogen. An assessment of C mineralization, showed that a one year maize crop did not reduce potential mineralizable carbon, when the opposite was true for a one year wheat crop and bare control soil

#### CONCLUSIONS

Resource poor farmers will often be interested to maximize crop productivity when land is converted from fallow to crop agriculture. This study reveals that integrating tree crops with maize can build up soil organic carbon, when farmers cannot afford fertilizer inputs. The marginal increase in carbon under continuous maize crops is a function of poor maize yields, hardly exceeding 1 T ha<sup>-1</sup> in farmers' fields. Fertilizer applications to maize with or without tree crops resulted in significant decreases of SOC. Although fertilizer inputs are required to increase crop productivity, the magnitude of C depletion can dramatically be amplified when crops receive initial fertilization. Further research is needed to explore the extent of C-loss and the magnitude of maize C and its effect on functional pools of nitrogen and phosphorus. A mid-season influx of a large pool of soluble C is hypothesized to provide the energy required by microorganisms to decompose organic matter. Sanchez et al. (2000) and in part this paper, present preliminary evidence of a) the existence of this carbon pool and b) an indirect link between N availability in the presence of maize roots and crop productivity. On conversion to maize cultivation, these data suggest that we may not expect tree crop carbon inputs to persist beyond a single cropping season when residual maize is planted after the tree crop season. Longer term tree crops or repeated seasonal rotation with trees as was done in this experiment may potentially mitigate this loss of C and instead, enhance C sequestration.

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Table 3.1. Maize ( $C_4$ ) and *Crotalaria* ( $C_3$ ) contributions onto surface layers and interior regions of soil aggregates (4-8mm across), during the

field study, (0-15cm depth).

Crop rotation		Maize	-Maize	Crotolaria -Maize		
		+P	-P	+P	<b>.</b> P	
		C (%)				
C-Source (Jul	y- <b>9</b> 8)					
C <sub>3</sub> -C	Exterior	nd	•	•	-	
	Interior	nd	•	8.1	15.2	
C4-C	Exterior	nd	55.4	51.8	49.8	
	<b>Interior</b>	nd	4.2	•	•	
C-Source (Se	ptember-98)					
C <sub>4</sub> -C	Exterior	nd	-	nd	-	
	Interior	nd	8.8	nd	12.6	
C-Source (Fel	bruary-99)			•		
C <sub>4</sub> -C	Exterior	nd	-	nd	•	
	Interior	nd	13.0	nd	19.7	

nd, not determined

<sup>-,</sup> not calculated when <sup>13</sup>C shift was in the opposite direction relative to crop input. Initial dates for C-source determination were, February, July and September 1998.

Table 3.2. Maize ( $C_4$ -carbon) and *Crotalaria* ( $C_3$ -carbon) contributions onto surface layers and interior regions of soil aggregates (4-8mm across), during a 5-week pot study, 1999.

Crop		ize	No Maize		
	+NPK	-NPK	+NPK	-NPK	
	C (%)				
Exterior	14	22	11	20	
<sup>1</sup> Interior	-	12	11	10	
			#	*	
Exterior	23	49	-	•	
Interior	-	17	-	-	
	<sup>1</sup> Interior	+NPK  Exterior 14  Interior -  Exterior 23	Exterior 14 22  1 interior - 12  Exterior 23 49	+NPK -NPK +NPK	

<sup>&</sup>lt;sup>‡‡</sup>, <sup>‡</sup>, Calculations of carbon contributions for these layers are excluded as pots without plant growth did not receive maize carbon input.

n = 5 (maize), n = 3 (no maize)

Table 3.3. Effect of fertilizer application on above and below ground biomass of maize grown in 7 liter pots during a 5 week study, 1999.

Sho	Shoots		Roots		Root:Shoot	
+NPK	-NPK	+NPK	-NPK	+NPK	-NPK	
	g p	ot <sup>-1</sup>				
22.5	3.5	9.5	3.0	0.4	0.9	
(2.2)	(0.3)	(2.3)	(0.6)	(0.1)	(0.2)	

Figures in parenthesis are standard deviations of the means Biomass is expressed as dry matter pot<sup>-1</sup> n=5, fertilized pots n=3, non-fertilized pots

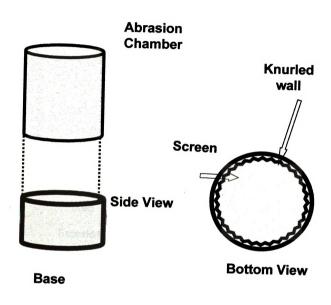
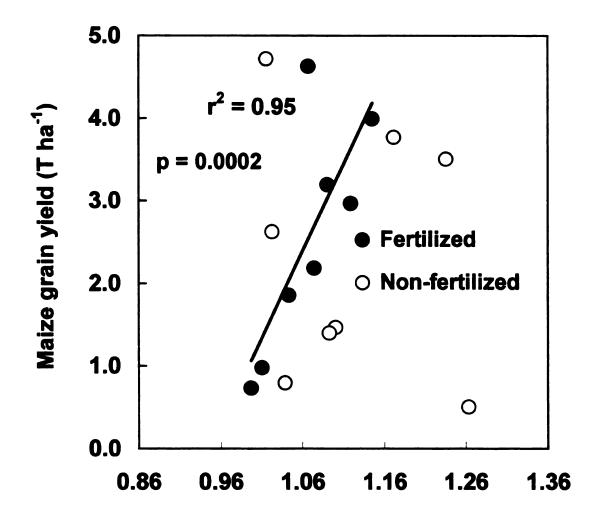


Fig 3.1 Diagramatic illustration of meso- Soil Aggregate Erosion (SAE) chamber



## Exterior to interior ratio (EIR) of total carbon

Fig. 3.2 Maize grain yield (Aug-98) as influenced by the EIR of total carbon within layers of soil aggregates (4-8mm across) for a Kenyan Oxisol. Aggregates were sampled in February 1998, from 0 15 cm depth of fertilized (annual 50 kg P ha<sup>-1</sup>) continuous maize and Crotalaria tree plots of an agroforestry management system. Regression excludes square data point. n = 4.

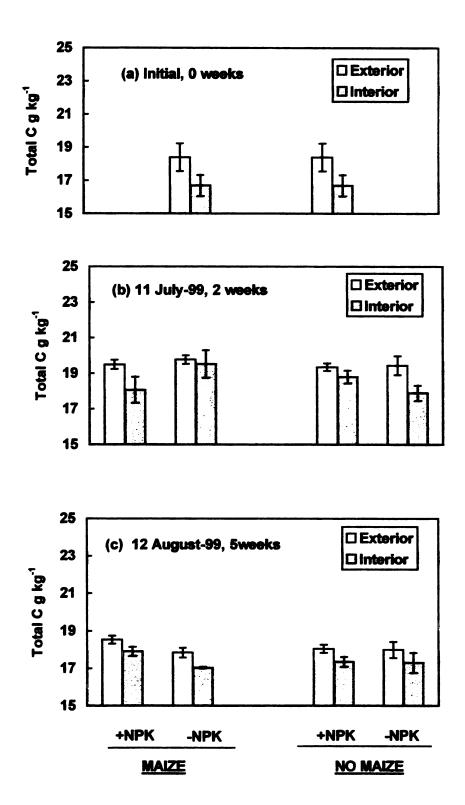


Fig. 3.3 Total carbon contributions to surface layers and interior regions of soil aggregates (4-8mm across) for a 5 week study in 7 liter containers. Bars are std. errors. n = 5 (maize), n = 3 (no maize)

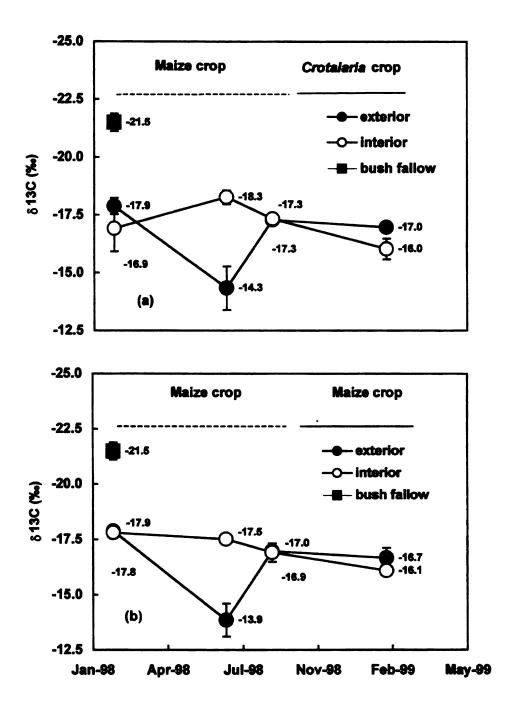


Fig. 3.4 Effect of crop rotation on seasonal variation in maize ( $C_4$ -source, -10.6 (‰), n=5) and *Crotalaria* ( $C_3$ -source, -26.1 (‰), n=4) carbon contributions to surface layers and interior regions of soil aggregates (4-8mm across, 0-15cm soil depth), of a Kenyan Oxisol. Reference soil was sampled form a bush fallow (9-12 years since conversion from agriculture) at Osita Farm in July 1999. Bars are std. errors for the difference in means. n = 4.

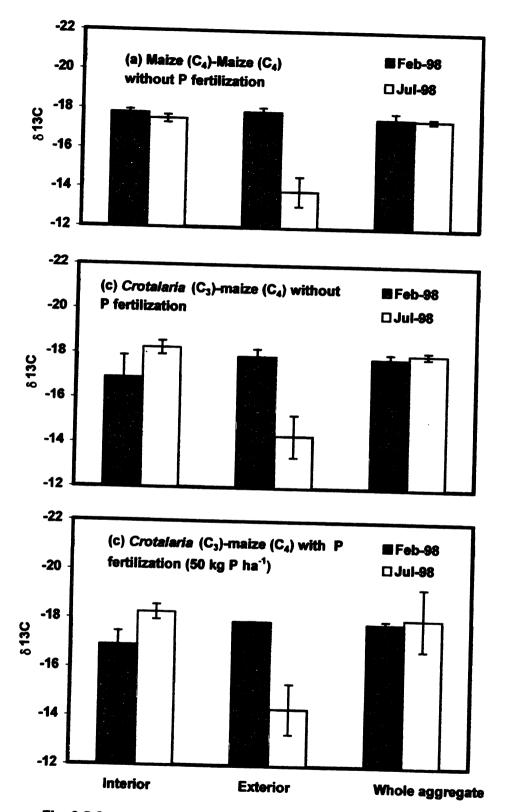


Fig. 3.5 Crop rotation and fertilization modifications of carbon deposition onto surface layers and interior regions of soil aggregates (4-8mm across), for a Kenyan Oxisol. n=4

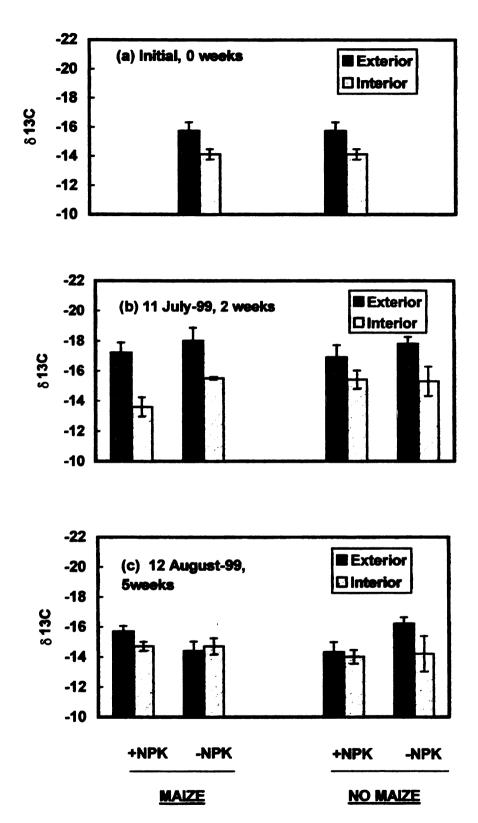


Fig. 3.6 Maize carbon contributions onto surface layers and interior regions of soil aggregates (4-8mm across) for a 5 week study in 7 liter containers. Bars are std. errors. n = 5 (maize), n = 3 (no maize)

#### Chapter 4

#### **SUMMARY AND CONCLUSIONS**

Integration of tree fallow crops with food crop farming in low input cropping systems provides a short term solution to managing carbon, nitrogen and phosphorus stocks in soils within small holder farms in Africa. When the overall goal is to sequester carbon in order to build the fertility status of the soils the use of shrubby and woody perennials has been attractive to farmers due to the abilities of the trees to fix large quantities of above and below ground biomass. Tree-crop rotation with maize did not lead to short-term reductions in SOC. Whereas the total biomass production for Crotalaria and Tithonia are comparable, their contributions to soil fertility greatly contrasts. Crotalaria returned significantly more biomass to the cropland (58% of the total) while Tithonia returned significantly less biomass (11.3% of the total. Although the tissue NPK concentrations for Tithonia are higher than Crotalaria, these differences in biomass production resulted in lower nutrient inputs (kg ha<sup>-1</sup>) recycled with Tithonia (36N, 4P and 36K) in contrast to Crotalaria (143N, 10P and 67K). Overall, these nutrient inputs are low when one considers maize crop requirements. It is estimated that a 2T ha<sup>-1</sup> grain and 3T ha<sup>-1</sup> stover maize crop would remove approximately 66 kg N ha<sup>-1</sup>, 18 kg P ha<sup>-1</sup> and 60kg K ha<sup>-1</sup>.

It can be concluded that, of the three systems studied, *Crotalaria*-maize rotations are the most appropriate for fertility maintenance of nutrient-depleted soils. However, *Tithonia*-maize rotations may still be important where fuel wood

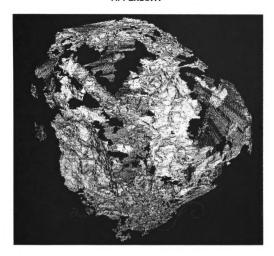
is a critical requirement for households. *Tithonia* yielded twice the amount of wood produced by *Crotalaria*. Supplemental inputs of inorganic P fertilizers applied during tree-crop establishment greatly enhanced tree fixation of C. Unfortunately, most of the biomass due to additional fertilizer is exported as wood resulting in a net mining effect. This is further evidence that suggests C losses were accelerated under modest fertilization. Greater N cycling by *Crotalaria*-maize rotation suggests the need for higher P fertilization in order to utilize these plant N inputs if yields were to be increased in this cropping system.

The lack of differences in plant available P among cropping systems for the duration of the study raises doubts whether current analyses of bulk soils are able to capture the effects of specific processes that appear to be mediated by aggregate structure and function. The locations of plant available P from recent applications of plant residues and fertilizer materials, and the resulting relationships with grain yield suggest that these processes associated with nutrient recycling are closely linked to the activities of growing plants. These interactions appear to be rapid with high seasonal turnover. The low P status of bulk soil and associated poor maize growth may erroneously suggest the need for more P fertilization when accumulations of plant available P are occurring within specific layers of soil aggregates. The large influx of maize carbon onto surface layers of soil aggregates observed during maize growth in the third study raises further questions on how plants acquire mineral nutrients. There is speculation that these C compounds contributed by maize are primarily made up of sugars and lactic acid (Jones et al, 1999) Perhaps this "maize effect" leads to

N immobilization in continuos cropping systems. In contrast, increased N inputs from biological N fixation and residue return, further lead to adequate supplies of available N in *Crotalaria*-maize rotations when the N is in excess of requirements by microorganisms involved in C respiration. These micro-nitche and rapid changes that seem to occur under tropical cropping conditions are concealed by current analyses of bulk soils. Integrating concentric separations of soil aggregate layers, using meso SAE chambers, into soil testing methods will greatly contribute to our understanding of short-term factors affecting management modifications of C, N and to maximize input utilization efficiencies of P into low input sustainable agriculture systems of the tropics.

**APPENDICES** 

#### APPENDIX A



- Source: Albee P B, Stockman G C and Smucker A J M, Advanced Photon Source at Argnne National Laboratory, January, 2000.
- Reconstruction of a microtomographic image of a soil aggregate (1.1 mm across) from Kenya, rendered by algorithms at the APS. Light areas represent surface breaking and internal pores >63 microns, while black areas are the solid portions of the aggregate and surrounding air. The light tubular area within the middle appears to be a root induced macropore (RIM). The aggregate was sampled from an Oxisol soil in July 1998.

