

**EFFECTS OF CHEMICAL COMPOSITION OF PLANT RESIDUES ON
NITROGEN RELEASE AND CROP UPTAKE**

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*A THESIS SUBMITTED IN PARTIAL FULFILMENT FOR THE AWARD OF THE
DEGREE OF MASTER OF SCIENCE, DEPARTMENT OF SOIL SCIENCE,*

UNIVERSITY OF NAIROBI.

SEPTEMBER, 1997.

DEDICATION

To Gladys

DECLARATION

This Thesis is my original work and has not been presented for a degree in any other University.

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This thesis has been submitted for examination with our approval as Supervisors.

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ABSTRACT

To supplement high cost of inorganic fertilizers, smallholder farmers in the tropics are likely to increase the use of plant residues as a suitable source of plant nutrients especially nitrogen (N) and phosphorus (P). Management of these organic N - sources demand that their N - release patterns coupled with synchronization of the released N with crop growth be fully understood. Consequently this study was undertaken to evaluate the effect of chemical composition of various residues on N - release, improve the N-release pattern of low quality organic materials by mixing them with those of high quality and synchronize the N released with crop production.

These objectives were achieved through a series of controlled experiments. The experiments involved incubation, in the laboratory for 12 weeks, of six selected plant residues which involved leaves of *Leucaena leucocephala*, *Croton macrostachyus*, *Calliandra calothyrsus*, *Tithonia diversifolia*, *Sorghum bicolor* and husks of *Oryza sativa*. Parallel to this, mixtures of *C. macrostachyus* (Cm) and *O. sativa* (Os) in various ratios were also incubated. Finally maize was grown in the glasshouse in pots whose soil had been amended with *C. macrostachyus*, *O. sativa*, *T. diversifolia* (Ts) and ½: ½ mixture of *Croton macrostachyus* and *Oryza sativa*. In the laboratory incubation, soil samples were taken after every 2 weeks for analysis of ammonium nitrogen ($\text{NH}_4^+\text{-N}$) and nitrate nitrogen ($\text{NO}_3^-\text{-N}$), while in the glasshouse experiment, harvesting of maize tops was done at 2 week intervals 8 weeks and the shoots' dry matter as well as N content determined.

Two patterns of N (NO_3^- -N + NH_4^+ -N) release were observed during the 12 week incubation period: Leaves of *C. macrostachyus*, *L. leucocephala*, *T. diversifolia* and *C. calothyrsus* had a net release throughout the incubation period while *S. bicolor* leaves and *O. sativa* husks showed significant net immobilization. Due to concurrent nitrification over the 12 week incubation period, 65 - 80% of the accumulated mineral N was in nitrate form.

The dynamics of N - mineralization of the various mixture of *Croton macrostachyus* (Cm) and *Oryza sativa* (Os) were in general not significantly ($p \geq 0.05$) different from those predicted from the isolates of *Oryza sativa* (Os) and *Croton macrostachyus* (Cm) alone with the exception of the 3/4 Cm + 1/4Os having depressed the incubated N - release in weeks 6 - 8 significantly while the 1/4Cm + 3/4Os stimulated the incubated N-release at weeks 2 and 12 weeks of incubation respectively.

Addition of plant residues increased maize biomass in the glasshouse significantly ($P < 0.05$) throughout the growth period. However, the study showed a sharp contrast of maize response from the results of the incubation. Mixture of plant residues in the pots had the highest contribution to maize dry matter yield and N uptake. It could be speculated that high C:N ratio *Oryza sativa* material in the mixture stimulated microbial activity in the rhizosphere leading to high organic material decomposition.

Of the chemical variables studied, initial contents of N and P as well as C:N and polyphenol:N ratios were significantly correlated with cumulative N mineralized. Nitrogen- release was best correlated with C:N ratio having $r = -0.84$ to -0.90 for most of the sampling periods. Polyphenol: N ratio also gave high correlation with cumulative

N mineralized with correlation coefficient (r) ranging from -0.65 to -0.95. Initial N and P contents showed positive significant correlation but not as high as those of the two ratios. These results show that the best predictors for N mineralization were residue C:N and Polyphenol:N ratios.

CHAPTER ONE

1.0 INTRODUCTION

Nitrogen is an essential nutrient element for plant growth and therefore needed in adequate supply for normal development of crops. However nitrogen is the most deficient nutrient in cultivated soils (Jones, 1982), the element that most frequently limits yield in the tropics (Sanchez, 1976) and generally the first element to become deficient in semi-arid and arid regions (Hagin and Tucker, 1982). Nitrogen usually occurs in small amounts ranging from 0.02 to 0.4% by weight in the plough layer of majority of cultivated soils (Barber, 1984). Unfortunately, most of this N is not available to crops at any one particular time because most of it is organically bound (Jones, 1982).

In traditional agriculture, which was characterized by low population density, the restoration of soil fertility was achieved through shifting cultivation and bush fallow systems (Nye and Greenland, 1960; Okigbo, 1977). In Kenya the maintenance of soil fertility prior to 1940s was dependent very largely on shifting cultivation (Graham, 1941). In later years, tremendous increase in human population (Jaetzold and Schmidt, 1982), caused a very high demand for land. Hence, shifting cultivation could no longer support pressure on land and maintain soil fertility in this region without decline in soil productivity (Bationo and Mokwunye, 1991).

In Kenya, growing of crops continuously without addition of manure or commercial fertilizers has resulted in depletion of nitrogen, phosphorus and potassium especially in Kisii and Machakos districts (Smaling, 1993; Okalebo *et al.*, 1993a). According to Stoorvogel and Smaling (1990), Kenya was among the countries with the highest nutrient

depletion in Sub-Saharan Africa in 1983 with depletion of > 40, 15 and 40kg ha⁻¹ year⁻¹ N, P and K respectively while the average N and P fertilizer consumption in 1986-1992 in Kenya was as shown in Table 1 below.

Table 1. Fertilizer consumption in Kenya, 1986-1992

Years	¹ Fertilizer consumption (MT)		² Total land under arable and permanent cropping (ha)	Fertilizer consumption (Kg/ha)	
	N	P ₂ O ₅		N	P
1986	35961	38027	2370000	15.17	7.01
1991	57000	51000	2440000	23.36	9.13
1992	51000	35600	2460000	20.73	6.32

Source:

¹ FAO Fertilizer Year book, 1993, Vol. 42, Rome, Italy

² FAO Production Year book, 1992, Vol 46, Rome, Italy.

From this data, it is evident that nitrogen is one of the most depleted nutrient in Sub-Saharan Africa including Kenya yet its replacement in Kenya, as indicated in Table 1 is below its depletion in kg/ha. In this situation, where soil is depleted in N, the use of commercial fertilizers to maintain high soil productivity becomes very crucial. Unfortunately, the smallscale farmers who are the majority in Kenya use commercial fertilizers below the recommended levels and some do not use them at all, (Jaetzold and Schmidt, 1982). This is probably due to economic reasons like continuous increases in prices of fertilizers as shown by FAO (1994) in Table 2.

Table 2. Prices¹ of Nitrogenous Fertilizer in Kenya, 1982 -1993

Years	Prices paid by farmers for Sulphate of Ammonia (SA), (KSh/MT)
1982	10362
1985	13771
1990	18119
1993	21000

Source: FAO Fertilizer Yearbook, 1994, Vol. 43 Rome, Italy.

¹ Constant 1982 prices

The high prices of fertilizers and unavailability means that cheaper alternative sources of N that could supplement commercial N fertilizers in crop production should be sought. The best option of maintaining soil fertility and productivity is through periodic additions of properly processed organic materials. These organic materials can contribute substantial amounts of macro- and micro-nutrients for crop growth, in addition to improving soil physical and biological properties (Young, 1989).

A wide range of plant residues introduced into the soil have varying effect on the subsequent crop production (Tian *et al.*, 1993). For proper crop management, more information is needed about the factors affecting the release of N from such materials. Most previous investigations on the decomposition of agricultural plant residues have focused on the net mineralization of N (accumulation of NH_4^+ + NO_3^-). Those

investigations have shown that the concentration of lignin, polyphenols, N and the C:N ratio of the plant residues are important characteristics governing decompositions. When the concentration of lignin-to-N ratio increases, there is slower decomposition and N-release (Melillo *et al.*, 1982). Polyphenol content or polyphenol-to-N ratio has been shown to serve as an index for short term immobilization of N pattern observed for legumes with relatively high polyphenol content (Palm and Sanchez, 1991; Oglesby and Fownes, 1992). When C:N ratio increases or the N concentration decreases, then the decomposition rate of the organic material and net mineralization of N decreases. Net mineralization of N from plant residues during laboratory incubation does not necessarily reflect their actual N contribution to the crops in the field. This was shown by Tian *et al.*, (1993) when he used a low quality plant material high in lignin and polyphenols but low in nitrogen contents e.g. *Acioa barteri*. During this study he found out that when the material was applied to maize in the field during low rainfall periods, the maize produced higher yields than expected from the N-released during laboratory incubation. The high yield was attributed to the low quality material having a greater modifying effect to the soil microclimate than the higher quality materials (materials high in nitrogen but low in lignin and polyphenol contents).

There is therefore need to find a means of selecting plant materials that can serve as efficient and sufficient sources of plant nutrients for crops. It is with this background information that Swift (1987) presented the following hypothesis: " high quality organic plant residues release N rapidly initially in excess of plant demand and that there is a risk of N released being lost through volatilization, leaching or denitrification. On the other

hand, low quality plant residues when incorporated into the soil leads to low N release or immobilization leading to deficiency. However, synchronising nutrient supply with crop's demand might be achieved by a mixture of high and low quality materials." This hypothesis has been supported by several authors in the past who have used N-fertilizer as a quick N-releasing material. For example, Amarasiri and Wickramasinghe, (1988) showed that rice receiving 60 kg ha⁻¹ fertilizer and straw give grain yields that were about as high as that receiving 90 kg ha⁻¹ as fertilizer alone. It was concluded that the straw may have reduced N losses from the fertilizer through immobilization and then released the N in consistence with rice demand. An example in which mixtures of high quality and low quality materials might lead to synchrony is found in the field experiment of Bandara and Anderson (unpubl. data) as is described in Myers *et al.*, (1994). In this field experiment, there was comparison of N-mineralization from cereal straw, from a legume (*Gliricidia sepium*) and various mixtures of the two. Their results showed that the cereal straw immobilized N in the first four weeks, followed by slow mineralization. The *Gliricidia* material released N rapidly from four weeks while the mixture released N at an intermediate rate, gradually increasing during the 12 week experiment. It was also observed that there was an enhanced nutrient uptake in the mixture treatment between weeks 6 and 10. Apart from this example, there is lack of information on the use of high quality in mixtures with low quality organic residues instead of N fertilizers. Also little is known on the synchrony of N released from the plant residue mixture with N uptake and yield by a maize crop.

1.1 Justification of the study

Since the cost of inorganic fertilizers are increasing as was shown in Table 2., and their future availability is uncertain due to world energy crisis, there might be an increase in demand for plant residues to serve as sources of plant nutrients. But, some of the plant residues are of very low quality (i.e having low nitrogen content and high lignin and polyphenolics). Due to this they might in some occasions lead to immobilization of available N in the soil leading to nutrient deficiency symptoms to some crops (Ocio, *et al.*, 1991; Singh and Singh, 1994; Beri, *et al.*, 1995). A long-term experiment in Kenya of soil amendment with low- quality resources e.g maize stover has shown that they have low nutrient contribution to the plants grown leading to nutrient, especially nitrogen, deficiency symptoms (Qureshi, 1987). There is need, therefore to carry out studies to characterize some of the commonly available residue resources on the farms, to obtain their mineralization patterns with an aim of synchronising N release with uptake by a test crop.

1.2 Objectives of the study

The broad objective of this study was to quantify the nutrient content of the farmer available plant residues, quantify N-released from selected residue materials as well as from their mixtures and their contribution to maize growth. This was to be achieved through the following specific objectives;

1. Determination of chemical composition: nitrogen, carbon, lignin, polyphenol and other plant nutrients (Ca, Mg, K and P) of various onfarm plant materials and industrial wastes.
2. Assessment of the effect of chemical characteristics of plant residues on the amounts and patterns of mineral-N release.
3. Evaluation of the effect of mixing low quality and high quality plant residues on amounts and patterns of mineral N- release.
4. Relate crop response to the N-released from various plant residues and their mixtures using maize as a test crop in the glasshouse.

CHAPTER TWO

2.0

LITERATURE REVIEW

2.1 Soil nitrogen

Soil nitrogen originates from various sources which include the fixation of elemental N by biological agents, the acquisition of ammonia and nitrate in rain water and addition of nitrogen fertilizers either in mineral or organic form (Smaling, 1993). The total N in the soil ranges from 0.02% to more than 2.5% in peat and 0.02 to 0.4% in plough layer of most cultivated soils (Brady, 1990). The amount of nitrogen present in a particular soil is determined by the interaction of environmental factors, microbial activities and man (Haynes, 1986).

Over 90% of nitrogen in the surface layer of most soils is organically bound (Jones 1982). The organic form of soil nitrogen contains amino-acids and traces of other nitrogenous organic compounds (like amino sugars and amines) that comprise more than 40% of the total N in most soils (Bremner, 1965). Organic nitrogen in soils is not directly available to plants until the organic nitrogenous compounds are mineralized to release the inorganic forms i.e Ammonium and Nitrate ions (Novoa and Loomis 1981). Interestingly, the organic N in soils resists microbial attack and hence at any one moment inorganic nitrogen accounts for less than 2% of the total soil nitrogen (Melillo, 1981). Thus, the need to improve the proportions of inorganic N in soils has attracted the profound interest of the soil biologists and fertility specialists.

2.2 Available (mineral) nitrogen ($\text{NH}_4^+ + \text{NO}_3^- - \text{N}$)

Agronomically, ammonium (NH_4^+), nitrate (NO_3^-) and nitrites (NO_2^-) are the most important forms of inorganic nitrogen in the soil (Haynes, 1986). Nitrate and nitrite occur exclusively as free diffusible ions in the soil solution whereas most of the ammonium occurs in exchangeable or non exchangeable form (Bajwa, 1982). Soils with variable charge mineralogy can show a net positive charge in the subsoil in which case NO_3^- ions are bound to the sites and accumulate (Sanchez, 1976). The ratio of NH_4^+ to NO_3^- found in the soil depends on the presence of satisfactory conditions for nitrification which is inhibited by low soil pH and anaerobic conditions (Barber, 1984). Nitrites accumulate in alkali soils after application of high rates of ammonium fertilizers because ammonium inhibits the oxidation of nitrites by nitrobacter bacterial species during nitrification (Keeney and Nelson, 1982).

2.2.1 Transformation of nitrogen between organic and inorganic phase

The transformation of organic nitrogen to plant available inorganic state is called nitrogen mineralization. Nitrogen mineralization occurs via two steps, namely, ammonification (production of ammonium from organic matter) and nitrification (conversion of ammonium to nitrate) (Jansson and Persson, 1982). Immobilization is the conversion of inorganic (NH_4^+ and NO_3^-) nitrogen to organic forms due to synthetic reactions associated mainly with microbial growth and metabolism. While mineralization releases mineral nitrogen for plant uptake, immobilization reduces the amount of available N through incorporation into microbial biomass. The mineralization of decomposing

residues is a major source of available nitrogen in highly weathered soils with little inherent soil fertility (Sanchez *et al.*, 1989).

The process of ammonification is performed by heterotrophic soil organisms that utilize nitrogenous substances as an energy source (Janssen and Persson, 1982). Enzymatic conversion of organic nitrogen substances yielding ammonium ions is considered the step limiting the rate of nitrogen mineralization (Stanford and Epstein, 1974). Ammonification is an important microbial process, not only because it supplies the raw materials for nitrification process but also a readily available nitrogen source for plants such as strawberry and certain grass species like rye (Gashow and Mugwira, 1981). It can also be regarded as a 'preferred' nitrogen form as it is less susceptible to leaching than nitrates, hence used more efficiently by plants (Harris, 1989).

Some of the ammonium ions released by the process of ammonification are converted to nitrate nitrogen. This is biological oxidation process by autotrophs which obtain their carbon from carbon dioxide and energy from NH_4^+ and NO_2^- oxidation (Schmidt, 1982). However, if the rate of nitrification is faster than plant demand, some of the nitrate ions so formed are easily lost from the soil through denitrification and leaching.

Net mineralization, therefore, needs to be slow when plant needs are low and rapid when plant needs are high; thus there should be synchrony between release and plant uptake for enhanced nutrient use efficiency in agricultural systems. This may be achieved by mixing high quality plant residues (those that release N very fast) with low quality plant residues (those that release N slowly).

2.2.2 Factors affecting nitrogen transformation between organic and inorganic phases

The rate of nitrogen mineralization depends largely upon the following factors: soil moisture, temperature, quality and quantity of organic resources incorporated in the soil, soil factors such as texture, mineralogy and acidity, biological activities and presence of other nutrients like phosphorus, calcium and magnesium (Myers *et al.*, 1994). These factors are now discussed:

2.2.2.1 Soil moisture

The effect of moisture content on mineralization depends on its effect on aeration. It has been noted that mineralization is positively correlated to moisture until the surface pores are about 50-60% water filled (Mazzarino *et al.*, 1991; Doran *et al.*, 1988). Higher moisture content decreases aeration and microbial growth (Walters *et al.*, 1992). Under such anaerobic conditions, decomposition of organic residues becomes increasingly dependent on anaerobic bacteria which inhibit the nitrification step and encourage denitrification.

Birch (1964) working in Makerere Uganda, with five grass species namely Meadow hay, Kikuyu grass (*pennisetum clandestinum*), Rye grass (*Lolium perenne*), tufted grass (*Pennisetum schiimperi*) and Rhonfa grass (*Phalaris tuberosa*), reported that intermittent drying enhanced the amount of mineral N released from four of the five grass species, with an increasing effect as the decomposition was at an advanced stage. Similarly, De Bruin *et al.*, (1989) observed that mineralization of N from soil during periods of

alternate wet and dry conditions was double that under continuously moist conditions. The rapid release of N following re-wetting is the result of lysis of soil microbes exposed to the sudden change in water potential (Kieft *et al.*, 1987). When the soil water potential increases suddenly, water moves into the cell faster than the organism is able to adjust its osmotic potential resulting in rupture of the cells (Scholes *et al.*, 1994). More than 17% of the soil microbial biomass can be lost in that fashion with each wetting event (Kieft *et al.*, 1987). Another explanation for the rapid release of N during intermitted drying is: re-wetting mineral soils may expose otherwise inaccessible nutrients by disrupting soil aggregates (Adu and Oades, 1978).

2.2.2.2. Temperature

In addition to sufficient available nutrients and water, soil microbial activity and hence decomposition is largely controlled by temperature. Scholes *et al.*, (1994) reported that N-mineralization and soil respiration rates increase exponentially with soil temperature over a range of 10-30°C. Many studies have quantified the influence of temperature on the rate of litter decomposition and soil respiration. Moore (1986) carried out a laboratory study to relate the decomposition rates of hardwood and coniferous leaf litter with temperature and moisture. Decomposition rate was found to be a linear function of the logarithm of water potential and approached a maximum near 40°C. The temperature dependence of litter decomposition was found to be consistent with a model based on irreversible heat inactivation of a rate-controlling enzyme. This indicates that decomposition, like any other enzyme controlled reaction, requires optimum temperature

to achieve a maximum rate. Nyhan (1976) studied the decomposition of blue gamma residues in relation to temperature. His results showed a positive linear relationship between %carbon loss and soil temperature over the range of 2°C to 60°C but CO₂ was released at maximum rate between 20 and 30°C.

These observations could be explained by the fact that common soil inhabitants are mesophyllic bacteria, actinomycetes and fungi which grow at a soil temperature range of 0 to 40°C (Alexander, 1977). Thermophilic bacteria and actinomycetes are minor soil inhabitants and have optimum temperature range of 45 to 65°C. However, thermophiles play an important role during composting of plant residues. In the composting system, heat is generated by decomposition process raising internal temperature to 70-80°C (Swift *et al.*, 1979); and is dominated by populations of thermophilic bacteria, actinomycetes and fungi. Similarly, the surface layers of exposed tropical soils may reach temperatures greater than 45°C (Lal, 1986). Similar conditions occur when ambient temperatures are high and catabolic activity is generating heat within well-insulated microsites. Under such circumstances, thermophilic microorganisms may be important in maintaining decomposer activities.

2.2.2.3 Soil pH

Soil pH is divided into two categories: local pH (pH of immediate microbial environment) and bulk pH (pH of wider environment) (Swift *et al.*, 1979). Of these two, the local pH determines the microbial activity while the bulk pH is useful as an indicator of microbial distribution in the soil. The lower the local pH the greater the concentration

of soluble aluminium, iron and manganese ions. These elements are not only toxic to decomposers but also react with phosphate ions making it unavailable to the decomposers (Tisdale *et al.*, 1990). Ishaque and Cornfield (1972) observed that other factors being constant, production of inorganic nitrogen is greater in neutral than in acid environment. This could be explained by less microbial activity at low pH environments. Acidification tends to depress but not eliminate N mineralization (Haynes and Swift, 1988). This is why whenever residue amended acid soils are limed a rapid release of mineral N is noted (Fu *et al.*, 1987).

2.2.2.4. Effect of organic resource quality

Quality of organic materials generally refers to the relative rates of decomposition and nutrient release of these materials, which is determined in large part by their chemical composition. Rapid decomposition and nutrient release are associated with high quality and conversely, immobilization or slow release refers to low quality (Swift *et al.*, 1979).

The initial nitrogen and carbon contents, both of which influence the C:N ratio, have been considered to be the most important factors governing decomposition and N-release from organic residues added to the soil (Frankenberger and Abdelmagid, 1985). According to Palm (1995), it has been suggested that for crop residues and cover crops, low quality materials are the ones that contain less than 1.73% N content or C:N ratio of more than 20. These materials if added to the soil will cause immobilization of soil nitrogen. Otherwise, if organic residues contain an N content more than 1.73% or C:N

ratio of less than 20, they will probably show net N mineralization. If material with N% greater than 2 leads to immobilization of N, then factors other than the C:N ratio are considered to be influencing their N release patterns.

Apart from the C:N ratio, there are other modifying factors that influence mineralization of organic materials. The lignin concentration or the lignin:N ratio improves the prediction of the N release patterns under natural ecosystems (Melillo *et al.*, 1982). In fact it was suggested by Berg and McClaugherty (1987) that N is not released from litter until decomposition of lignin commences. The role of lignin as a regulator in the decomposition process can be attributed to the fact that: lignin is a recalcitrant substance, that is, it is highly resistant to microbial decomposition, (Melillo *et al.*, 1982; Spain and Le Feuvre, 1987). It has been found out that lignin with two phenolic hydroxyls could bind considerable amounts of N, part of it which was resistant to 72% sulphuric acid or strong alkali extraction (Bennet, 1949). Lignin is capable of reducing the availability of both carbohydrates and proteins by complexing them. Based on this, Swain (1979) suggested that lignin could also influence N release rates.

With high N content, leguminous plant materials are expected to release N with ease. however, Vallis and Jones (1973) found that this did not hold true with respect to *Desmodium intortum* pasture legume. They associated this phenomenon to the high polyphenol content of the legume. Similar effects of polyphenols lowering N-mineralization patterns of organic residues were reported by Sivapalan *et al.*, (1985). At Yurimagus, Peru, Palm and Sanchez (1991) examined a range of ten plant leaf materials to determine how their chemical composition (lignin, N, and polyphenol contents) could

affect their N mineralization. After an eight week incubation period, they found that regression against polyphenolic:N ratio explained 76% of the variation in N release. They concluded that polyphenol:N ratio is the best index for N release patterns. Similarly, results of Oglesby and Fownes (1992) indicated the importance of polyphenol:N ratio in controlling N mineralization from legumes. In their work, they found that in spite of the small correlation with nitrogen or polyphenol content alone, the relation of N-mineralized with polyphenol:N ratio appeared stronger and more consistent than polyphenol alone. Tian *et al.*, (1992), working with a range of tropical woody and herbaceous residues, found their decomposition was strongly correlated with N, lignin and polyphenol concentration.

A study conducted by Fox *et al.*, (1990), using the shoots of many tropical legumes, showed that net N release after a 12-week incubation period, ranged from 11% to 47% of the initial N content of the shoots. In this study, the best chemical characteristic that predicted N mineralization was (lignin + polyphenol) to N ratio. The influence of polyphenols on N mineralization could be associated with one or all of the following three reasons: some polyphenols are capable of binding with plant protein (Haslam, 1989), thus reducing the release of N from decomposing plant materials. Polyphenols are known to be disinfectant and bacteriacide (Stokes, 1977). A high polyphenol content in plant residues can therefore slow down the decomposition by lowering the activities of microorganisms and enzymes especially where soil bacteria are the majority of decomposing organisms. Azhar *et al.*, (1986) found that phenolic compounds bind mineral N in nitro and nitroso forms in soil humus leading to less availability of the N to the crops.

Based on these analyses, Palm (1995) suggested that N content is the primary factor while polyphenol content is the secondary factor in determining the percentage of initial N released from decomposing organic materials. She proposed that polyphenol to N ratio may serve as an index for short term N immobilization patterns commonly observed with legumes having relatively high polyphenol content while lignin plus polyphenol to nitrogen ratio may serve as an index for longer term release patterns.

2.3 Nitrogen uptake by plants

The soil solution contains both nitrate and ammonium ions. Both these ions are absorbed by plants via an energy dependent uptake mechanism (Haynes, 1986). Plants may have seasonal preference for either ammonium or nitrate depending on plant species, plant development stage and environmental factors (Abbes *et al.*, 1995; Serna *et al.*, 1992). However, for most agricultural soils, nitrate is the main source of N since ammonium is readily absorbed on soil colloids and does not normally move as readily, by mass flow, as nitrate does to the root environment (Barber, 1984). Utilization of nitrate in plants include uptake, storage, translocation, reduction and incorporation of N into plant organic forms. Nye and Tinker (1977) emphasized the relative role of mass flow in supply of nitrogen to the plant roots and suggested that only a small proportion of nitrogen supply to the roots may come from diffusion. This is confirmed by Foth and Ellis (1988) who estimated that 79% of nitrogen at the root surface was there through mass flow, 20% through diffusion and only 10% through interception. In cases where ammonium is the only nitrogen source available for plant uptake, cation uptake exceeds

anion uptake and hydrogen ions are released from the roots into the rhizosphere creating an acid environment around the root environment. On the other hand, if nitrate is the only source, anion uptake will exceed cation uptake and OH^- and HCO_3^- ions are secreted into the rhizosphere leading to increase in soil pH (Barber, 1984).

Temperature and solution pH are some of the factors that influence relative rates of uptake of ammonium and nitrate ions. Ammonium ion absorption increases as pH increases, whereas nitrate ion uptake tend to decrease with increasing pH (Haffaker and Rains, 1978). When both ions are present, absorption of ammonium ions is greater than nitrate ions at 8°C but ammonium influx reaches maximum at media temperature of 25°C (Novoa and Loomis, 1981).

2.4 Types of organic resources in Kenya.

There are several organic resources in Kenya that could be used as organic fertilizers. The most common ones can be grouped into four categories according to Janssen (1993).

- (i) Farm wastes: crop residues like cereal straws, green manures and sugarcane trash.
- (ii) Agro-industrial wastes from processing plants: fibres from paper industry; husks from coffee, barley and rice processing industries; wood material like bark, sawdust and wood chips and other residues from sugar industry like filtermud.
- (iii) Agroforestry plant prunings especially from alley cropping and live fences: *Leucaena leucocephala*, *Calliandra calothyrsus* and *Croton megalocarpus*.
- (iv) Municipal wastes: composted household refuse and sewage sludge.
- (v) Household animal wastes eg cattle, poultry, pig and rabbit manures.

For the above materials to be used as options for improving soil fertility, there is need to understand their biophysical and socioeconomic factors that affect their use (Swift *et al.*, 1994) The biophysical factors referred here are biological processes that regulate their nutrient release while socioeconomic factors are the determinants that make their use socioeconomically practical within the context of the farm management system e.g their availability, cost, accessibility and acceptability by the farmer. However, entry into their socioeconomic determinants comes only after an extensive process of their characterization and nutrient release patterns have been conducted.

2.5 Organic resource use as a source of plant nutrients

The results of long-term additions of organic materials to soil have been shown to increase soil organic matter, crop productivity and soil biological activity (Collins *et al.*, 1992). High rates of animal manure can sustain crop yield (Bouldin *et al.*, 1984). However, studies utilizing typical farm-scale management practices have shown that replacement of inorganic with organic (animal and green manures) N sources results in unacceptable yield reduction during the first few years of transition (Doran *et al.*, 1987). This low yield could be because the N use efficiency of plant residues of a first crop is low (5-25%) compared 60% of N applied in organic fertilizers (Myers *et al.*, 1994). The possible reason for the low N use efficiency is the partitioning of the incorporated organic residue-N into mineral N and humic N pools with a major proportion going into organic matter pool (Ladd *et al.*, 1981; Feller *et al.*, 1983). Most of the humic N goes to labile organic matter fraction making it more readily mineralizable in the subsequent seasons

(Janzen *et al.*, 1990). Thus for a successful transition from inorganic to organic N sources, there must be enough short term N availability from organic N sources to maintain crop productivity (Fauci and Dick, 1994). Consequently short term N mineralization from a variety of organic resources needs to be fully understood for better management of agroecosystems by use of these organic resources.

2.5.1 Use of important crop residues

Common crop residues in Kenya include maize stover and bean husks; rice, sorghum, millet, and wheat straws; groundnut and sugarcane trash and potato vines together with roots of these crops (Karanja, Personal comm.; Amolo, 1995). Rice straw and husks could be important in rice growing areas like Ahero, Mwea Tebere and their surroundings. Rice straws have been used in other research work where they have shown marked improvement in crop performance both in the greenhouse (Broadbent and Nakashima, 1965) and in the field experiment with flooded rice (Amarasiri and Wickramasinghe, 1988).

Unlike leguminous tree prunings and forest litter where nitrogen release is controlled by polyphenol and lignin contents respectively, the N release from crop residues is generally controlled by their C:N ratio because of low lignin and polyphenol contents (Palm, 1995). If crop residues of high C:N ratio (greater than 20) like maize stover are incorporated into the soil, there is nitrogen immobilization during the initial stages of decomposition. Though this immobilization seems undesirable, it has some beneficial aspects to nitrogen management. This is because the immobilized N is not lost

and may later be remineralized. This is from the evidence of Feller *et al.*, (1983) who reported that after adding ^{15}N -labelled maize stover to a sandy soil, 25% of the ^{15}N was found in the maize plant during first season with the remaining ^{15}N in the labile fraction of soil organic matter. The ^{15}N in organic matter was found to be released in subsequent seasons. Another beneficial effect of immobilization is reduction in mineral-N being lost by leaching, gaseous loss or erosion.

Data from Kisii district showed that complete removal of harvested plant parts and crop residues accounts for 55% of nitrogen depleted per ha per year (Smaling *et al.*, 1992). Sugarcane trash, materials being left behind after harvesting of sugarcane, have been shown to contain 100kgN/ha when still green (Wood, 1986). Trash retention has been shown to increase soil N within a single cropping cycle (Myers, 1987). Apart from providing N, it has been established that leaving sugarcane trash on the surface reduces direct sun strikes on the soil surface leading to reduced soil water evaporation and incidence of weeds (Clement, 1980).

2.5.2. Use of agroforestry for improving crop production.

Agroforestry has been defined as 'a system combining agricultural and tree crops of varying longevity (ranging from annual through biennial and perennial plants), arranged either temporarily (crop rotation) or spatially (intercropping) to maximize and sustain agricultural yield' (Lal, 1989a). Apart from the potential benefit of recycling nutrient elements from deep soils, prunings from the agroforestry woody legumes generally have a high N content because many of them fix nitrogen symbiotically with rhizobia

(Rubaduka *et al.*, 1993). Hence, crops may benefit from the N supplied when leaves are used as green manure or when composited and applied to soil. Palm (1995) reported that 4 t leaf biomass of *Leucaena leucocephala*, *Erythrina poeppigina*, *Inga indulis* and *Senna siamea* contain 154, 132, 142 and 105 kg N respectively. Thus addition of 4 tonnes per ha per year of any of these biomass to the soil will supply enough nitrogen to meet about 80kg N per ha per year requirement for the production of two tonnes grain of maize and three tonnes of associated stover.

Somewhat an encouraging example of the effect of alley cropping with *Leucaena leucocephala* has been available in Philippines (Lal, 1989b). The results obtained showed yield increases of 0.7 t ha⁻¹ when *Leucaena* was applied compared to only an increase of 0.3 t ha⁻¹ when N fertilizer was applied.

In another experiment conducted by Singh *et al.*, (1986) in India, it was observed that there was a significant decrease in yield whenever grain crops were grown in association with tree crops in regions prone to drought. The low grain yield was attributed to root competition for water.

Nevertheless, agroforestry tree pruning used as either mulch or incorporation as green manure minimizes rate of soil structure breakdown, water runoff, soil erosion and soil temperature increases. In addition, there is increased soil microbial activity and improved soil organic matter content (Insam *et al.*, 1991). Also agroforestry can be used for reclamation of severely degraded areas. For example near Mombasa, coral limestone left bare by quarrying has been restored, with humic to soil developing, through planting of *Casuarina equisetifolia* (Young, 1989). In sylvopastoral practices agroforestry trees act

as wind breaks and shelterbelts especially in semi-arid zones (Baumer, 1983). Consequently, agroforestry remains one of the most important low external soil conservation input available to resource poor farmers.

2.6 Use of industrial and town wastes

Use of cattle and other livestock manures as a source of plant nutrients has been found to be popular in the Kenyan highlands (Karanja, personal comm.). Addition of manure to the soil has been found to increase the labile pool of soil organic matter (Wander *et al.*, 1994). Influence of manure application on leaf water potential and yield was pronounced in loam and sand than in clay soil (Materechera and Mehuys., 1991). Okalebo *et al.*, (1993a) reported a significant grain yield increase in maize from 1189 to 2639 kg/ha in one instance due to manure application in Makueni and Machakos districts of Kenya. In spite of the above benefits, aerobically decomposed manures, commonly used by farmers, have low N fertilizer effect on the soil in the short term (Murwira and Kirchmann, 1993). This could be attributed to the fact that during aerobic decomposition, organic materials of high stability and low amounts of inorganic N are formed, reducing the value of the manure as an N fertilizer in the short term (Kirchmann, 1989). Also cattle manures have nutrient imbalance especially for phosphates and this requires supplementing manure treated soils with phosphates (Brady, 1990).

Town waste like sewage sludge and garbage are also potential organic resources that could be used by farmers living near towns. However, these resources have high concentration of heavy metals like lead, zinc and chromium (Wood, 1989). These heavy

metals are toxic to soil microorganisms as well as concentrating in the food chain. This limits the use of these residues as a source of plant nutrients. Nonetheless, these materials have been used in Netherlands by De Haan (1986) who used air dried sewage sludge in the period 1972-1983 on rye plot. He found that uptake of N from sludge by the crop was generally lower than from fertilizers. It was concluded that the low uptake of N was due to lower mobility of sludge NO_3^- in soils compared with fertilizer NO_3^- . This was attributed to high heavy metal content in the sludge e.g. manganese (Mn) and lead (Pb) that formed compounds with nitrates.

2.7 Alternative uses of organic residues

Organic residue management has very important socio-economic implications. In many countries, the soil benefits must be weighed against use of organic residues as livestock feed, fuel or construction material (Myers, 1987). For instance, a survey carried out by Karanja (Personal comm.) showed that in Kenya, apart from incorporation into the soil, large quantities of crop residue materials are usually fed to animals or sold. It was also found that, in areas with large farms like in Machakos and Thika districts, the use of litter materials for feeding livestock was minimum since animals graze freely. This indicated that in densely populated areas like Kiambu district, crop residues like maize stover, beans husks and banana leaves and stalks are fed to livestock then manures from these animals are applied to the land. The nutrient composition of this manure is variable depending on the livestock and manure management. But generally percent nutrients of Pen manure range from 1.03-2.12 for N, 0.49-1.30 P_2O_5 and 2.33-5.54 K_2O (Webster and Wilson, 1992).

Leguminous wood species of agroforestry trees and shrubs provide a high protein animal feed that complements low protein cellulytic materials like mature grasses and cereal straws (ILCA, 1985). Leaves of legumes like *Leucaena leucocephala* and *Gliricidia sepium* are the most nutritionally available plant parts of the materials consumed by animals. This is because they have high nitrogen content (around 3.2%) (Reynold and Adediran, 1988). Ironically, this is also the high quality plant part that should be incorporated into the soil to supply nitrogen. For instance Jabbar *et al.*, (1992), working in S.W. Nigeria found that use of fodder foliage of *L. leucocephala* and *G. sepium* as mulch gave higher maize yields than when twigs were included.

2.8 Incubation methods of determining soil available nitrogen.

Now that agricultural production of crops by small-holders is becoming more dependent on mineralization of N from plant residues, animal manure and soil organic matter, numerous biological and chemical methods attempting to provide a simple, reliable index of N availability have been proposed (Gianelle and Bremner, 1986; Stanford, 1982; Constantinides and Fownes, 1994a). It is generally accepted that the most reliable methods currently available are those involving determination of inorganic nitrogen produced by incubation of soil samples under aerobic and anaerobic conditions (Stanford, 1982). Various incubation methods have been used and these include incubating soils and organic residues *in situ* (use of column) and incubating the soil samples in polythene bags in the laboratory.

In situ field methods employing the incubation of undisturbed cores theoretically offers the best estimate of nitrogen mineralization (Raison *et al.*, 1987). This is because they exhibit transient flow of water and aeration in soil as in field situations. The field method is however prone to problems of compaction and water-logging (both leading to denitrification), especially in clayey soil (Anderson and Ingram, 1993).

Incubating soil samples in polythene bags in laboratories, though rather unnatural, has however been proved to give more approximate inorganic N available in a given season that has correlated well with crop performance (Stanford and Smith, 1972; Stanford, 1982). Recent research work of Tian *et al.*, (1993) has also shown that there is very high correlation between inorganic N released during incubation with field crop performance. In this method soil samples for incubation studies are weighed and placed in polythene bags. After moisture is adjusted to the required level, the bags are sealed and incubated. The polythene bags have an advantage in that they allow air exchange and minimize water loss. Gordon *et al.*, (1987) working with forest floor material in Ontario, Canada, showed that the thickness of polythene bags did not affect mineralization of nitrogen. In the same experiment, it was shown that all thicknesses remained impermeable to water loss. This ensured constant moisture content over the duration of the experiment. Since there was no leaching possibility in this experiment, all the available nitrogen was analyzed. This was the part of total mineralized nitrogen that exceeded the microbial demand. This method, adapted for analyzing the effect of organic materials to soil by Palm and Sanchez (1991), is easy to carry out hence it is used in this study.

CHAPTER THREE

3.0. MATERIALS AND METHODS

3.1. Site characterisation

The soils used in this study were collected from Malava smallholder farm in Kakamega district of Western Kenya. The soil was collected from this area because KARI had been carrying there a three year research project on fertilizer and manure use thus it was easy to locate a plot that had not received fertilizer or manure for the past three years. This was important to avoid residual nitrogen fertilizer effect in this experiment.

According to Jaetzold and Schimdt (1982), the site is situated in a high agricultural potential area which has an average annual precipitation of 65-80% of potential evapotranspiration. It falls under Agro-ecological zone LM 2 (Lower midland 2). The soil was collected from Chapman Keree's farm which is situated about 3 km from Malava trading centre, at an altitude of 1,595 m above sea level (a.s.l).

The rainfall distribution at the site is bimodal i.e it normally comes in two seasons: early March to July, referred to as the long rains (L.R) and beginning of September to November referred to as short rains (S.R). The land is used for growing maize, H622, during the L.R, sorghum and sunflower for S.R and sugarcane and pineapple the whole year.

The soils are well drained, very deep. dark reddish brown to dusty red, friable clays in places bouldery. The soils are classified as nito-rhodic Ferralsols according to Jaetzold and Schmidt (1982). The geology of this area is basic igneous rocks, mainly basalts.

3.1.1. Soil collection

Soil sampling was done during a dry period about two weeks prior to the beginning of long rains (5/3/95). Soil samples were taken from the surface layer (0-20 cm) using a spade on the plot that had not received any fertilizer or manure for the past three years. A random procedure was utilized in taking the samples in order to cover the field. The plot was divided into 10m by 10m subplots. From each subplot, 15 soil samples were taken randomly. The samples were bulked and about 200 kg soil brought to the laboratory. The soil was air-dried in the laboratory and about 2 kg ground and sieved through 2 mm screen to be used for physical and chemical analysis while samples for total nitrogen and total organic carbon determination were ground further to pass through a 60 mesh sieve (0.25 mm). The rest of the soil sample was kept in polythene bags for use in incubation and greenhouse experiments.

3.1.2. Soil particle size analysis

Particle size distribution was determined using the Bouyoucos hydrometer method described by Okalebo *et al.*, (1993b). Thus fifty grammes of air dried soil (2mm) which had been ground was transferred to a shaking bottle and 250 ml of distilled water added together with 10 ml of 10 % Calgon solution and the bottle corked tightly. 10% calgon was added as a dispersing agent (i.e. to separate the soil mass into its primary particles). The soil samples, together with distilled water and calgon, were shaken in the air-tight bottles overnight. The suspension was transferred to a graduated cylinder, hydrometer inserted and then distilled water added to 1130 ml mark and the hydrometer removed.

After mixing the sample in the cylinder vigorously with a plunger, the hydrometer was inserted and the first hydrometer reading taken 40 seconds later. The second hydrometer reading was taken 2 hours later. Before each hydrometer reading, the temperature of the soil suspension was recorded. It was important to know and make correction for the temperatures of the suspension because greater temperatures result in reduced velocity due to liquid expansion leading to a more rapid descent of the falling particles (Okalebo *et al.*, 1993b). The % sand, silt and clay was calculated as described by Okalebo *et al.*, (1993b).

3.1.3. Determination of soil pH

The soil reaction was determined following the procedure outlined by Okalebo *et al.*, (1993b). Ten grammes of air dried soil (2 mm) was weighed into plastic specimen bottles and 25 ml of deionized water added. The mixture was stirred for 10 minutes with a glass rod and allowed to stand for 30 minutes. It was stirred again for 2 minutes and the pH of the soil solution measured by inserting electrode of a pH meter into the suspension. The pH was then recorded as soil "pH in water" pH (H₂O), 1:2.5 ratio.

3.1.4 Moisture determination for data correction

The soil under plant material moisture was determined following the procedure describe by Anderson and Ingram (1993). Sample material weighing about 5gm for soil samples and 1gm of for plant material were put into dry containers of known weight (W1) and the total weight (W2) recorded. The materials were dried at 105 °C over night

in case of soil and 65°C for 2 hours if plant material. The materials are then allowed to cool in a desiccator and reweighed giving weight (W3).

Calculation

$$\% \text{ Dry material} = \frac{(W3-W1) \times 100}{(W2-W1)}$$

Corrected data to dry weight basis was obtained by multiplying the uncorrected data by (100/% dry material).

3.1.5 Determination of gravimetric soil water content at field capacity

The gravimetric soil water content at field capacity was determined according to the method described by Anderson and Ingram (1993). A 5 litre beaker was filled with soil and a shallow hole made at the centre of the soil. The hole was filled with water and refilled as necessary until about 50 % of the soil was water soaked. The beaker was covered with parafilm for 3 days to prevent evaporation of water from the soil. After 3 days, 5 replicated soil samples from near the centre area were taken and bulked. Then 250g of the bulked wet soil was put in a container of known weight (W1), weighed to give wet weight (W2) and then dried at 105°C for 48 hours. The dry soil and the container were reweighed to give dry weight (W3).

Calculation

Gravimetric soil water content at field capacity (%)

$$= \{(W2-W3)/(W3-W1)\} \times 100 .$$

3.1.6. Total organic carbon

The Nelson Sommers procedure outlined in Okalebo et al (1993b) was used to estimate total organic carbon in the soil. In this method organic carbon is oxidized by concentrated sulphuric and aqueous potassium dichromat ($K_2Cr_2O_7$) mixture. After complete oxidation from the heat of solution and external heating, the unused $K_2Cr_2O_7$ (in the oxidation) is titrated against ferrous ammonium sulphate. The used $K_2Cr_2O_7$, obtained from the difference between added and residual $K_2Cr_2O_7$, gives a measure of organic carbon content in either soil or plant material (Nelson and Sommers, 1975).

Dry soil (passed through a 0.25 mm sieve) weighing 0.3 g was put into digestion tubes. Five millilitres of 1 N potassium dichromat (1N $K_2Cr_2O_7$) and 7.5 ml concentrated sulphuric acid were added. The tubes were placed in a pre-heated block at 150°C for exactly 30 minutes, removed and allowed to cool. The digests were quantitatively transferred to 100 ml conical flasks and 0.3 ml of 1,10 phenanthroline monohydrate-ferrous sulphate indicator added. Using a magnetic stirrer, to ensure proper mixing, each digest and blanks were titrated with 0.2 M ferrous ammonium sulphate solution to end point (colour changed from green to brown). The % organic carbon in the soil was calculated as described by Okalebo *et al.*, (1993b).

3.1.7. Exchangeable bases

Exchangeable bases in the soil were determined using 1M NH_4OAc (ammonium acetate) as an extractant (Okalebo *et al.*, 1993b). Five grams of air dry soil (< 2mm) was weighed into clean plastic bottles. One hundred millilitres of 1M NH_4OAc adjusted

to pH 7.0 was added and the bottles stoppered. The contents were shaken for 30 minutes and filtered through No.542 Whatman papers. The filtrate was used for Na, K, Ca. and Mg determinations. Amounts of sodium and potassium in the extracts were determined by flame photometry while calcium and magnesium contents were analyzed by atomic absorption spectrophotometry. In Ca analysis, 26.8 % Lanthanum chloride ($\text{LaCl}_3 \cdot 7\text{H}_2\text{O}$) was added as a releasing agent to prevent formation of refractory compounds, which could have interfered with the determination.

3.1.8. Available phosphorus

Available phosphorus in the soil was determined by the Bray No. 2 method (Bray and Kurtz, 1945) as described by Okalebo *et al.*, (1993b). This method was selected because the soil reaction determination had indicated that the soil was acidic. Hence the combination of HCl and NH_4F in the Bray P2 solution is designed to remove easily acid-soluble forms of P, largely the Ca phosphates and a portion of Al and Fe phosphates which could not be extracted by the Olsen method (page, 1982).

Air dry soil (< 2 mm) weighing 2.5 g was placed into clean plastic bottles. Fifty millilitres of prepared Bray P2 extracting solution was added, the contents shaken for 5 minutes and filtered through No. 542 Whatman paper. Ten millilitres of each soil extract and prepared P standard solutions were pipetted into 50 ml volumetric flasks. Twenty millilitres of distilled water and 5 ml of 0.8M H_3BO_3 were added. Finally 10 ml of freshly prepared ascorbic acid was added and each flask topped with distilled water. The flasks were left to stand for 1 hour for full colour development. The available phosphorus

was estimated colorimetrically using a CE 202 UV/visible spectrophotometer, model Cecil 202, Cambridge U.K.

3.1.9. Total nitrogen

Total nitrogen was determined by the semi- micro Kjeldahl method as described by Okalebo *et al.*, (1993b). The organic N in the soil sample under analysis was converted to ammonium nitrogen ($\text{NH}_4^+ - \text{N}$) by digestion under heat using concentrated H_2SO_4 and 30% hydrogen peroxide (H_2O_2). Selenium was used as a catalyst while lithium sulphate ($\text{Li}_2\text{SO}_4 \cdot \text{H}_2\text{O}$) was added to raise the boiling point of the mixture. After the completion of digestion, the soil digest solutions were diluted with water and then treated with aqueous sodium hydroxide (NaOH) and distilled with steam. The ammonia (NH_3) liberated by distillation was collected in dilute boric acid (H_3BO_3)-indicator. The amount of nitrogen in the soil samples was estimated by titrating the distilled ammonia with dilute HCl as described by Okalebo *et al.*, (1993b).

3.2 Characterization of plant materials

3.2.1 Plant material sampling and preparation for analysis

On-farm and industrial plant materials were collected from Kabete Campus (University of Nairobi), Muguga area (Kiambu District) and Bungoma district and Maseno area in Western Kenya. Leaves (blades and petioles) from several indigenous woody trees and herbaceous agro-forestry trees, crop residues and agro-industrial plant

wastes for example barley, coffee, and rice husks and bean and sugarcane trash, as listed in Table 4, were sampled. The green leaf samples were obtained by selecting 5 fully matured trees randomly selected on farms that had been randomly picked in the areas mentioned above. Three branches were selected from each tree: one at the bottom most, second at the middle and the third at the topmost of the canopy. The leaves were removed completely from each branch and the sub-samples mixed to make a composite sample. Following the procedure described by Anderson and Ingram (1993), 5 kg fresh sample was taken for drying in the laboratory. For industrial wastes, 10 subsamples from one week old industrial heaps were taken randomly, mixed to make a composite sample and then 2 Kg taken for use in the laboratory through the quartering procedure.

The samples were held in labelled paper bags No. 25 and transported to the laboratory. The samples in bags were dried in a forced-air oven at a maximum temperature of 30°C to avoid loss of soluble polyphenols (Constantinides and Fownes, 1994a). After drying, the samples were ground by electric plant grinder to pass through a 2 mm size sieve and kept in plastic containers for analysis of nutrient contents (N,P,K,Mg,Ca,lignin and polyphenol) as outlined in the following sections.

3.2.2. Tissue analyses for total N , P , Ca , Mg , K

In order to analyze for total N, P, Ca, Mg, K in the plant tissues, the samples were re-dried at 65°C so as to express the nutrients on dry matter basis as recommended by Okalebo *et al.*, (1993b). The wet digestion procedure was then used to bring the nutrients into solution using the procedure described by Okalebo *et al.*, (1993b). This

procedure was selected because it is fast, accurate and only a single digestion is required to bring all the above nutrients into solution. The re-dried samples weighing 0.3 g were put into labelled, dry and clean digestion tubes to which 4.4 ml of digestion mixture containing sulphuric acid, selenium powder, lithium sulphate, and hydrogen peroxide (H_2SO_4 -Se- Li_2SO_4 - H_2O_2) was added. Included in the digestion were two standard samples and two blanks to check and correct the results for any errors during analysis and impurities. The samples and blanks were placed in the Fisons LE11 digestion block and digested at 360°C, starting from 100°C and raising the temperature gradually to 360°C to avoid frothing, till the solution was colourless. The digests were allowed to cool after which about 25 ml distilled water was added and mixed well until no more sediments dissolved. The digests were transferred into labelled 50 ml flasks, topped and the contents thoroughly mixed. The sample solutions in these flasks were used for total N, P, K, Ca and Mg determinations.

3.2.2.1 Total nitrogen analysis

Total nitrogen was estimated using semi-micro Kjeldahl method as described by Okalebo *et al.*, (1993b) where 5 ml of the sample solution was transferred to the reaction chamber of the Markham still and 10 ml of 40% NaOH added. The contents in the chamber were steam distilled immediately into 5 ml of 1 % boric acid containing 4 drops of a mixed indicator. The distillation was continued for 2 minutes from the time the indicator turned green. The distillate was removed and titrated with N/70 HCl to the end point and the volume (ml) of the standard HCl required noted. The blank digest was

distilled and titrated as the sample. The volume (ml) of N/70 HCl required for the blank was subtracted from the micro burette reading of the sample giving a corrected volume of N/70 HCl (V).

Calculation

$$\% \text{ N in plant sample} = \frac{V \times 0.2}{\text{weight of sample}}$$

Where the weight of sample is 0.3g

3.2.2.2. Determination of total phosphorus content.

Determination of phosphorus in the plant residue samples was done by the Murphy-Riley ascorbic acid method with no pH adjustment procedure as outlined by Okalebo *et al.*, (1993b). Ten millilitres of the clear digest solution were pipetted into 50 ml flasks. Twenty millilitres of distilled water and 10 ml of ascorbic acid reducing agent were added to each flask of sample and standards. The flasks were topped with distilled water, shaken well and left to stand for 1 hour to permit full colour development. The standards and sample absorbances were measured at 880 nm wavelength setting in a CE 202 UV/visible spectrophotometer, model Cecil 202, Cambridge, U.K.

Calculations :

A plot of absorbance against standard concentration was done from which the solution concentration for each unknown and the two blanks were determined. A

subtraction of the mean blank concentration from the unknown gave the corrected concentration (C). The P content of the plant residue samples was calculated as follows:

$$\% \text{ P in plant residue sample} = \frac{C \times 0.025}{\text{weight of sample}}$$

Where the weight of sample is 0.3g.

3.2.2.3. Determination of potassium and magnesium.

One millilitres of the wet-digested sample solution (obtained from section 3.2.2) was pipetted into 50 ml volumetric flasks and made to the mark. The potassium in the samples was determined by flame photometer while magnesium content was estimated, from the same sample solution, by atomic absorption spectrophotometer Pye Unicam SP 919 model. The calculation of the two nutrients was as follows :

$$\% \text{ K or \% Mg in sample} = \frac{C \times 0.025}{\text{weight of sample}}$$

C is the amount of K or Mg present in the unknown solution obtained from the calibration curve prepared by plotting transimissions against K or Mg standard concentrations.

3.2.2.4. Determination of calcium content

Determination of calcium content in the plant material was done using atomic absorption spectrophotometer. Ten millilitres of the sample solution was pipetted into 50

ml volumetric flasks. Ten millilitres of 0.15% Lanthanum chloride were added to the flask to check phosphate and sulphate interference during determination. One millilitre of dilute ammonia was added to suppress interference from mutual excitation between elements. The contents in the flask were made to the mark with distilled water and then well shaken. The standard blanks and sample solutions were then sprayed into atomic absorption spectrophotometer flame with a Ca lamp. A graph of transmission versus Calcium concentrations from the standard solutions was obtained on the computer whereby the Ca concentration (C) of the unknown sample solutions were read.

$$\% \text{ Ca in the sample} = \frac{C \times 0.025}{\text{weight of sample}}$$

Where the weight of sample is 0.3g

3.2.3 Total carbon determination in plant materials

Total Carbon in the plant materials was determined by Nelson and Sommers method as described for soil samples (see section 3.1.5) with the exception that smaller samples (0.02 g) were used for the plants materials to avoid frothing.

3.2.4. Determination of lignin content

The lignin content in the plant residues was determined by the acid detergent fibre method (Van Soest, 1963). This method was chosen because the permanganate-acid detergent fibre (ADF) treatment of Goering and Van soest (1970) fails to give full lignin recovery for samples with ADF > 30%, especially for litters (Rowland and Robert, 1994). Oven-dried (30°C) materials weighing 0.500 g (W1) were put into 500 ml

beakers. One hundred millilitres of cetyltrimethylammonium bromide (CTAB) solution was added to each beaker and a few drops of Octan-2-ol anti-foam agent. The contents were connected to a condenser and refluxed for 1 hour. The refluxed contents were filtered hot through pre-weighed (W2) sinters (No.2) under gentle suction. The residues were washed with 3 x 50 ml aliquots of boiling water followed by acetone and filtered dry. The residues on the sinters were oven dried at 105°C for 2 hours, cooled in a desiccator and weighed (W3). This was used to calculate % ADF.

The sinters with dried residues were then half filled with cooled 72% H₂SO₄, stirred with glass rod and placed on small beakers to catch the acid as it drains through from the sinters. The sinters were refilled with the drained acid and stirred, as acid drains away. After 3 hours, the acid was filtered off under vacuum and contents washed with hot water until free of acid. The sinters with washed residues were oven dried at 105°C for 2 hours, cooled and weighed to give weight (W4). The dry residue in the sinters were then ignited at 550°C for 2 hours, cooled in a desiccator and re-weighed to give weight (W5). Lignin was calculated as follows:

Calculations

$$\% \text{ Lignin} = \frac{(W4 - W5)}{W1} \times 100$$

Where W1 was the corrected weight of the sample on dry weight basis after determination of residual moisture in the samples by oven-drying them at 105°C overnight. W4 and W5 are the weights described in the text under this section.

3.2.5. Extractable polyphenols

Soluble polyphenols were determined by the procedure similar to that described by Anderson and Ingram (1993) with the following slight modifications by Constantinides and Fownes(1994b):

(i) A plant tissue-to-solvent ratio of 5mg/ml was used instead of 37.5 mg/ml. This was because the higher tissue-to-solvent ratio may not extract all polyphenols.

(ii) Folin-Ciocalteu reagent was used instead of Folin-Denis reagent because the former is more sensitive to reduction by polyphenolics and less prone to precipitation than the latter.

Plant materials weighing 0.1 g were put into 50ml beakers and 20 ml of 50% methanol added. The beakers were covered with paraffilm paper and placed in a pre-heated water bath at 77-80°C for 1 hour. The extracts were filtered quantitatively through Whatman No.1 filter papers into 50 ml volumetric flasks using 3 ml 50% methanol to rinse, made upto the mark with distilled water and well shaken. One millilitre of the unknown or the standards was pipetted into 50 ml volumetric flasks, 20 ml water added followed by addition of 2.5 ml Folin-Ciocalteu reagent and 10ml sodium carbonate (17%). The flasks were made to the mark with distilled water mixed well and left to stand for 30 minutes. The standards and unknown absorbances were colorimetrically read at 760 nm wavelength using a CE 202 UV/visible spectrometer model Cecil 202. Cambridge U.K.

Calculations

A regression of absorbance against standard concentrations was calculated and solution concentrations for each unknown and blanks determined. Subtracting the mean blank value from unknowns gave a value for corrected concentration, C.

$$\begin{aligned} \text{Total extractable polyphenols (\%)} \\ = \frac{C \times 5}{w} \end{aligned}$$

where w is the corrected sample weight on oven dry weight basis.

3.3. Incubation experiments

3.3.1. Procedure for incubating plant materials

The incubation procedure was adapted from Palm and Sanchez (1991). Six plant residues of varying qualities were selected following the chemical characterization data (Table 5). The materials are: *Croton macrostachyus*, *Leucaena leucocephala*, *Calliandra calothyrsus*, *Tithonia diversifolia*, *Sorghum bicolor* and the rice (*Oryza sativa*) husks. The selection was based on contrasting nitrogen, lignin and soluble polyphenol contents (Table 3). These qualities were considered from the work of Palm (1995)

Table 3: Qualities of various plant materials used for incubation

Treatment	Name of plant	Quality
1	<i>Croton macrostachyus</i> (Cm)	High N; Low Lignin; Low Polyphenol
2	<i>Leucaena leucocephala</i> (Ll)	High N; Medium Lignin; High Polyphenol
3	<i>Calliandra calothyrsus</i> (Cc)	High N; High Lignin; High Polyphenol
4	<i>Tithonia diversifolia</i> (Td)	High N; Medium Lignin; Medium Polyphenol
5	<i>Sorghum bicolor</i> (Sb)	Low N; Low Lignin; Medium Polyphenol
6	<i>Oryza sativa</i> husks (Os)	Low N; High Lignin; Low Polyphenol
7	Control	

3.3.2. Experiment 1: Nitrogen mineralization from different materials.

For each of the six plant materials: 1.14 g of the material was mixed with 500g (dry weight basis) soil, which had been air dried and passed through 2mm sieve. This was to approximate a rate of 5 tonnes dried plant materials ha⁻¹. This is because a rate of 3 tonnes ha⁻¹ of plant material was found not to respond well by Palm and Sanchez (1991). After thorough mixing of the plant residues and the soil, the mixtures were placed in 500 gauge polythene bags. A soil only treatment was included as a control. Deionized water was added to each mixture to reach gravimetric soil water content (25% soil moisture at field capacity) as was determined in section 3.1.4. This water was used to avoid water logging that could have lead to anaerobic conditions which would have

limited mineralization in this experiment Anderson and Ingram (1993). The soil and plant mixtures in polythene bags were tightly tied with strings and stored in a dark place in the laboratory at room temperature of approximately 21°C. Each treatment was replicated 3 times and laid down in a completely randomized design. Moisture content of the incubated soils was maintained by periodic weighing of the samples and moisture loss made by addition of deionized water. This was accomplished by weekly weighing of the samples and the difference in weight from the initial sample weight made up by addition of equivalent weight of deionized water.

The experiment was run for twelve weeks with sampling and analysis being done after every two weeks as describe in sections 3.3.5- 3.3.7.

3.3.3. Experiment 2: Nitrogen release from organic mixtures.

C. macrostachyus (Cm) and *O. sativa* (Os) were mixed in various ratios on weight basis of the ground materials to give the following ratios:-

- (i) 1/2 Cm : 1/2 Os
- (ii) 3/4 Cm : 1/4 Os
- (iii) 1/4 Cm : 3/4 Os
- (iv) 1 Cm: 0 Os
- (v) 0 Cm: 1 Os
- (vi) Soil (control)

These treatments were also incubated following the procedure described in section 3.3.2

3.3.4. Experimental design and layout

The two experiments (1) and (2) above were laid down in a completely randomized design with 7 and 6 treatments respectively. Each treatment was replicated three times.

3.3.5. Sampling for NO_3^- - N and NH_4^+ - N determination

Mineralization and nitrification processes were assessed through the changes of ammonium nitrogen (NH_4^+ -N) and nitrate nitrogen (NO_3^- -N) contents in the soil. The ions were measured immediately after the addition of the plant residues and at 2, 4, 6, 8, 10 and 12 weeks after incubation. Moisture content was also being estimated at each sampling period as shown in section 3.1.4. Subsamples weighing 10g were taken from each bag for mineral nitrogen extraction. The subsamples were extracted by shaking in 50ml of 0.5 M K_2SO_4 for 30 minutes then filtering through Whatman No. 542 paper and the filtrate used for the analysis of NH_4^+ -N and NO_3^- -N.

3.3.6. Nitrate nitrogen (NO_3^- -N) analysis

The NO_3^- -N in the samples was determined colorimetrically following the salicylic acid procedure outlined by Anderson and Ingram (1993). Each prepared (as shown in section 3.3.5.) standard and sample solution amounting to 0.5 ml was pipetted into test tubes. One millilitre of 5% salicylic acid solution was added to each tube and mixed thoroughly using a vortex mixer then left to stand for one hour for complete cooling. Ten millilitres of 4 M NaOH solution was then added to each test tube, mixed well and left to stand for 1 hour for full colour development. The standards and sample absorbances were read at 410 nm using a CE 202 UV/visible spectrophotometer model Cecil 202, Cambridge U.K.

3.3.7. Ammonium nitrogen (NH_4^+ - N) analysis

Analysis of NH_4^+ - N from the extracted samples was done colorimetrically according to the method described by Anderson and Ingram (1993). Using a micro pipette, 0.10 ml of each standard and sample solution were transferred into suitably marked test tubes. Five millilitres of a reagent obtained by mixing sodium salicylate, sodium citrate, sodium tartrate and sodium nitroprusside solution (N_1) was added to each test tube, mixed well and left to stand for 15 minutes, then 5 ml of a reagent containing NaOH and sodium hypochlorite solution (N_2) was added to each test tube, mixed well and left to stand for 1 hour. Standard and sample absorbances were read at 655 nm wavelength on a spectrophotometer.

Calculations

A regression of absorbances against standard concentrations for either $\text{NH}_4^+\text{-N}$ or $\text{NO}_3^-\text{-N}$ was done from which concentrations ($\mu\text{g/ml}$) for each unknown and blanks were determined. A subtraction of the mean blank value from the unknowns was done to give corrected concentrations of the samples, C.

$$\text{NO}_3^-\text{-N or NH}_4^+\text{-N } (\mu\text{g/ml}) = \frac{C \times V}{w}$$

where C = corrected concentrations ($\mu\text{g/ml}$)

V = extracting volume (ml)

W = corrected weight (dry weight basis) of the sample(g).

3.4. Experiment 3: N uptake by *Zea mays* (maize).

The glasshouse experiment was set up to follow the response of maize to organic materials of *C. macrostachyus*, *O. sativa*, *T. diversifolia*, a mixture of 1/2 Cm and 1/2 Os, and soil only as a control. Standard plastic pots with a base diameter of 8" with holes at the bottom were underlined with double layer of Whatman No 5 filter papers to hold the soil. To each pot 2.5 kg of soil (oven dry weight basis) was added and then pots were placed on plates. At the beginning of the experiment each pot received a basal application of 114 mg K as K_2SO_4 and 114 mg P in the form of $\text{Ca}(\text{HPO}_4)_2$ to supply equivalent of 100 kg/ha K and P respectively. Three plant materials and one mixture were used in the green house because: Incubation experiment had shown that *C. macrostachyus* released the highest amount of N, *O. sativa* immobilized N while *T. diversifolia* and the 50% Cm & 50% Os mixture released intermediate amounts of N throughout the sampling

period. Based on this fact, the three materials were picked to find out if their contrasting N release patterns could be reflected in maize growth in the pots.

To each pot, containing 2.5 kg soil, 5.7 g (equivalent to 5 tonnes dry plant materials ha^{-1}) of ground (< 2mm) plant residues were added and mixed thoroughly. Two maize seeds, variety H 512 were sown in each pot to a depth of 2.5 cm. The soil moisture was adjusted to gravimetric soil water content (i.e 25% soil moisture at field capacity) as was determined in section 3.1.4. Two weeks after planting (2 WAP), the maize seedlings were thinned to one plant per pot. During the growth period, the pots were maintained in the glasshouse and rotated periodically. Depending on sunshine intensity and the prevailing ambient temperature, soil water content was adjusted, to replace the loss, three times a week. Each treatment was replicated 9 times to allow for three harvests during the experimental period.

For assessment of maize dry matter yield and nutrient uptake, maize plants were sampled at 3, 5 and 7 WAP. Three pots per treatment were randomly selected and maize plants excised at soil level, placed in standard labelled paper bags No.2 and dried at 65° C for 48 hours then weighed for dry matter (DM) measurements. The dry shoots were then ground to pass 60 mesh sieve for nutrient (N, P, K, Ca and Mg) analysis. The procedures for nutrient analysis were those described by Okalebo *et al.*, (1993b) as was explained above in plant characterization section (3.2.2)

The dynamics of the mineral N (NO_3^- -N and NH_4^+ -N) concentration in remaining soil in pots were monitored by sampling soil samples from each pot, where the plants had been harvested and all the roots removed from the soil, These soil samples were extracted

in 50 ml 0.5M K₂SO₄ for 30 minutes. The NO₃⁻-N and NH₄⁺-N in the extracts were analyzed as described in incubation experiment above (section 3.3).

3.5. Statistical analyses.

Data on N release was analyzed by standard ANOVA procedure for completely randomized experimental design. Main effect means were separated by LSD at the P=0.05 level. Correlation analysis between chemical contents of plant tissues was done to assess the underlying generalities on their chemical characteristics. Contrasts were used to test the significance of differences in response parameters due to mixing high quality and low quality plant residues in various ratios. For contrast analysis, the amount of N-released during incubation of mixtures was compared to the predicted N-released obtained from the means N-released by *C. macristachyus* and *O. sativa* (Os) individually i.e (amount of N by Cm + amount of N by Os)/2. Treatment differences in the glasshouse experiment were analyzed with general linear model procedure (GLM) (SAS, 1985). Under GLM, the sources of variation in the experiments included residue chemical contents and sampling periods. A t-test was employed to determine the effect of site of sampling on quality of the plant materials.

CHAPTER FOUR

4.0 RESULTS AND DISCUSSION

4.1. Chemical and physical characteristics of soil and plant residues.

The chemical and physical characteristics of the soil from Malava (Kakamega district) are given in Table 4. The soil is strongly acidic, moderate in sodium, high in potassium and magnesium but low in calcium based on the gradings of Okalebo *et al.*, (1993b). It was also moderate in nitrogen and total carbon contents. The soil textural class was found to be sandy clay.

The soils of this area are developed from basic parent materials, mainly basalts (Jaetzold and schmidt, 1982). Thus, it was expected that the soil pH(H₂O) would be higher than 5.2 (Deckers, 1993). The strong acidity of the soil in this area could attributed to leaching of bases from the top soil to deeper horizons as the area is a high rainfall zone. These soils, being Ferralsols, are highly weathered with soil materials consisting of kaolinite, quartz and hydrated oxides (Ssali *et al.*, 1986). Thus, the capacity of these soils to supply nutrients to the plants as well as their capacity to retain nutrients are both low. From a soil fertility point of view, the low nutrient retention capacity has marked consequences for fertilizer management especially nitrogen which is normally leached before plants utilize it (Deckers, 1993).

Table 4: Physical and chemical characteristics of soil from Malava, Kakamega district.

Parameters of surface soil(0-20 cm)	Values
pH (H ₂ O)	4.49
Organic Carbon (%)	1.78
Total Nitrogen (%)	0.16
Bray P (ppm)	17.14
Exchangeable Cations (me/100g soil)	
Calcium (Ca ²⁺)	0.14
Magnesium (Mg ²⁺)	0.31
Sodium (Na ⁺)	0.07
Potassium (K ⁺)	0.26
Texture (%)	
Sand	49.5
Clay	48.4
Silt	2.1
Textural class	Sandy clay

The chemical characterisation data obtained from plant materials collected from Kabete Campus (University of Nairobi), Muguga area (Kiambu district) and Bungoma district and Maseno area in western Kenya are shown in Table 5. Large differences in the chemical composition of the 33 plant residues were observed. Nitrogen content ranged from 0.47% in sugarcane trash to 3.74% in *L. leucocephala* leaves. Phosphorus content found in these materials ranged from 0.06% in *H. saligna* leaves to 0.36% in sweet potato vines.

Table 5: Chemical characteristics of plant materials

Plant Name	Chemical characteristics (%)							
	Site	N	Ca	Mg	P	K	Poly	Lignin
Banana leaves	A	1.30	1.02	0.27	0.10	1.72	1.78	11.99
	C	2.50	1.30	0.36	0.13	2.66	0.49	9.51
Barley husks	A	2.97	0.07	0.12	0.32	1.97	1.36	1.92
Coffee husks	A	1.63	0.51	0.12	0.14	4.45	1.37	27.61
<i>Ficus sp</i> (Mugumo leaves)	A	1.97	1.30	0.28	0.24	3.29	-0.11	43.46
	B	1.37	1.28	0.12	0.27	2.87	11.86	7.46
<i>Croton megalocarpus</i> leaves	A	3.30	1.06	0.45	0.29	4.03	1.37	8.40
	B	2.53	1.50	0.74	0.25	3.12	2.19	15.57
	C	2.60	1.26	0.45	0.18	1.48	6.25	16.54
<i>Croton macrostachyus</i> leaves	A	3.30	1.06	0.45	0.29	4.03	1.37	8.40
	B	2.77	1.19	0.35	0.31	3.87	1.25	10.92
	C	3.30	1.47	0.41	0.27	3.42	4.41	13.87
Bamboo leaves	A	1.80	0.45	0.19	0.18	2.21	0.56	7.74
	B	1.37	0.29	0.16	0.12	1.72	0.57	8.471

Plant Name	Chemical characteristics (%)							
	Site	N	Ca	Mg	P	K	Poly	Lignin
<i>Eucalyptus sp.</i> leaves	A	1.73	1.04	0.41	0.12	0.72	2.08	34.40
	B	1.13	1.10	0.23	0.07	0.89	5.59	30.13
<i>Markhamia lutea</i> leaves	A	1.87	0.94	0.18	0.21	2.21	1.99	28.14
	B	1.97	1.07	0.25	0.16	1.55	4.24	25.24
	C	2.47	1.51	0.27	0.17	1.23	4.56	18.79
Coffee leaves litter	A	2.30	1.00	0.31	0.18	3.45	5.06	30.29
	C	2.80	1.17	0.28	0.19	3.26	5.10	13.65
	C	2.03	1.11	0.22	0.18	2.75	3.94	16.88
<i>Spathodea canipulata</i>	A	1.63	1.67	0.37	0.18	1.88	3.09	31.34
	B	2.27	1.54	0.33	0.24	1.88	4.72	27.52
	C	2.03	3.41	0.37	0.15	1.23	5.57	14.64
Sweet potato vines	A	1.73	0.84	0.46	0.48	6.63	2.38	13.35
	C	2.90	1.12	0.33	0.24	2.75	5.80	5.93
<i>Leucaena leucocephala</i>	B	3.74	1.16	0.33	0.26	3.37	6.13	14.68

Plant Name	Chemical characteristics(%)							
	Site	N	Ca	Mg	P	K	Poly	Lignin
<i>Calliandra calothyrsus</i>	B	3.64	0.66	0.24	0.22	1.88	5.36	28.71
	H	2.77	0.58	0.15	0.17	0.97	15.24	14.18
Neem leaves	B	2.97	2.57	0.18	0.20	1.90	2.75	9.01
<i>Hakea saligna</i> leaves	B	1.00	0.47	0.13	0.06	0.64	6.19	32.89
Napier grass	B	1.97	0.31	0.12	0.14	3.85	0.96	4.82
<i>Lantana camara</i> leaves	D	2.50	0.87	0.51	0.27	1.80	9.42	17.13
	E	2.40	1.03	0.49	0.25	2.05	6.95	25.53
	F	2.60	0.86	0.44	0.25	2.38	8.24	15.94
	G	2.53	1.06	0.53	0.25	2.38	8.42	16.03
<i>Tithonia diversifolia</i> leaves	D	3.17	1.23	0.58	0.30	4.28	3.61	13.76
	E	3.27	1.14	0.44	0.32	4.61	2.92	14.05
	F	3.44	1.50	0.37	0.26	4.69	3.74	15.25
	G	3.97	1.53	0.39	0.31	4.44	3.39	13.68
<i>Psidium guajava</i> leaves	D	2.17	0.88	0.31	0.22	1.57	14.03	19.01
	E	1.97	1.05	0.36	0.18	1.31	13.80	17.27
	F	2.03	1.03	0.31	0.19	1.48	14.02	18.11
	G	2.13	0.79	0.31	0.21	1.82	13.60	21.71
Creeping vines	D	3.50	0.78	0.48	0.25	2.41	2.65	4.95
	G	3.17	0.85	0.48	0.24	2.58	2.64	5.91

Plant Name	Chemical characteristics(%)							
	Site	N	Ca	Mg	P	K	Poly	Lignin
<i>Acanthus sp</i> leaves	D	3.13	1.17	0.51	0.27	2.58	3.64	6.96
	E	3.44	0.94	0.54	0.25	3.09	2.36	9.90
	F	3.20	1.17	1.73	0.28	4.94	4.16	9.59
	G	3.23	1.16	0.54	0.27	3.09	3.38	7.36
<i>Grevillea robusta</i> leaves	H	1.37	1.22	0.09	0.06	0.64	5.63	23.72
<i>Sorghum bicolor</i> leaves	C	0.63	0.49	0.14	0.10	1.40	2.91	4.23
<i>Oryza sativa</i> husks	C	0.63	0.08	0.04	0.17	0.38	0.00	16.66
Bean trash	C	0.80	0.89	0.37	0.07	1.57	0.23	11.77
	I	0.77	0.73	0.20	0.06	1.31	0.29	11.29
Tea leaves	C	2.40	1.03	0.33	0.31	1.57	12.21	31.62
Tea litter	C	1.37	1.18	0.27	0.30	1.23	7.32	37.04
Sunflower leaves	C	2.03	2.36	0.20	0.17	3.76	3.66	15.47
Banana stalks	C	0.73	0.39	0.20	0.18	4.10	0.00	5.49
Sugarcane trash	C	0.47	0.24	0.06	0.06	1.23	0.34	1.00
Russian comfrey	I	2.97	1.84	0.57	0.23	5.37	4.45	1.76

A=Kabete (Nairobi-Central Kenya); B= Muguga (Kiambu-Central Kenya); C= Bungoma (Western Kenya); D= Ochinga farm (Western Kenya); E= Musinde farm (Western Kenya); F=Julius farm (Western Kenya); G=Abneri farm (Western Kenya); H= Maseno ICRAF (Western Kenya) I= Gatwanyaga (Thika-Central Kenya)

It is interesting to note that there were two exotic and one indigenous plant species whose leaves had both high N and P; These were the indigenous, *C. macrostachyus* containing 3.12% N and 0.29% P, and the exotics, *T. diversifolia* with 3.46% N and 0.29% P, and *Acanthus sp.* with 3.25% N and 0.27% P content. The three species could be potential organic inputs that can be pruned and incorporated into soil to supply N and P to the crops.

Potassium content in the plant residues ranged from 0.38% in *O. sativa* husks to 4.51% in *T. diversifolia* leaves. The leaves obtained from *Eucalyptus sp.*, *H. saligna* and *G. robusta* contained lower K levels as compared to other species. Calcium levels of residues ranged from 0.07% in barley husks to 2.57% in Neem tree leaves. Magnesium contents ranged from 0.04% in *O. sativa* husks to 0.58% in leaves of *Acanthus sp.* Lignin contents of the plant materials ranged from 1.92% (barley husks) to 37% (tea litter). The banana stalks and *O. sativa* husks did not have any soluble polyphenol whereas *P. guajava* leaves contained 13.93%.

The characterization of plant materials and N-mineralization data of this study showed that the following plant materials should be applied at the indicated rates for them to supply enough N (i.e., 80 kg N, 18 kg P, 66 kg K, 15 kg Ca and 10 kg Mg) to meet a yield of 2 tonnes grain (plus 3 tonnes stover) ha⁻¹ (Palm, 1995):- *L. leucocephala* (4.2 t ha⁻¹), *C. macrostachyus* (4.5 t ha⁻¹), *C. calothyrsus* (5.5 t ha⁻¹) and *T. diversifolia* (8 t ha⁻¹).

It was observed that the sites from where the plant materials were collected had an influence on the N, polyphenol and lignin content of the same plant species. For

example, t-test analysis indicated that leaf samples from (1) banana, (2) *C. megalocarpus*, (3) *M. lutea*, (4) Coffee, (5) *S. canipulata*, and (6) Sweet potato vines collected from Bungoma (Western Kenya) tended to have significantly ($P < 0.05$) higher nitrogen and polyphenol contents but lower lignin than those from Kabete (Central Kenya), as shown in Table 6 and Appendix I.

Table 6: The mean differences in chemical composition of same species of plant materials sampled from Bungoma (Western Kenya) compared to those from Kabete (Central Kenya)

Chemical characteristics	Mean differences of chemical contents (%)
Nitrogen (N)	0.65**
Polyphenol (Polyp)	2.14*
Lignin (L)	-8.04*
Phosphorus (P)	-0.04ns

**P < 0.01; *P < 0.05; ns=not significant

Negative value indicate less chemical concentration in materials from Bungoma than those from Kabete.

The high nitrogen concentration in materials collected in Bungoma could be attributed to the high evapo-transpiration of the area (Appendix II). Observations of Bray (1983) showed that during N transportation in the xylem from the roots, the stem tissues progressively absorb some of this nitrogen leading to progressive decrease in the nitrogen content of the xylem sap further up the stem resulting in a reduction in N available to the leaves. In case of high evapotranspiration rates, there is concentrating effect of the diluted N supplied to the leaves, thus in areas with high evapotranspiration like Bungoma, P/Eo

of 94%, plants materials would be expected to contain high N concentrations than those collected in Kabete with a lower rate of evapotranspiration of 54%. The high polyphenol content in plant materials from western Kenya (Bungoma) could be due to the low soil nitrogen which was found to favour the production of polyphenolic compounds like anthocyanins (Ribereau-Goyon, 1972).

On comparing leaves and litter of the same species collected from the same site; fresh leaves from coffee and tea had higher nitrogen and polyphenol contents but lower lignin levels than the litter from the same plants (Table 5). Similar variations were reported by Constantinides and Fownes (1994a) and Negi *et al.*, (1979) who reported that litter/or older leaf treatments of several plant species had lower initial polyphenol and nitrogen concentrations but higher lignin than fresh leaf samples.

The reduction in nitrogen content with age of plant parts, as presented in Table 5 is probably due to the retranslocation of leaf nitrogen into branches before leaf fall and partly due to dilution factor with expansion and maturity of leaves (Khosla *et al.*, 1992). The former strategy adapts the plant for conserving nitrogen when the plant sheds leaves especially during unfavourable climatic conditions. The conserved nitrogen assist the plant to regenerate when favourable conditions set in.

The difference in polyphenol content between fresh leaves and litter/or older leaves indicate that phenolic constituents are not metabolically inactive storage products simply accumulating during the whole life of a plant but are subject to relatively rapid turnover and degradation as plant parts mature. This quantitative reduction in polyphenol content with aging of plant parts was explained by Barz and Hoasel (1979) who suggested

that the change is a process that involves: Oxidative decarboxylation of phenolic compounds as plant parts mature leading to formation of naturally occurring hydroquinones and other polymers. Also some polyphenols undergo glycosylation and other conjugation processes into various structural forms like malic acid, palmitic acid or malonates. Another process by which litter can have lower polyphenol content than the fresh leaves is through leaching of the soluble compounds after the litter has fallen to the ground (Swift *et al.*, 1979). These processes lead to low polyphenol in older leaves and litter than in fresh and young leaves.

Based upon the results of this study (Table 5), it is necessary that the leafy materials of agro-forestry plants be pruned and incorporated into the soil before leaves senescence to obtain maximum nitrogen content and high N mineralization. However it may not be practical to remove field crop residues, for instance maize leaves, prior to senescence to incorporate into soil to reap of high total N and low lignin contents.

Correlation analysis between chemical contents of the plant tissues indicated that polyphenols had a weak but positive correlation with the N and carbon contents, both cases having a correlation coefficient of 0.62 (Table 7). However, lignin did not correlate with any of the other measured chemical properties. Initial P concentrations had a strong and positive correlation with N concentrations ($P < 0.001$). The ratios; Carbon to nitrogen (C:N), Lignin to nitrogen (L:N), and (lignin + polyphenol):N were negatively correlated with P contents of various plant tissues, while polyphenol:N ratio had a significant ($P < 0.05$) negative correlation with carbon content.

Table 7: Correlation coefficients of various initial chemical properties.

	Nitrogen (N)	Lignin (L)	Polyp (PP)	Carbon (C)	Phosphorus (P)	C:N	L:N	(L+PP):N	PP:N
N	1.00	0.43	0.62*	0.45	0.86***	-0.88***	-0.63*	-0.69*	-0.20
L		1.00	0.36	-0.04	0.10	-0.38	0.24	0.18	-0.32
PP			1.00	0.62*	0.30	-0.39	-0.47	-0.39	-0.49
C				1.00	0.23	-0.16	-0.66*	-0.55	-0.67*
P					1.00	-0.87***	-0.68*	-0.78*	-0.36

* P < 0.05 ; ** P < 0.01 ; *** P < 0.001.

The composition of plant residues selected for use in the incubation and glasshouse experiments is presented in Table 8. As expected N concentration of legume species were 5 - 6 times higher than that of cereals. The legumes also generally had higher concentration of P, K, Ca and Mg. The C:N ratios of the cereal were consequently much greater than those of the legumes. Polyphenol concentration however tended to be higher in legumes than in the cereal residues. Similar results were expressed by Lefroy *et al.*, (1992) who found that leguminous trees and shrubs contained higher polyphenol, astringent compounds and ash content than grasses. The higher N in the legumes could be due to the accumulation of the symbiotically fixed N (Rubaduka *et al.*, 1993). The high N content could make the legumes have better cell physiology than grasses (Bray, 1983) hence enhanced uptake of cations from the soil, making legumes to have higher contents of K, Ca and Mg.

Table 8: Chemical characteristics of the six selected plant residues

Species	tissue chemical content										
	%										
	C	N	Ca	Mg	P	K	Poly	Lignin	C:N	Lig:N	(L+P)
<i>C. macrostacyus</i>	37.50	3.30	1.02	0.45	0.29	4.03	1.37	8.40	11.36	2.55	2.96
<i>L. leucocephala</i>	36.00	3.74	1.16	0.33	0.26	3.37	6.13	14.68	9.63	3.92	5.56
<i>C. calothyrsus</i>	38.70	3.64	0.66	0.24	0.22	1.88	5.36	28.71	0.63	7.89	9.36
<i>T. diversifolia</i>	39.00	3.97	1.53	0.40	0.31	4.44	3.39	3.68	9.82	3.45	4.30
<i>S. bicolor</i>	39.90	0.63	0.49	0.14	0.10	1.40	2.92	4.23	63.33	6.71	11.35
<i>O. sativa</i>	33.00	0.63	0.10	0.04	0.14	0.38	0.00	16.66	52.38	26.44	26.44

Where: Poly= polyphenol; C:N= Carbon to nitrogen ratio; Lig:N= lignin to nitrogen ratio
(L+PP):N= (lignin+polyphenol) to nitrogen ratio; PP:N= polyphenol to nitrogen ratio

4.2. Incubation studies to assess N-mineralization patterns by selected plant materials.

4.2.1. Nitrogen mineralization patterns.

Nitrate (NO_3^- -N) constituted over 72% of the extractable inorganic N for most of the organic materials incubated throughout the 12 weeks (Table 9). However, where *Sb* leaves were added, percentage NO_3^- N

Table 9: The quantity of NO_3^- -N released by soil treated with different organic residues as percentage of total extracted inorganic N

Residues	TIME (WEEKS)					
	2	4	6	8	10	12
Cm	72.22ab	87.51ab	73.14ab	79.44bc	81.61a	79.05a
Ll	88.54a	89.78ab	82.59a	84.63ab	85.10a	75.59ab
Cc	79.09a	84.79ab	74.21ab	86.95a	81.08a	72.602a
Td	59.06bc	75.54b	79.89a	81.77abc	83.96a	77.48a
Sb	39.63c	21.90c	28.33d	49.40d	50.103c	39.18d
Os	81.41a	73.94b	66.55bc	75.06c	69.92b	64.97c
Control	88.22a	100.00a	61.93c	74.83c	68.86b	69.80bc
F test	***	****	****	****	****	****
LSD 0.05	19.55	15.92	11.14	7.14	7.84	6.65
R ²	0.76	0.91	0.91	0.92	0.91	0.94

Within a column, means followed by the same letter are not significantly different according to Fisher's LSD at a 0.05 probability level .

*** P < 0.001 ; **** P < 0.0001

Cm = *Croton macrostachyus*; Ll = *Leucaena leucocephala*; Cc = *Calliandra calothyrsus*; Td = *Tithonia diversifolia*; Sb = *Sorghum bicolor*; Os = *Oryza sativa*.

ranged between 21.90-50.10% throughout the incubation period. The high NO_3^- proportion observed in the accumulated mineral N occurred because no leaching occurred and this experiment was set up in an environment that was aerobic thus most of the ammonium released during mineralization was nitrified to nitrates.

The cumulative mineral N in the control treatment showed a gradual increase of 19.67 to 57.33 $\mu\text{g N g}^{-1}$ soil during the 12 week incubation period (Table 10). Rapid increase in cumulative soil mineral N was observed in soils in which *C. macrostachyus*, *L. leucocephala*, *C. calothyrsus* and *T. diversifolia* were incorporated. On mixing *O. sativa* husks and *S. bicolor* leaves individually with the soil, cumulative mineral N release was lower (16.0-41.33 $\mu\text{g N g}^{-1}$ soil for *S. bicolor* and 17.67-58 $\mu\text{g N g}^{-1}$ soil for *O. sativa* husks) than that of soil alone (19.67 - 57.33 $\mu\text{g N/g}$ soil) during the incubation period (Table 10).

Incorporation of *S. bicolor* leaves resulted in net N immobilization throughout the 12 week incubation period. Addition of leaves of *O. sativa* husks led to a net immobilization between 2 -10 weeks and thereafter a slow N release was observed.

The net inorganic N released after 12 weeks of incubation varied significantly among species. The net N released ranged from -16.33 $\mu\text{g N g}^{-1}$ soil (net immobilization) for *S. bicolor* to 43.33 $\mu\text{g N g}^{-1}$ soil (net mineralization) for *L. leucocephala* (Fig. 1). When *L. leucocephala* and *C. macrostachyus* leaves were added to the soil, a significantly ($P < 0.05$) higher net inorganic N was released followed by *C. calothyrsus* and *T. diversifolia*.

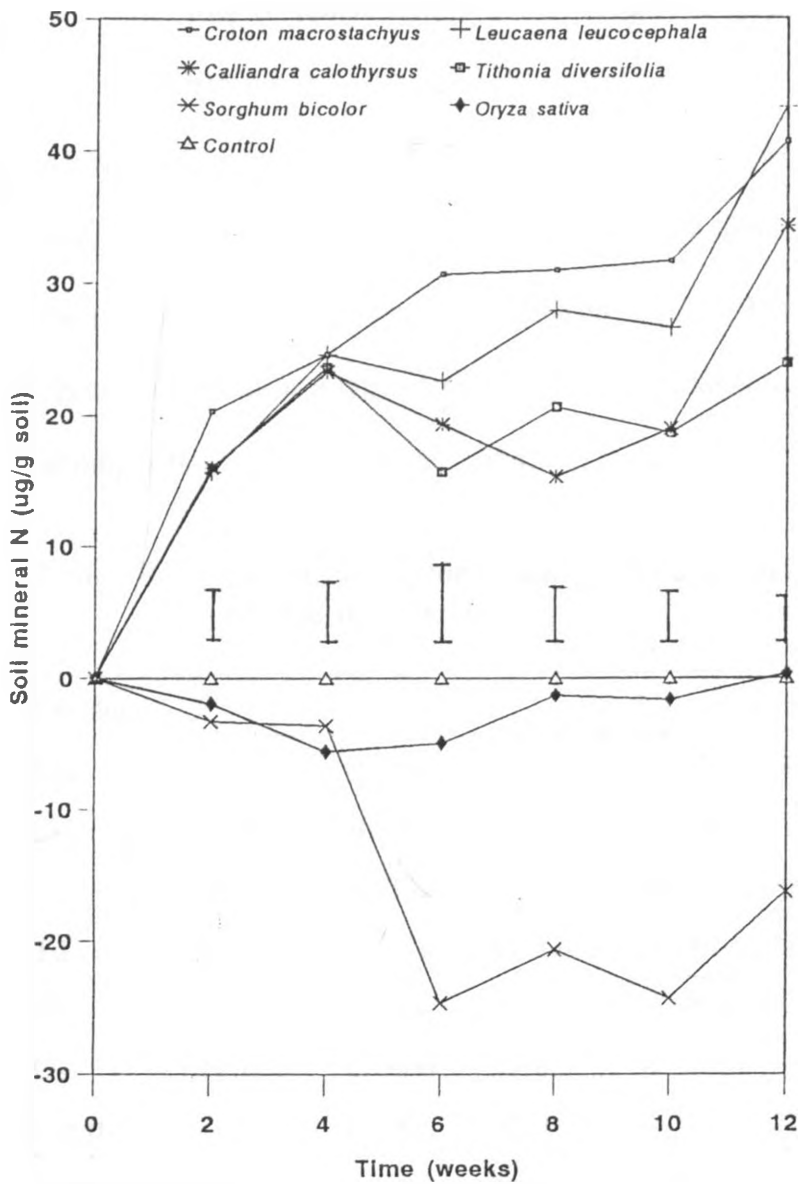


Fig. 1: Changes in net inorganic soil N (treatment inorganic N - control) during 12 week incubation as affected by addition of plant residues of contrasting chemical composition. Bar represents LSD at 0.05 probability level.

The low N-mineralization pattern observed in presence of *T. diversifolia* was unexpected because the organic resource had high initial N content and medium lignin and polyphenolics compared to *L. leucocephala*. The fact that not all polyphenol compounds have equivalent capacity to bind protein or nitrogen may explain the unexpected observation (Martin and Martin, 1982). Hence, *T. diversifolia* may have a higher proportion of a type of polyphenol that binds nitrogen which delayed the rate of decomposition and net mineralization.

Table 10: Effect of organic residues on cumulative mineral N ($\text{NO}_3^- + \text{NH}_4^+$) release in $\mu\text{g/g}$ soil)

Residues	2	4	6	8	10	12
Cm	40.00a	46.00a	74.67a	74.33	81.33a	98.33a
Li	35.00a	46.00a	67.00a	71.33a	76.00a	101.00a
Cc	35.67a	44.33a	63.33b	58.67b	68.33b	92.33b
Td	35.67a	45.67a	59.33b	64.00b	68.33b	81.33c
Sb	16.00b	18.00c	18.33d	23.99d	25.33d	41.33e
Os	17.67b	27.00b	39.00c	42.67c	47.67c	58.00d
Control	19.67b	21.00	44.0c	43.67c	49.33c	57.33d
F test	****	****	****	****	****	****
L.S.D 0.05	7.35	11.72	9.622	5.68	6.17	6.05
R ²	0.88	0.92	0.94	0.98	0.98	0.98

(1) Within a column means followed by the same letter are not significantly different according to Fisher's LSD at 0.05 probability level.

(2) *** P < 0.001

Cm = *Croton macrostachyus*; Li = *Leucaena leucocephala*; Cc = *Calliandra calothyrsus*; Td = *Tithonia diversifolia*; Sb = *Sorghum bicolor*; Os = *Oryza sativa*.

The net immobilization of N during incubation of *O. sativa* and *S. bicolor* leaves with soil could be explained by their high C:N ratio. Usually net mineralization will occur when the C:N ratio of the organic resource is equal to or lower than that of the decomposer organisms which is usually about 20 (Swift *et al.*, 1979). In the case of *O. sativa* and *S. bicolor* with C:N ratios of 52 and 63 respectively, the decomposer microbial community had narrower C:N ratio than the resources leading to a high demand of N by the decomposers. In fact the demand was so high that N was being imported from the soil solution in contact with the decomposing resources making the N-mineralization curves of the two resources to be below that of soil alone (Fig. 1). Comparing treatments with the same amount of N (*S. bicolor* and *O. sativa*), *S. bicolor* with a higher C:N ratio resulted in more immobilization of mineral N than *O. sativa*, though immobilization was taking place in both cases. This is due to increased incorporation of soil N into the microorganism tissues in the soil mixed with *S. bicolor*. Similar trends of immobilization after amendment of soil with carbon sources of higher C:N ratios were observed by Ladd *et al.* (1977) and Azam *et al.* (1986, 1988).

In this study net mineralization for *O. sativa* treatment was detected before the end of the 12th week. However, the trend of nitrogen release which the treatment with *S. bicolor* had started showing indicate that release of N is expected sometimes after the 12th week (Fig. 1). This could be explained by either or both of the following: Firstly, during mineralization of an organic material, carbon (C) in the material usually mineralizes faster than the nitrogen (N) component hence with time the C:N ratio of the material decreases (Sanchez, 1976). Based on this, Stevenson (1982), suggested that net

N immobilization from an organic material with a high C:N ratio mixed with soil, lasts until the C:N ratio of the decomposing material has been lowered to approximately 20. In the case of this study, the C:N ratios of *O. sativa* and *S. bicolor* were 63 and 52 respectively. Accordingly, it was expected that *O. sativa* husks would start showing net release before *S. bicolor* leaves, which was confirmed by this experiment. Secondly, presence of higher polyphenol content in *S. bicolor* leaves might have consisted largely of macromolecules with higher stability that were only slowly utilized by the microorganisms (Quemada and Cabreta, 1995).

The percentage of N recovered from the added plant materials in form of inorganic N after 12 weeks of incubation was significantly high ($P < 0.05$) in presence of *C. macrostachyus* (54%), *L. leucocephala* (51%) and *C. calothyrsus* (42%) but relatively low where *T. diversifolia* (26%) and *O. sativa* (2.7%) and *S. bicolor* (-114%) were added (Table 11).

Based on the initial quantity of N added to the soil, the results of this experiment and the quantities recovered at the end of twelve weeks showed that not all of the mineralized N was recovered and this might have been due to fixation of NH_4^+ on the clay lattices that was not accounted for in the final balance (Ladd et al, 1977).

Table 11: Fraction of initial quantity of N recovered from plant materials in form of mineral N during 12 week incubation period.

Residues	TIME (WEEKS)					
	2	4	6	8	10	12
Cm	0.27a	0.33a	0.41a	0.41a	0.43a	0.54a
Ll	0.19ab	0.29a	0.27a	0.33ab	0.31a	0.51a
Cc	0.19ab	0.28a	0.23a	0.18b	0.23ab	0.42a
Td	0.18ab	0.26ab	0.17a	0.23b	0.21ab	0.26ab
Sb	-0.25d	-0.24c	-1.78c	-1.44d	-1.68d	-1.14c
Os	-0.14cd	0.39a	-0.35b	-0.08c	-0.13c	0.03b
F test	***	**	*****	*****	*****	*****
LSD 0.05	0.21	0.26	0.46	0.16	0.28	0.31
R ²	0.78	0.74	0.92	0.98	0.96	0.93

Within a column, means followed by the same letter are not significantly different according to Fisher's LSD at 0.05 probability level. ** P < 0.01 ; *** P < 0.001 ; ***** P < 0.0001

Cm = *Croton macrostachyus*; Ll = *Leucaena leucocephala*; Cc = *Calliandra calothyrsus*; Td = *Tithonia diversifolia*; Sb = *Sorghum bicolor*; Os = *Oryza sativa*.

4.2.2. Correlation of N released with quality of the organic resources.

Correlation analysis was used in an attempt to identify the residue characteristics that best predict the N release. The total amount of mineral N accumulated was positively correlated with initial N and P concentrations of the organic residues during each time

period apart from week 4 (Table 12). At week four the correlation between amount of mineral N accumulated and the initial N and P was positive but insignificant. Initial soluble polyphenol, lignin and carbon contents were not correlated with the mineral N accumulation in the soil. The N accumulation was negatively correlated with the C:N and polyphenol:N ratios. Also, there was a negative correlation between the cumulative N and (lignin + polyphenol):N ratio at week 2.

The correlation between the initial N and P contents with N accumulation over the incubation period agrees with the findings of Frankenberger and Abdelmagid (1985) and Tian *et al.*, (1992). This is probably because nutrient concentration particularly N and P commonly limit the activity of the decomposer organisms which require these two elements in large quantities for their growth and development (Swift *et al.*, 1979). Hence if these two nutrients are in high concentration in the residues added to the soil, they will supply adequate amounts of N and P needed for the decomposer organisms stimulating their growth and activity. This would lead to increased N mineralization and release to the soil from such organic material making them be of high quality.

The correlation coefficient between lignin and N accumulation was low (ranging from -0.27 to -0.49). This indicates that, although lignin has been shown to be an important factor by other researchers like Tian *et al.*, (1992) and Oglesby and Fownes (1992), it cannot alone explain the differences in the N release between the different plant residues added to the soil. This lack of correlation could be explained by the fact that the function of lignin in determining and regulating the rate of decomposition is through its physical proximity to the cellulose molecules thus retarding the enzymatic attack on the cellulose.

Table 12: Correlation coefficients (r) of cumulative N mineralized at each time interval versus

Time (wks)	Cum.Min.N($\mu\text{g/g}$ soil)	Nitrogen	Lignin	Poly	Carbon	Phosphorus	C:N
2	28.52	0.63*	-0.27	-0.05	-0.14	0.78**	-0.90***
4	35.43	0.38	-0.49	-0.25	-0.58	0.46	-0.59*
6	52.24	0.71**	-0.45	-0.10	-0.23	0.75**	-0.90***
8	53.95	0.63*	-0.42	-0.06	-0.30	0.69*	-0.86***
10	59.48	0.60*	-0.43	-0.00	-0.36	0.67*	-0.84***
12	75.67	0.59*	-0.45	-0.02	-0.37	0.68*	-0.84***

* $P < 0.05$; ** $P < 0.01$; *** $P < 0.001$

Where: Poly = polyphenol; C:N = Carbon to nitrogen ratio; L:N = lignin to nitrogen ratio; (L+PP):N = lignin plus polyphenol to nitrogen ratio; PP:N = polyphenol to nitrogen ratio; Cum. Min. N = Cumulative N released at each time interval

The rate of the decomposition curve in most cases is determined by the decomposition of cell wall polysaccharides, i.e, cellulose and hemicellulose which account for 70 - 90% of the dry weight of most plant residues (Swift et al, 1979). May be in these residues, the proportion of cellulose and hemicellulose were too high to be interfered with by the lignin association. Hence there is need to find the effect of lignin-to-cellulose ratio on the effect of N release. These results agree with those of Heal *et al.*, (1978). However, Palm and Sanchez (1991), Tian *et al.*, (1992) and Constantinides and Fownes (1994a) found lignin to be weakly correlated to mineral N accumulated during incubation.

Initial polyphenol content was found to have no influence on N release from various plant residues but the polyphenol:N ratio had a negative influence on N mineralization (Table 12). Constantinides and Fownes (1994a) reported similar correlation of polyphenol:N but not polyphenol alone on N-mineralization. Thus, the polyphenol:N ratio could be used to predict N-mineralization of the various plant residues.

Lack of correlation between N mineralized and polyphenol content but high correlation with polyphenol:N ratio could be explained by the fact that the effect of polyphenol on N-mineralization is basically due to interaction of the polyphenolics with N (Palm and Sanchez, 1991). For instance, some polyphenolics form complex structures by H-bonding with basic N-containing groups, others form stable cross linkages with amino groups making the material resistant to decomposition (Swain, 1979). Phenolics are also readily oxidized to quinones that react with nitrogen in amino acids and amino sugars to form stable polymers that cannot be broken down during decomposition (Martin and Haider, 1980).

4.3. Mixtures of high quality and low quality organic residues.

The effects of mixing *C. macrostachyus* (Cm) and *O. sativa* (Os) on the amount of mineral N released varied greatly depending on the amount of Cm added. In absence of Cm, addition of Os in the soil suppressed the amount of N-mineralized to less than that of soil alone. When Cm and Os mixture was incorporated into the soil, a net N release was recorded and the release patterns were intermediate to those of Cm and Os (Fig.2).

When Cm and Os were mixed in a 1/2: 1/2 ratio the N release pattern was very close to that predicted by getting the means of the amounts of the N released from individual residues. On mixing 1/4 Cm with 3/4 Os, the N release pattern of Cm was suppressed considerably and the release pattern was closer to that of the soil alone. However, the observed N release pattern was higher than that of the theoretical(predicted) mixture between 0-4 weeks and lower than the predicted between 4 -10 weeks. When 3/4 Cm and 1/4 Os mixture was added, the amount of N released was more than that predicted from 0 - 4 weeks and thereafter lower for the rest of the incubation period.

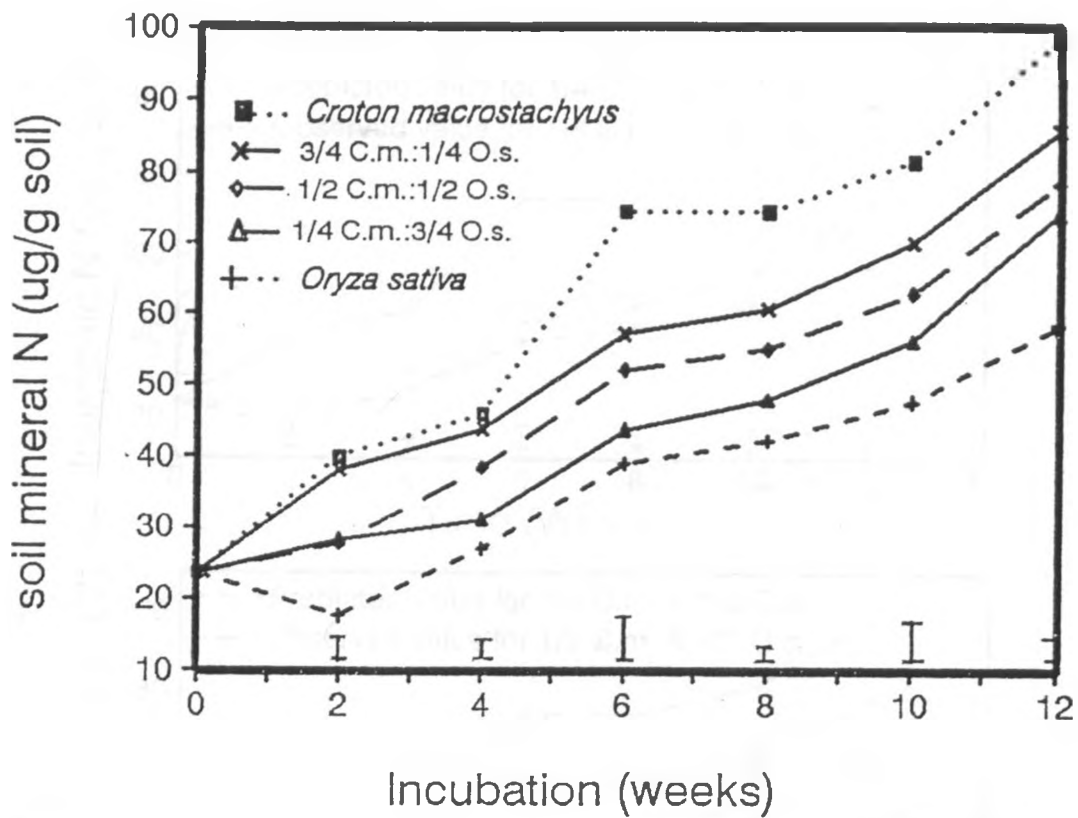


Fig. 2: Changes in soil inorganic N release during 12 week incubation as affected by addition of mixtures of *C. macrostachyus* (Cm) and *O. sativa* (Os) in various ratios to the soil. Bar represents LSD at 0.05 probability level.

Croton macrostachyus (C.m.)
 Oryza sativa
 Control

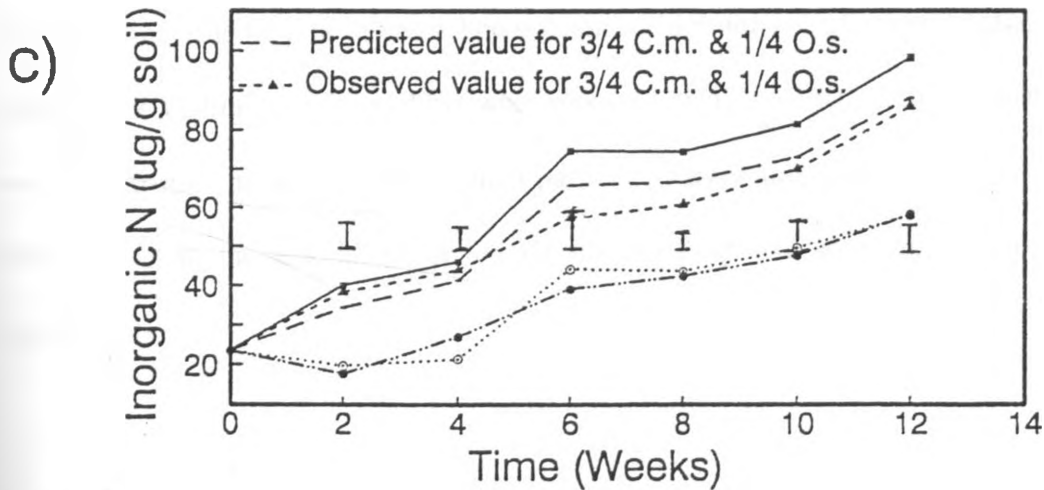
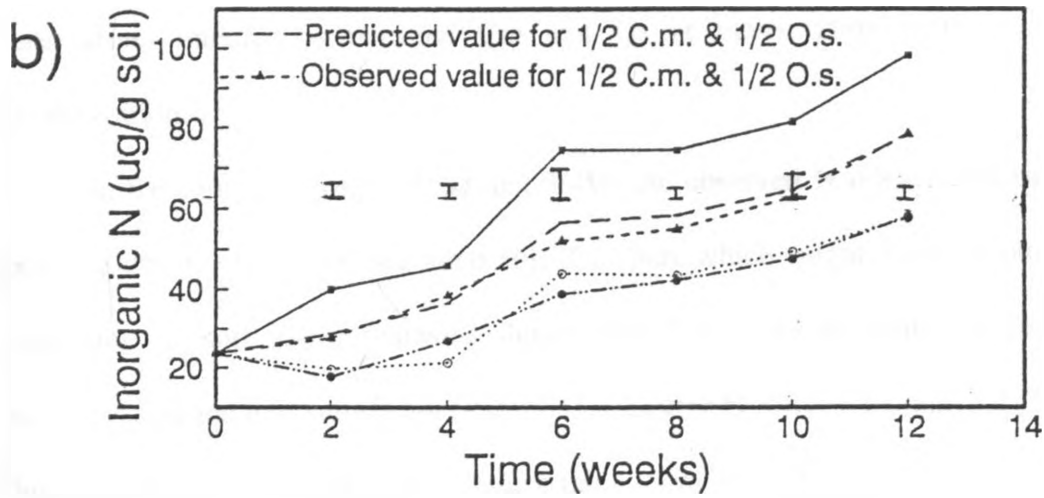
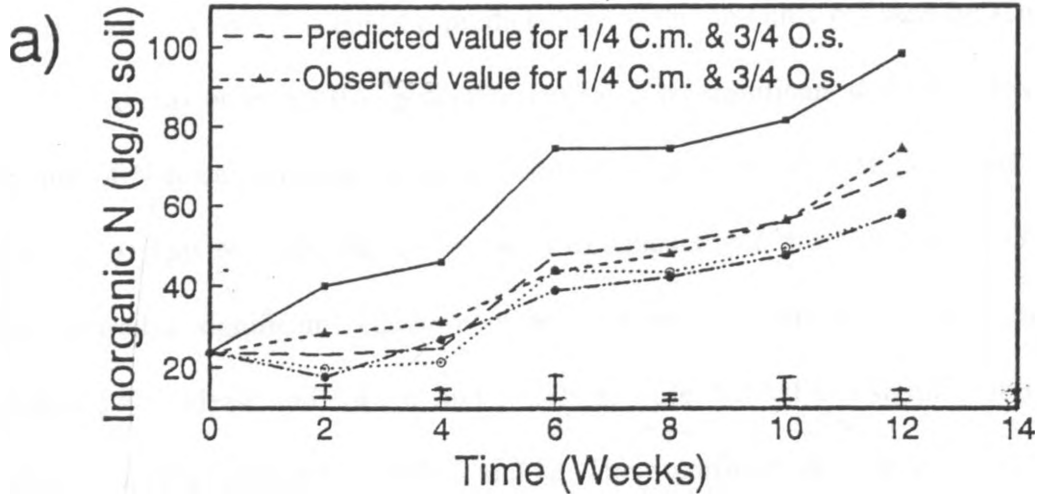


Fig. 3: Predicted and observed soil inorganic N changes during 12 week incubation of (a) 1/2 C.m. & 1/2 O.s., (b) 1/4 C.m. & 3/4 O.s., (c) 3/4 C.m. & 1/4 O.s. and individual C.m & O.s. Bar represents LSD at 0.05 probability level.

A contrast was computed to determine if the those of N accumulated from the mixtures of Cm and Os could be predicted from the amounts released by Cm and Os residues. It was observed that generally there was no significant difference between the amount of N released during the incubation of the mixtures and the predicted N-release (Table 13). This was with the exception of treatments with 3/4 Cm and 1/4 Os mixture that showed a significant reduction of N - release at weeks 6 - 8 compared to the predicted N - release and 1/4 Cm and 3/4 Os mixture that led to a significant higher N - release than the predicted at weeks 2 and 12. This significant differences meant that there were strong interactions between Cm and Os at these ratios at these stages of decomposition.

In the case of mixing 1/4Cm and 3/4Os, the observed N release pattern (Fig.3a) could be due to the easily decomposable Cm part which might have encouraged the flourishing of soil microorganisms during the first 2 weeks hence increasing the decomposition and thus significant ($P < 0.05$) release of N above the predicted (Table 13). This is because mixing Cm and Os reduce the C:N ratio of Os from 52.38 to 25.25 thus contributing to the high N-release. The speculated high microbial activity agreed with the findings of Collins *et al.*, (1990) who reported that in a 30 - day incubation, CO₂ evolution from a mixture of wheat plant parts (ie. leaves and stems) increased upto 25% with respect to the amount predicted by summing up CO₂ evolution from individual components.

Table 13: The amount of mineral N released during incubation above those predicted by mixing Os and Cm ($\mu\text{g/g}$ soil).

Time (wks)	1/2 Cm + 1/2 Os	1/4 Cm + 3/4Os	3/4 Cm + 1/4Os
2	-1.17	5.08*	3.92
4	2.17	-0.75	2.75
6	-4.83	-3.92	-8.75**
8	-3.5	-2.42	-5.58**
10	-1.67	0.00	-2.67
12	-0.17	5.92**	-2.58

(1) * $P < 0.05$; ** $P < 0.01$. (2) Where: Cm = *Croton macrostachyus*; Os = *Oryza sativa*; (3) In the table, a contrast that did not show significant difference from the predicted N-release (i.e. {amount of N by Cm + amount of N by Os}/2) indicated that the values for Cm and Os mixtures can be predicted from the sum of Cm and Os release curves.

After 4 weeks, the easily decomposable Cm had been completed and the microorganisms were subjected to the remaining less decomposable Os with a high C:N ratio. The reduced easily available nutrients could have led to bacterial population going dormant with an effect of reduced decomposition hence N - release was reduced to below that predicted (Struwe and Kjoller, 1986). However as the C:N ratio of Os kept on decreasing with decomposition (Sanchez, 1976), N release increased though below the predicted values. The results of this experiment are consistent with those obtained by Struwe and Kjoller (1986) who found that when nitrogen rich alder litter was mixed with nitrogen poor ash litter, there was initially high population of starch and gelatin utilizing bacteria but bacteria population declined when the alder litter was depleted. These authors suggested that the population shift was related to the progressive decomposition and consequently to the depletion of easily available nitrogen and energy sources.

4.4. N uptake by *Zea mays* (maize)

4.4.1. Maize dry matter yield

The effect of plant residues on maize growth was evident from the early stages (Table 14). For example, at 3 weeks after planting (WAP) the above ground dry matter was 29 - 58% higher in pots that had received plant residues and at 5 WAP it was 24 - 62% higher and 7 WAP, 3 - 38% higher than in the control pots. Similar trends have been shown in the past by Mulongoy and Van der Meersch (1988) using *L. leucocephala* and maize stover as organic amendments. Weight of above ground dry matter in all treatments increased gradually until 3 WAP and rapidly thereafter (Table 14).

Table 14: Dry matter production by maize plants grown in soils treated with various plant residues.

Treatment	Maize dry weight (g/pot)		
	Time in weeks after planting (WAP)		
	3	5	7
Control	0.24a	0.68a	1.45a
<i>C. macrostachyus</i> (Cm)	0.36b	0.95bc	1.70b
<i>O. sativa</i> (Os)	0.31ab	0.84ab	1.48a
1/2 Cm + 1/2 Os (mixture)	0.38b	&1.10c	2.00c
<i>T. diversifolia</i> (T.d)	0.34b	0.93bc	1.81bc
LSD (0.05)	0.09	0.22	0.22

Within a column, means followed by the same letter are not significantly different at LSD at 0.05 probability level.

Addition of plant residues significantly ($P < 0.05$) increased maize biomass above the control throughout the growth period. At 7 WAP the highest response to residue application was observed in treatments with $\frac{1}{2}\text{Cm} + \frac{1}{2}\text{Os}$ mixture followed by Td and Cm respectively. Where Os husks alone were added, there was no significant difference in dry matter production when compared with soil alone treatment throughout the growth period.

A contrast was computed to determine if *C. macrostachyus* and *O. sativa* interacted to give significant increase in dry matter yield in the mixed residue. The results in Table 15 showed that the observed dry matter yield obtained in the mixture was significantly larger than that predicted from isolated Cm and Os yields throughout the growth period apart from 3 WAP. These results indicate that there was a significant positive interaction between Cm and Os on maize dry matter yield.

The yield increase was probably not as a result of N released from the mixture since N release from the mixture during laboratory incubation was lower than that released from *C. macrostachyus*. The high yield in the pots receiving the Cm and Os mixture could have been due to the low quality *O. sativa* portion in the Os and Cm mixture having effectively improved the soil physical conditions that contributed to better plant growth (Yamoah *et al.*, 1986).

Table 15: Maize dry matter yield production above that predicted from *C. macrostachyus* and *O. sativa* mixture at each sampling time.

Time	yield (g/pot)		
	observed	predicted	(observed-predicted)
3	0.38	0.33	0.05
5	1.10	0.89	0.21*
7	2.00	1.59	0.41**

* $P < 0.05$; ** $P < 0.01$

In the table, predicted yield was obtained by (Dry matter from Cm treatment + Dry matter yield from Os treatment)/2.

Another possible explanation is that, the high carbon portion of *O. sativa* in the mixture may have enhanced the activity of N_2 -fixing bacteria living in the rhizosphere in association with maize plant. Similarly, Martin *et al.* (1989), using high carbon millet straw and low carbon *Gliricidia sepium* in a field of maize, observed that millet straw, unlike *Gliricidia*, significantly enhanced the activity of N_2 fixing bacteria in the maize rhizosphere. This enhanced microbial activity could have led to more mineral N available to the maize plants during the turnover of the bacteria than was the case with *C. macrostachyus*. These results agreed with the research work of Van Der Meersch *et al.* (1993) who found that in cases where a highly decomposable organic material has been added to the soil, the ability of microorganisms living in the soil to fix nitrogen is reduced.

4.4.2. Dynamics of soil mineral N in the presence of maize crop.

In the control treatment mineral-N content fluctuated between 79.2 $\mu\text{g N/g}$ soil at 3 WAP and 43.9 $\mu\text{g N/g}$ soil at 7 WAP (Fig 4). The soil solution mineral-N increased by 128% with addition of *C. macrostachyus* by the 7th week of the experiment. Addition of $\frac{1}{2}$ Cm and $\frac{1}{2}$ Os mixture and *O. sativa* alone increased soil mineral N by 76% and 65% respectively at the end of the 7th week. There was a general decrease in extractable N possibly due to the plant uptake as reported by Tian *et al.*, (1993) and Franzluebbbers *et al.*, (1994).

There was net immobilization of mineral-N in the laboratory incubation where *O. sativa* residue was applied but a net N release was recorded in the pot experiment (Fig. 4). Some short-term immobilization was expected in pots with *O. sativa* having a high C:N ratio of 52 (Iritani and Arnold, 1960). However, the high day temperatures of over 35°C in the glasshouse compared to the low laboratory incubation temperatures (21°C-room temperature), may have increased microbial activities leading to the net N release.

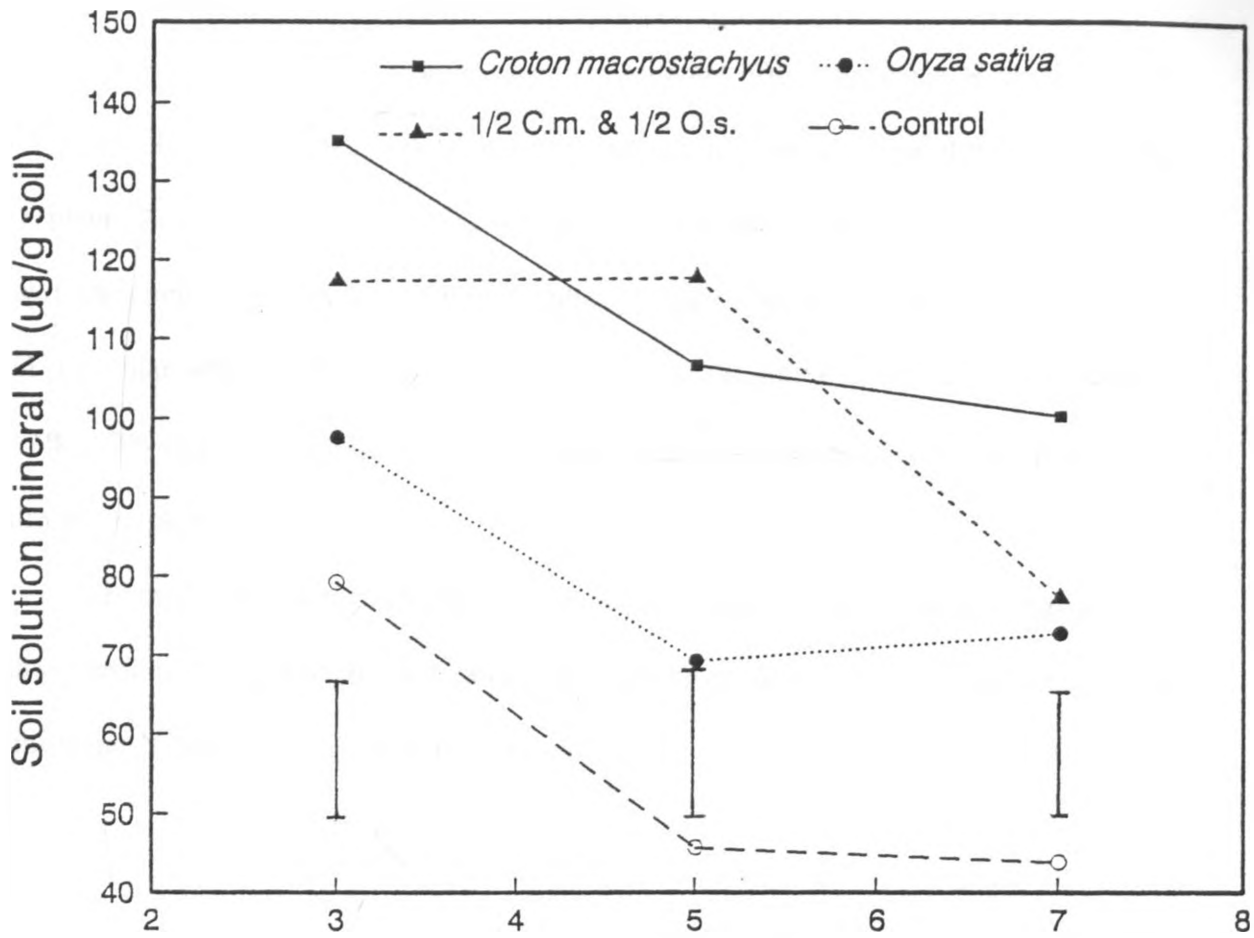


Fig. 4: Changes in extractable soil N as influenced by different plant materials in the glasshouse maize N uptake experiment. Bar represents LSD at 0.05 probability level.

4.4.3. Nitrogen uptake by *Zea mays* (maize).

Addition of organic residues had a significant effect on maize N uptake (Table 16). The control treatment gave low N uptake values ranging from 9.29 to 20.60 mg N/plant. Addition of *O. sativa* did not improve N uptake significantly when compared with the control. However, addition of other residues enhanced N uptake by maize and in particular when *C. macrostachyus*, $\frac{1}{2}$ Cm + $\frac{1}{2}$ Os and *T. diversifolia* were added. 62%, 59% and 44% of total available N above the control respectively was taken up by the maize plant.

Increase in N uptake by maize in presence of plant residues implies that some of these residues are potential candidates that could be used as sources or supplements of inorganic-N fertilizers in maize production.

Table 16: Influence of organic residue addition on N uptake by *Zea mays* (maize)

Treatment	N uptake in mg/plant		
	weeks after planting (WAP)		
	3	5	7
Control	9.29a	14.55a	20.60a
<i>C. macrostachyus</i> (Cm)	13.34b	21.09b	33.32c
<i>O. Sariva</i> (Os)	11.32ab	16.78a	19.98a
1/2 Cm + 1/2 Os (mixture)	14.86c	23.82bc	29.76b
<i>T. diversifolia</i>	12.99bc	24.46c	32.76bc
LSD (0.05)	2.43	4.84	2.98

Within a column, means followed by the same letter are not significantly different at LSD at 0.05 probability level.

A simple regression analysis was carried out to obtain the relationship between N uptake and extractable N at 5 and 7 WAP. Significant co-efficients of determination (R^2) were found with equations generated which could be used for estimating N uptake and are presented below:-

(i) 5 WAP: $N \text{ uptake} = 0.809 + 0.014 * \text{Soil N}$ $R^2 = 0.87^{**}$

(ii) 7 WAP: $N \text{ uptake} = 0.783 + 0.025 * \text{Soil N}$ $R^2 = 0.71^*$

The significant R^2 indicated that the N supply and N uptake are inter-linked processes. The high correlation between N uptake by maize and the change in soil mineral N content imply that knowledge of soil mineral-N levels is a reliable predictor of the level of plant N uptake from organic residues with known qualities. Similar high correlation were obtained by Olf and Werner (1994) after planting rye grass in a grass-clover residue amended soil and Fauci and Dick (1994) in maize grown in pots which had received various levels of organic amendments.

CHAPTER FIVE

5.0 Conclusions and recommendations.

5.1. Conclusions

The site from which plant materials were collected had considerable influence on their chemical composition. Plant materials from western Kenya (Bungoma) had higher nitrogen and polyphenols content but low lignin than those from central Kenya (Kabete-Nairobi).

The contents of N and P as well as C:N and polyphenol:N ratios were correlated with N release under controlled incubation experiment proving their importance in residue decomposition and N release. Thus N release patterns of plant residues can be predicted from analysis of N and P content and C:N and polyphenol:N ratios in different plant materials.

Generally mixing low quality and high quality plant materials resulted in higher N release during the first four weeks. Incorporation 3/4 Cm:1/4 Os mixture caused a significant suppression of the N released from the high quality residue at the 6 - 8th week. This effect is likely to improve N use efficiency through reduction of N losses with a positive N residual effect on the succeeding crops.

S. bicolor leaves released N which was less than that of soil alone throughout the study period, indicating that some N was being immobilized from soil due to the presence of this residue. This is important for optimising crop management in Malava area of Kakamega district where the predominant crop rotation practised is maize followed by

sorghum and then maize. The immobilization after incorporation of sorghum residue into the soil during short rains may lead to N unavailability to the following maize which is planted less than 2 months after harvesting sorghum. Therefore the amount of N immobilized should be accounted for when evaluating N requirement of the following maize crop and be supplied by another quick N- releasing organic material or by inorganic fertilizers.

Although results from incubation studies can be used to predict N release patterns and accumulation from various plant residues, they may not work out well for the estimation of plant uptake and response to N released from these residues. This is exemplified by the experiment of maize dry matter yield and N uptake responses to various plant residues. It was shown that the treatment with $\frac{1}{2}$ *Croton macrostachyus* + $\frac{1}{2}$ *Oryza sativa* contributed more positively to maize production than the high quality *C. macrostachyus* alone. However, during the incubation experiment, *C. macrostachyus* treated soil released more N than that treated with the mixture. The high response of maize dry matter yield in the mixed residue treatment implies that mixing low quality and high quality plant residues in a proper ratio is a prerequisite for synchronizing soil nutrient supply and crop nutrient demand.

Determination of N levels in the soil solution during the plant growth period was shown to be a better tool for evaluating the possible N contribution of plant residues to the crops as N uptake in maize was correlated with the extractable soil N.

5.2. Recommendations and guidelines for further research.

In areas where *O. sativa* husks are available, it is recommended that the husks should be mixed with either *C. macrostachyus* or *L. leucocephala* in a ratio of ½ to ½ before they are incorporated into the soil for them to supply sufficient N for maize growth. However, availability of the suggested high quality plant materials that are to be mixed with the low quality materials was beyond the scope of this study. Consequently it is recommended that research to assess availability, acceptability of the organic resources by the farmers and their decay patterns in the field should be carried out. Where, farmers have high amounts of *S. bicolor* residues, they should not be incorporated in the soil for the results of this study have shown that *S. bicolor* immobilises N for more than 12 weeks hence rendering N unavailable for most of the annual crops. However *S. bicolor* residues could supply some N to the crops if applied at least four months before planting. The appropriate time to apply these residues for efficient N release to the crops by these residue materials was not done during this study and is an area recommended for future research.

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APPENDIX I

THE EFFECT OF SITE OF SAMPLING ON THE CHEMICAL COMPOSITION OF PLANT MATERIALS: t- TEST ANALYSIS TABLE.

N			Lignin			Polyphenols			P		
Y ₁	Y ₂	D	Y ₁	Y ₂	D	Y ₁	Y ₂	D	Y ₁	Y ₂	D
2.50	1.30	1.20	9.51	11.99	-2.48	0.49	1.78	-1.29	0.13	0.10	0.03
2.60	1.93	0.67	16.54	25.67	-9.13	6.25	1.84	4.41	0.18	0.15	0.03
3.30	3.30	0.00	13.87	8.40	5.47	4.41	1.37	3.04	0.27	0.29	-0.02
2.47	1.87	0.60	18.79	28.14	-9.35	4.56	2.00	2.56	0.17	0.21	-0.04
2.80	2.30	0.50	13.65	30.29	-16.64	5.41	5.06	0.35	0.19	0.18	0.01
2.03	1.63	0.40	14.63	31.34	-16.71	5.57	3.09	2.48	0.15	0.18	-0.03
2.90	1.73	1.17	5.93	13.35	-7.42	5.80	2.38	3.42	0.24	0.48	-0.24
18.6	14.0	4.54	149.8	92.92	-56.26	32.49	17.53	14.97	1.59	1.33	-0.26

$$\sum [Y_{1j} - Y_{2j}]^2 = \sum D^2 \dots \dots \dots 1$$

$$S_D^2 = \frac{\sum D^2 - \frac{[\sum D]^2}{N}}{n-1} \dots \dots \dots 2$$

$$S_D^2 = \frac{S_D^2}{n} \dots\dots\dots 3$$

$$t = \frac{\bar{D}}{S_D} \dots\dots\dots 4$$

Y_1 = Chemical contents of plant materials from Sangalo (Bungoma)

Y_2 = Chemical contents of plant material from Kabete.

APPENDIX II

Soil and Climatic Characteristics of Sangalo (Bungoma District) and Kabete areas.

	Sangalo	Kabete
Soil Classification	Orthic Ferralsols	Humic nitisols
Soil physical characteristics		
Sand (%)	52	5.0
Silt (%)	16	23
Clay (%)	32	72
Chemical characteristics		
pH (H ₂ O)	5.2	4.9
% C	1.44	2.47
% N	0.12	0.18
C:N	12	14
P (ppm)	6	12
Climatic characteristics		
Mean annual temp.(°C)	22	18
Potential Evapotranspiration (P/E _o) (%)	94	56

Source: Muchena and Siderius (1977); Where P is precipitation while E_o is evapotranspiration.

APPENDIX III

Analysis of variance procedure: Cumulative N mineralized at various weeks.

Week 2					
Source of Variation	Degree of freedom	Sum of squares	Mean squares	F value	
Treatment	6	1886.57	14.43	17.85	
Error	14	246.67	17.62		
Total	20	2133.24			
	R-Square	C.V	Root MSE		NMIN M
	0.88	14.72	4.20		28.52

Week 4					
Source of Variation	Degree of freedom	Sum of squares	Mean squares	F value	
Treatment	6	2971.81	493.30	28.73	
Error	14	241.33	17.23		
Total	20	3213.14			
	R-Square	C.V	Root MSE		NMIN M
	0.92	11.72	4.15		35.43

Week 6					
Source of Variation	Degree of freedom	Sum of squares	Mean squares	F value	
Treatment	6	6861.14	1143.52	37.88	
Error	14	422.67	30.19		
Total	20	7283.81			
	R-Square	C.V	Root MSE		NMIN M
	0.94	10.51	5.49		52.24

Week 8

Source of Variation	Degree of freedom	Sum of squares	Mean squares	F value
Treatment	6	6095.62	1015.94	96.54
Error	14	147.33	10.52	
Total	20	6242.95		
	R-Square	C.V	Root MSE	NMI
	0.98	6.01	3.24	5

Week 10

Source of Variation	Degree of freedom	Sum of squares	Mean squares	F value
Treatment	6	6947.24	1157.87	93.16
Error	14	174.00	12.43	
Total	20	7121.24		
	R-Square	C.V	Root MSE	NMI
	0.98	5.93	3.53	5

Week 12

Source of Variation	Degree of freedom	Sum of squares	Mean squares	F value
Treatment	6	9877.33	1646.22	137.73
Error	14	167.33	11.95	
Total	20	10044.67		
	R-Square	C.V	Root MSE	NMI
	0.98	4.57	3.46	7

APPENDIX IV
Analysis of variance of mixed residues at various weeks
Week 2

Source of Variation	Degree of freedom	Sum of squares	Mean squares	F value	P
Treatment	4	986.93	246.73	13.71	0
Error	10	180.00	18.00		
Total	14	1166.93			
	R-Square	C.V	Root MSE		NMIN M
	0.85	39.52	4.24		10.73

Week 4

Source of Variation	Degree of freedom	Sum of squares	Mean squares	F value	P
Treatment	4	804.67	201.17	14.03	0
Error	10	143.33	14.33		
Total	14	948.00			
	R-Square	C.V	Root MSE		NMIN M
	0.85	23.66	3.79		16.00

Week 6

Source of Variation	Degree of freedom	Sum of squares	Mean squares	F value	P
Treatment	4	2288.27	572.17	13.47	0
Error	10	424.67	42.47		
Total	14	2713.33			
	R-Square	C.V	Root MSE		NMIN M
	0.84	69.82	6.52		9.33

Week 8

Source of Variation	Degree of freedom	Sum of squares	Mean squares	F value	P > F
Treatment	4	1875.07	468.77	47.51	0.0001
Error	10	98.67	9.86		
Total	14	19.73			
	R-Square	C.V	Root MSE		NMIN Mean
	0.95	25.06	3.14		12.53

Week 10

Source of Variation	Degree of freedom	Sum of squares	Mean squares	F value	P > F
Treatment	4	1968.40	492.10	14.25	0.0004
Error	10	345.33	34.53		
Total	14	2313.73			
	R-Square	C.V	Root MSE		NMIN Mean
	0.85	41.58	5.88		14.13

Week 12

Source of Variation	Degree of freedom	Sum of squares	Mean squares	F value	P > F
Treatment	4	2655.07	663.77	43.48	0.0001
Error	10	152.66	15.27		
Total	14	2807.73			
	R-Square	C.V	Root MSE		NMIN Mean
	0.95	18.49	3.91		21.13

APPENDIX V

Computation of contrasts for N-released from mixed residue experiment

Week 2

Source of variation	Degree of freedom	Type I SS	Mean squares	F value	P > F
Treatment	4	986.93	246.73	13.71	0.0005
Parameter	Estimate	T for HO:	Pr > T	Std Error of Estimate	
1/2Os + 1/2Cm	-1.17	-0.39	0.71	3.00	
3/4Os + 1/4Cm	5.08	1.63	0.13	3.12	
1/4Os + 3/4Cm	3.92	1.25	0.24	3.12	

Week 4

Source of variation	Degree of freedom	Type I SS	Mean squares	F value	P > F
Treatment	4	804.67	201.17	14.03	0.0004
Parameter	Estimate	T for HO:	Pr > T	Std Error of Estimate	
1/2Os + 1/2Cm	2.17	0.81	0.44	2.68	
3/4Os + 1/4Cm	-0.75	-0.27	0.79	2.79	
1/4Os + 3/4Cm	2.75	0.99	0.35	2.79	

Week 6

Source of variation	Degree of freedom	Type I SS	Mean squares	F value	P > F
Treatment	4	2288.67	572.17	13.47	0.0005
Parameter	Estimate	T for HO:	Pr > T	Std Error of Estimate	
1/2Os + 1/2Cm	-4.83	-1.05	0.32	4.61	
3/4Os + 1/4Cm	-3.92	-0.82	0.43	4.79	
1/4Os + 3/4Cm	-8.75	-1.82	0.10	4.78	

Week 8

Source of variation	Degree of freedom	Type I SS	Mean squares	F value	P >
Treatment	4	1875.07	468.77	47.51	0.0
Parameter	Estimate	T for HO:	Pr > T	Std Error of Esti	
1/2Os + 1/2Cm	-3.50	-1.58	0.15	2.22	
3/4Os + 1/4Cm	-2.41	-1.05	0.32	2.31	
1/4Os + 3/4Cm	-5.58	-2.42	0.03	2.31	

Week 10

Source of variation	Degree of freedom	Type I SS	Mean squares	F value	P >
Treatment	4	1968.40	492.10	14.25	0.0
Parameter	Estimate	T for HO:	Pr > T	Std Error of Esti	
1/2Os + 1/2Cm	-1.67	-0.40	0.69	4.15	
3/4Os + 1/4Cm	0.00	0.00	1.00	4.32	
1/4Os + 3/4Cm	-2.67	-0.62	0.55	4.32	

Week 12

Source of variation	Degree of freedom	Type I SS	Mean squares	F value	P >
Treatment	4	2655.07	663.77	43.48	0.0
Parameter	Estimate	T for HO:	Pr > T	Std Error of Esti	
1/2Os + 1/2Cm	-0.17	-0.06	0.95	2.76	
3/4Os + 1/4Cm	5.92	2.06	0.06	2.88	
1/4Os + 3/4Cm	-2.58	-0.90	0.39	2.87	