EFFECTS OF ORGANIC MATERIALS AND INORGANIC FERTILIZER ON SOIL MICROBIAL BIOMASS AT KABETE, KENYA

BY

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DECLARATION

This thesis is my original work and has not been presented for degree examination in any other university

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DEDICATION

This work is dedicated to my dear husband Simon Njii Mwangi, children and parents for their support, encouragement and sacrifice. It is also dedicated to my siblings and friends who stood with me and sincerely prayed for me when things were too tough.

To you I say thank you and may the LORD GOD bless you.

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ABSTRACT

Recently, the issue of sustainable agro ecosystem production has been of great concern. This has led to the need of understanding and developing management strategies, which conserve and protect the soil resources. Micro biota is one of the critical agents in relation to soil fertility and plant growth, due to among other reasons; its participation in nutrients cycling and in the formation and stabilization of soil aggregates. In order to develop sustainable and productive agroecosystems, there is therefore need to understand the dynamics of soil microbial biomass. This study was designed to evaluate the influence of organic inputs, inorganic fertilizers and their combinations on soil microbial biomass. The experiment was set-up at Kabete, Kenya, which is an on going trial established in 1999 (Kabete Nitrogen Management N1). It involved use of different combinations of *Tithonia diversifolia, Senna spectabilis, Calliandra calothyrsus* as the organic nutrient resources and urea as the nitrogen source. The experimental design was a complete randomized block design with 10 treatments each replicated 4 times. Soil Samples were collected to a depth 0-10 cm for two maize cropping seasons and this was done before incorporating the inputs and every two months thereafter within the season and also at harvesting time. The soils were analyzed for microbial carbon and nitrogen content using the chloroform fumigation extraction and incubation methods at ICRAF laboratory. Plant samples (maize ears and stovers) were also collected for yield data. Analysis of variance (ANOVA) was conducted on the data and means separated using LSD at 5% significance difference using Genstat for Windows Version 6. All treatments gave higher soil microbial biomass N (Nmic) than the control treatment throughout the two maize cropping seasons. Further, organic+urea treatments resulted in higher values of Nmic than sole organic treatments. Microbial biomass carbon (Cmic) in soils treated with organic and organic+urea inputs was higher than either control or urea treatments in both seasons. Treatments receiving organic+urea inputs gave higher values of Cmic than their corresponding sole organic treatments. Plant growth seemed to stimulate Cmic growth, as there was an increase in Cmic at the peak of plant growth. Moreover, Cmic was found to be positively correlated with total organic carbon and maize yield. Soil respiration in the control and urea treatments was lower than all the other treatments in both seasons and was found to be significantly lower than sole tithonia and tithonia+urea in the month of October 2003. Further, organic+urea treatments gave higher soil respiration than their corresponding sole organic treatments though the difference was not significant. A decrease in basal respiration was noted after addition of organic materials indicating a less stressed microbial community. The results of this study indicated that application of sole organic materials increased soil microbial biomass. However, organic+urea treatments did increase microbial biomass above sole organic treatments, suggesting that combining organic materials and urea is a better option for optimum soil production. Therefore, the advocacy should be to add organic resources where finances are limiting for the purchase of urea, otherwise combining of organic and inorganic inputs would be ideal for a productive and sustainable ecosystem.

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ABBREVIATION

C	Carbon
Ca	Calcium
Calliandra	*Calliandra calothyrsus*
CFE	Chloroform Fumigation Extraction
CFI	Chloroform Fumigation Incubation
CIAT	Centre for International Tropical Agriculture
Cmic	Soil microbial biomass carbon
EA	Exchangeable acidity
ICRAF	International Centre for Research in Agroforestry
K	Potassium
K_2SO_4	Potassium sulphate
KARI	Kenya Agriculture Research Institute
KCL	Potassium Chloride
LRS	Long rain season
Mg	Magnesium
N	Nitrogen
N1	Nitrogen Management I
NARL	National Agriculture Research Laboratory

NH^{+}_{4}	Ammonium
Nmic	Soil microbial biomass nitrogen
NO^{-}_{3}	Nitrate
Senna	*Senna spectabilis*
SMB	Soil Microbial Biomass
SOM	Soil Organic Matter
Tithonia	*Tithonia diversifolia*
SRS	Short rain season
TSBF	Tropical Soil Biology and Fertility

CHAPTER ONE: INTRODUCTION

1.1 Background

Microbes constitute about one quarter of all living biomass on earth and are responsible for significant nutrient transformations involving both macro and micro nutrients (Alexander, 1994) and therefore influence nutrient availability and ultimately soil health and quality. They are the main driving force in the decomposition of organic materials, and frequently used as an early indicator of changes in soil chemical and physical properties resulting from soil management and environment stresses in agricultural ecosystems (Transar-cepeda et al., 1998; Brookes, 1995; Jordan et al., 1995). Though 1-3% of total soil C is microbial biomass C and 5% total soil N is soil microbial biomass N, they are the most labile pools in soils (Jenkison and Ladd, 1981) and therefore, nutrient availability and productivity of agro ecosystems mainly depends on the size and activity of the microbial biomass (Friedel et al., 1996).

The turnover of soil microbial biomass is a dynamic process, and responds relatively quickly to changes in environmental conditions, i.e., climate, input of nutrients, and disturbance. Determination of soil microbial biomass can therefore provide estimates of the net flux of carbon, nitrogen, phorsphorous and sulfur through microbial pools and thus reflect the contribution of soil microorganisms as both a source and a sink of carbon and nitrogen in soil ecosystems. Authors have reported the identification of biological indicators of soil quality as critically important (Elliott et al., 1996; Doran and Parkin, 1994), and the rationale for the use of microbial and biochemical parameters as soil fertility indicators is their central role in the cycling of C and N (Visser and Parkinson, 1992) and their sensitivity to change (Brookes,

1995). Because soil microorganisms carry out many below-ground processes, estimates of microbial biomass may be useful for comparisons of ecosystems with similar soil management practices and land use histories.

Addition of organic wastes into the soil has become a widespread soil management practice, due to the fact that they are a source of nutrients (Perucci et al., 2000), and therefore the effects of such inputs on soil microbial biomass (SMB) should be taken into account. However, a few studies have addressed the effects of different factors on the microbial community than the effects of abiotic factors (Sarrantonio, 2003), and therefore the roles of macro and micro organisms in soil productivity, especially transformations and availability of nutrients remain to be fully understood (Zhenli et al., 2003). This study was therefore set to evaluate the effects of organic and inorganic resources on soil microbial biomass carbon and nitrogen.

1.2 Problem statement

The danger of losing land productivity due to changes in the soil created disturbances is paramount. The most significant of these changes are occurring from deforestation, desertification and agricultural production, which lead to biological, chemical and physical instability in soils because of massive loss of C and altered nutrient cycling (Perucci et al., 2000). While nutrients cycles tend to be less "leaky" in undisturbed ecosystems, an important characteristic of agroecosystems is that they export large inputs in the crop biomass. This calls for residue addition to promote stable supply of C and energy for organisms, which in turn, will affect nutrient cycling. One of the drawbacks is the choice of appropriate management

options, which will provide enough energy to sustain adequate level of microorganisms activities. Although soil microbial biomass carbon and nitrogen parameters reflect the soil's ability to recycle nutrients and energy, and to buffer external changes (He, 1997), there are no established benchmarks, critical or threshold values against which soil quality can be evaluated (He et al., 2003). Therefore, this study aimed at quantifying the effect of organic materials of differing quality and inorganic fertilizer on soil microbial biomass over time.

1.3 Research questions

The study sought to answer the following questions:

1. Are there any differences in soil microbial biomass resulting from application of organic materials?
2. How does the soil microbial biomass between the organically and inorganically treated soils compare?
3. What effects do combinations of organic and inorganic inputs have on soil microbial biomass?
4. Is soil microbial biomass determined by chloroform fumigation extraction and chloroform fumigation incubation equal?

1.4 Hypotheses

The working hypothesis was: ''soil microbial biomass can be influenced differently by addition of organic and inorganic inputs'': The study aimed at testing the following specific hypotheses:

1. Application of organic materials will increase soil microbial biomass.
2. Application of inorganic fertilizer can lead to a decrease soil microbial biomass.

3. Combining organic materials and inorganic fertilizers will decrease soil microbial biomass.

4. There is no difference between soil microbial biomass determined by chloroform fumigation extraction and chloroform fumigation incubation methods

1.5 Objectives of the study

The main objective of the study was to investigate the effects of organic and inorganic inputs and their combinations on SMB.

The specific objectives were:

1. To evaluate the effects of organic inputs (*Tithonia diversifolia, Senna spectabilis* and *Calliandra calothyrsus)* on soil microbial biomass.

2. To compare the effects of organic and inorganic inputs on soil microbial biomass.

3. To investigate the effects of combined use of organic materials and inorganic fertilizers on soil microbial biomass.

4. To compare results from the chloroform fumigation extraction and chloroform fumigation incubation methods for estimation of soil microbial biomass.

1.6 Justification and significance of the study

Increasing concern about the long-term productivity of agro ecosystems has emphasized on the need to develop management strategies that maintain and protect soil resources. The issue is directly related to maintaining the quantity and quality of soil organic matter (SOM), which is a critical component of soil productivity. Soil microbial biomass is considered an important and labile fraction of SOM involved in energy and nutrient cycling and therefore its size and turnover rates are very promising indicators of soil quality. Friedel (1996) indicated that

nutrient availability and productivity of agroecosystems mainly depend on the size and activity of the microbial biomass. Therefore, an understanding of the mechanism controlling the composition and function of microbial communities, and their population dynamics in soil is important.

The data on SMB will lead to the possibility of enhancing the growth and activity of desirable microorganisms or suppressing the undesirable ones (Kunc, 1994), and quantifying external factors that may influence them (Ritz et al., 1994). The results will give a direction on the application of appropriate agricultural management practices, which will increase land productivity. The findings will also form a meaningful tool necessary for studying the flux of energy and material through soil population, contribution of biomass to global cycles of carbon, nitrogen, phosphorous, sulphur and other elements, turnover of biomass and SOM and the role of microbes as a reservoir of nutrients (Kunc, 1994). Also important is that the extension personnel and farmers can use the results to devise practical measures of soil quality.

CHAPTER TWO: REVIEW OF LITERATURE

2.1 The importance of soil microbial biomass

The soil microbial biomass (SMB) accounts for only 1-3% of soil organic C but is the "eye of the needle" through which all-organic materials that enter the soil must pass (Jenkinson, 1977). Thus, it is viewed as an entity and regarded as a transformation station, whereby materials are taken up, converted into new products and subsequently released actively or passively. Under suitable environmental conditions the extent of the turnover will mainly be controlled by the size and activity of the microbial biomass. In order to elucidate intricate relationship and controlling mechanisms of input/output fluxes of nutrients and energy in the soil ecosystems, a reliable quantification of the SMB is required from which valuable information on biomass growth, turnover time, death rate and the efficiency of C use can be derived.

Soil microorganisms are the primary driving forces for many chemical and biochemical processes and thus affect nutrient cycling, soil fertility, and global carbon change (He et al., 2003). They play a major role as a catalyst in the decomposition of soil organic matter (SOM) and release of inorganic nutrients to the bulk soil where they become available for plant uptake (Smith, 1994). Microbial turnover rates of C, N and P are defined as the time required for synthesizing or decomposing the amount of microbial C, N, or P equivalent to original "stand crop" at steady state and this time has been found to be related to the size of the SMB, land use and management, and soil texture (Chen et al., 2002a, b; Yao and He, 1998). The turn over of microbial C, N, or P can therefore be used to estimate the annual C, N, or P flux in the soil (Chen et al., 2002a, b; Yao and He et al., 1999; Brookes et al., 1984) and the calculated annual fluxes may provide a rough estimate of N, and P available to plants.

2.2 Organic residues and microbial activity

Residues can have a significant effect on SOM, SMB levels, and decomposition rates and nutrients dynamics. Though the presence of adequate food for microorganisms boosts their numbers and contributes more to mineralization (Sande et al. 2001), there is need to consider quality of organic residues for soil fertility and yield improvements (Ayuke, 1999). Extensive literature on the importance of the quality of organic inputs on the dynamics (C and N) of soil organic matter is available. For example, Palm (1995) reported that organic materials from most multipurpose trees (e.g. *Leucaena leucocephala, Senna spectabilis*, *Inga edulis* and *Erythrina poeppigiana*) are good sources of nutrients especially nitrogen. Mugendi et al. (1999) reported that the highest amounts of mineralized N were obtained only at four weeks after planting maize with some species like *Calliandra calothyrsus* and *Leucaena leucocephala*. However, literature on how such resources affect SMB is scarce.

In order to evaluate C storage in alternative systems, multiple factors have to be interpreted. These factors include microbial biomass which plays a role in C cycling and partitioning as well as being a dynamic soil constituent and thus, it is thought that a single linear hypothesis rarely seems to explain complex interactions in soil ecosystems (Bell et al., 2003). However, little is understood on the effect of organic fertilization on microorganisms despite being the key component in the release of the nutrients from the organic materials.

2.3 Pool sizes and turnover of microbial biomass in soils

Soil Microbial Biomass has been the subject of intensive research at least in arable, aerated soils. It constitutes both a labile sink and source of plant nutrients that is easily remobilized (Inubushi et al., 1997a, b; Kieft et al., 1987). Given the small size of easily remineralizable N pool, it seems almost inevitable to consider SMB, as the most labile source and sink of N in soil, as well as a primary pool of soil N (Kirik and Olk, 2000). The relative size of SMB pool in relation to total organic C content of the soil is affected by soil and crop management (Kirik and Olk, 2000). For example, Witter and Kanal (1999) found that pool size increases with the organic C content in soil, while soils from long-term experiments of the temperate zone gave significantly higher ratios of microbial biomass C to total organic C in continuous rotation crops versus continuous monoculture systems (Anderson and Domsch, 1989). On the other hand, Mendes et al. (1999) found that microbial responses to cover cropping not only depended on aggregate size, but also on the type of winter cover crop. Distribution of Rhizobium serotypes among aggregates was also found to be affected by different crop treatments (Mendes and Bottomley, 1998).

Such results indicate that soil microorganisms and their activities can be influenced in as-of-yet unpredictable ways because of the complex interaction among environmental factors, substrate quality and time that occur in aggregate micro-habitats. Since the microbial community or functional changes in micro-aggregate in response to management or time will affect significantly the overall activity of the whole soil, understanding the heterogeneity of soil biological properties contributed by micro-habitats will be beneficial particularly when the distribution of the soil aggregate sizes is altered by alternative management practices or soil degradation (Schutter and Dick, 2002).

2.4 Microbial cycling and plant N uptake

Nitrogen cycling through the SMB has a major influence on plant productivity and ecosystem development. While distribution of N in the soil system is largely a function of microbial biomass, size and activity, soil microorganisms potentially compete with plant roots for N when available organic substrate has low N content (Korsaeth, 2001; Kaye and Hart, 1997; Schimel et al., 1989; Jackson et al., 1989). The competition presents potential implications both for plant productivity and for microbial C and N transformations. However, there seems to be evidence that microbial N release can be synchronized to coincide with the plant need for N at least in some agricultural areas, but the question remains how to exactly do this to make agriculture more efficient and ecologically sound (Hartfield and Stewart, 1993).

2.5 Microbial biomass and crop productivity

The productivity and stability of soil as a medium for plant growth depend greatly on the balance between living and nonliving components. Soil microbes perform many functions that are important for field crop production (e.g., nutrient cycling and soil aggregate formation). Energy from the sun and nutrients essential for growth stored in fabric of crop plants, are recovered for reuse through decomposition activities of micro and macro organisms in soil (Doran et al. 1987). The soil organic matter formed during this process serves both as a continuous nutrient supply and a factor stabilizing soil physical environment.

To maintain productivity, soluble nutrients removed from soil through plant growth and harvest must be replaced, either as fertilizers or through biological decomposition of organic and plant matter in the soil (Doran et al. 1987). In management systems in which synthetic chemical use is reduced or eliminated, it follows that the action of soil microorganisms becomes a major

determinant of nutrient cycling and plant growth (Doran et al. 1987). The importance of organic agricultural management lies in the recycling of nutrients for crop production and the critical effect of proper balance between organic matter, soil organisms, and plant diversity on productivity of soils. A basic importance of organic farming is that an ecologically balanced soil environment results in healthy, vigorous plant and stable soil environment (U.S. Dept. of Agriculture, 1980). However, few studies have compared the effects of organic and inorganic inputs on soil microbial properties and crop yield.

2.6 Microbial activity and N mineralization

The soil microbial biomass and its activity are governed by the amount and metabolic availability of C, which for SMB is the most limiting element in most ecosystems. The inorganic N pool fluctuates with mineralization/immobilization processes and it has been hypothesized that the SMB is usually in a steady state, fluctuating only during periods when there are large C inputs added into the soils (Smith and Paul, 1990; Smith and Paul, 1986). If the C pool is not readily metabolized by the SMB, then N mineralization will be slow regardless of the C/N ratio of the substrate, and the concept of the substrate quality is therefore introduced as the controlling factor on microbial growth and N mineralization.

Different plant residues decomposing in the same environment may show significant differences in the rates of breakdown and nutrient release (Swift et al., 1994) and these differences can be explained by regulation of microbial activities by factors such as variation in N, lignin and polyphenol contents (Mafongoya et al., 1996; Tian and Brussaud, 1992a). For example, the low level of polyphenol content in tithonia (2.2%) leads to faster decomposition rate than either senna (2.6%) or calliandra (7.7%) and subsequently releasing higher N (Gachengo et al., 1999) as

compared to senna and calliandra (Lehmann et al., 1995). Although organic materials are well known for their value as sources of nutrients, little information is available on whether the released nutrients are as effective to plants and microbes as the inorganic inputs.

2.7 Carbon dioxide evolution

Carbon dioxide evolution from soils has been measured by ecologists for more than a century (Pettenkofer, 1871). The processes controlling soil carbon (C) cycling are of particular interest because, on a global basis, soils contain twice as much C as the atmosphere (Adams et al. 1990). Monitoring the dynamic of soil C is key to managing soil organic matter to enhance soil quality and its ecological functions, and reduce the impact of agricultural soils on the global warming (Campbell et.al., 1999). Agricultural systems contribute to carbon emissions through the direct use of fossil fuels in food production, the indirect use of embodied energy in inputs that are energy-intensive to manufacture, the cultivation of soils and/or soil erosion resulting in the loss of soil organic matter. The direct effects of land use change (including forest loss and conversion of use) have led to net emission of 1.7 Gt C yr^{-1} in the 1980s and 1.6 Gt C yr^{-1} in the 1990s (IPCC, 2000). *In situ* soil respiration (CO_2 evolution) is a useful measure of relative biological activity (microbial, roots and fauna) of contrasting sites or contrasting treatments applied to the same site (Weber, 1990; 1985; Schlentner and Van Cleve, 1985; Leith and Ouellette, 1962). However, agriculture can also sequester carbon when organic matter accumulates in the soil or aboveground woody biomass acts as a permanent sink or is used as energy source that substitute for fossil fuels. The latest empirical data on agricultural carbon emissions and carbon sequestration opportunities in agricultural systems are reviewed and the necessary land use and management practices that will need to be employed to optimize carbon sequestration are considered. In fact, annual carbon

balances are considered a principal concern for measurement of net sources or sinks of carbon and therefore this study shall shed light on how organic and inorganic inputs affect carbon dioxide evolution.

2.8 Chloroform fumigation extraction and chloroform fumigation incubation methods of measuring soil microbial biomass

As early as 1908, Stormer described and interpreted the effects of biocidal fumigants on soils (Stomer, 1908).

He postulated that: -

- The observed effects of improved plant growth after a transient treatment of soils with toxic fumigants are caused by a liberation of additional N. This N originates from the bodies of the organisms killed by the toxicant, and
- After treatment of the soils, an increased proliferation of bacteria can be observed, which degrade the killed organisms and liberate the N fixed in the cell mass.

The explanation for this observed phenomenon though accepted now as correct did not find the general acceptance it deserved and was overlain by other explanations.

It took 10 years from 1966 for chloroform fumigation incubation (CFI) method of quantifying soil microbial biomass carbon (Cmic) to be presented (Jenkinson and Powlson, 1976a, b; Jenkinson, 1976; Powlson and Jenkinson, 1976). The method has been widely used for measuring Cmic in soils of different types or under different management practices. However, this method is subject to limitations with soils at pH values below 5. Low pH impedes development of bacterial populations in soil and thus results in invalid constant k_C (a constant factor used to convert C

flushes into Cmic) because of reduced mineralization of the killed microorganisms (Vance et al., 1987a).

The chloroform fumigation extraction (CFE) was an improvement on CFI (Wu et al., 1990; Vance et al., 1987b) and has the following advantages over CFI:

(1) Applicability to a wide range of soil types, including acidic soils (pH below 5) (Martikainen and Palojarvi, 1990; Vance, 1987b).

(2) Versatile use in soils with newly added substrate and for submerged soils (Inubushi et al., 1991; Bremer and Van Kessel, 1990).

(3) Determination of C flush from fumigated soils is more accurate and convenient with the automated analyzer (Wu et al., 1990), where the facility is available.

However, re-calibration of the k_C, which is used to convert C flushes to Cmic is essential for different types of soils (Ross, 1990). The study aimed at finding out whether CFE which is easier and faster method may be used in place of the traditional CFI method.

2.9 Comparison of methods of measuring soil microbial biomass

Several alternative and complementary methods for measuring soil microbial biomass have been described and all are subject to different interpretations and require careful standardization for specific soils. Among the biochemical and physiological methods developed for estimating soil microbial biomass are chloroform fumigation-incubation (CFI), chloroform fumigation-extraction (CFE) and substrate induced respiration (SIR). These methods have been widely used to evaluate the influence of agricultural practices, land use and contaminants on soil microorganisms and nutrient cycling (Giller et al., 1998; Dalal, 1998); Martens, 1995). Chloroform fumigation

incubation (CFI) being one of the first methods developed, has been used as a baseline for correlations, but CFE is gaining acceptance because of its greater simplicity and lack of problems with interpretation of the control treatment. However, both the CFI and CFE assay components of microbial biomass are necessary in the interpretation of nutrient-cycling processes, soil organic matter, cultural practices, and inputs associated with agronomic and natural systems (Weaver et al., 1994). The study aimed at finding out whether soil microbial biomass measured using CFE method is equal to that determined by CFI method.

2.10 Gaps in literature

Agroforestry farming which can be defined as all practices that involve a close association of tress or shrubs with crops, animals and/or pasture has gained popularity in the recent past due to its multipurpose benefits (e.g. soil improvement, maintenance of favorable microclimate and animal feeds) (Rocheleau et.al. 1988). The greatest potential of the agroforestry practices lies in the provision of diverse and abundant organic resources to benefit soil microorganisms, which when added to the soil are important in priming of microbial biomass and may have a long term effects on C and N stocks. If agroforestry is to serve people's needs in a variety of rural settings, it is important to see it as an approach to land use, rather than as a fixed arrangement of plants or a particular combination of species. However, a brief review of the full range of agroforestry practices will provide a basis for thinking about agroforestry systems that could usefully be introduced in different environments and especially with special attention to effects of litters of different quality on soil microbial biomass for maximum soil productivity.

CHAPTER THREE: RESEARCH METHODS

3.1 Site Description

This study was carried out at the Kenya Agricultural Research Institute (KARI), Kabete which is part of the on-going work established in 1999 by the Tropical Soil Biology and Fertility Institute of CIAT (TSBF) in conjunction with Kenya Agricultural Research Institute (KARI) and Kenyatta University on Nitrogen Management Trial (N1). The station is located at 36° 46' E and 01° 15' S and at an altitude of 1650 m above sea level. It is in a semi-humid climatic zone with a total bi-modal rainfall of 937 mm per year received in two distinct rainy seasons; the long rains (LR) (mid March to June) and short rains (SR) (mid October to December). The soils are trachyte geological material typically Humic Nitisols (according to FAO-UNESCO, 1990), deep and well weathered and with moderate amounts of C, Ca, Mg, and K but low in available N and P (Kimetu, 2002). The original objective of the study was to determine nitrogen fertilizer equivalency values of *Tithonia diversifolia* (tithonia), *Calliandra calothrysus* (calliandra) and *Senna spectabilis* (senna) and the investigation of nitrogen use efficiency from combined organic and inorganic inputs (Kimetu, 2002).

3.2 Experimental treatments

The experiment was established in 1999 as a Randomized Complete Block Design (RCBD) with ten treatments replicated four times. The current study considered eight treatments for the evaluation of soil microbial biomass under the influence of organic and inorganic resources as indicated in Table 1.

Table 1: Selected experimental treatments from N1 experiment at Kabete, Kenya

Treatment	Quantity of organic nutrient (N) applied (kg N ha^{-1})	Quantity of inorganic nutrient (N) applied (kg N ha^{-1})
Control	0	0
Sole tithonia	60	0
Tithonia + urea	30	30
Sole urea	0	60
Sole senna	60	0
Senna + urea	30	30
Sole calliandra	60	0
Calliandra + urea	30	30

The nitrogen application was at the recommended rate of 60 kg N ha^{-1} in maize and this was applied for all the treatments except the control (0 inputs). The quantity of organic materials applied was calculated on dry matter basis to give N at 60 kg N ha^{-1} and 30 kg N ha^{-1} for sole organic and organic+urea application, respectively.

3.3 Selection of plant material

The choice of the three organic materials was on the basis of their contrasting amounts of lignin and polyphenols as shown in Table 2 and their decomposition rate that had been observed to follow the sequence *Tithonia> Senna> Calliandra* (Palm et al., 2001).

Table 2: Chemical properties for the organic materials used in N1 Kabete

% Content	Tithonia	Senna	Calliandra
Nitrogen	4.4	3.4	2.7
Phosphorous	0.5	0.2	0.1
Polyphenols	2.2	2.6	7.7
Lignin	7.4	10.8	16.0

(Source: Kimetu, 2002)

3.4 Land preparation, inputs application and planting

Land+tilling was done before planting using hand hoes to a depth of about 10 cm in both seasons of the year. The inputs, which were freshly picked leaves of the organic resources and urea as the inorganic source, were applied each season by broadcasting, and incorporating prior to planting. Urea was applied according to farmer's practice of two splits (i.e. 20 kg ha^{-1} at planting and applied 40 kg ha^{-1} 5 weeks after planting). Maize (hybrid 513) was planted at a spacing of 0.75 m by 0.25 m between and within rows, respectively, in each of the 8 plots measuring 5.25 m by 5 m. Since organic materials have been shown to supply other nutrients like potassium (K) and phosphorous (P) that ultimately affect maize yields (Jama et al. 2000), both were applied in all the

plots during the two seasons in non-limiting quantities (i.e. 100 kg P ha^{-1} as triple super phosphate (TSP) and 100 kg K ha^{-1} as muriate of potash (MOP)). It was therefore assumed that nitrogen (N) was the only macronutrient limiting maize yields.

3.5 Soil sampling

Soil samples were collected at planting (before incorporating materials) and every two months thereafter within the maize cropping season and also at harvest. The samples were collected from 5 cores (diagonally and at the center) to a depth of 0-10 cm since this is where most impact of the added organic materials is felt as well as where most maize roots are found. Soil was then composited together and mixed thoroughly while removing all visible plant debris and soil fauna to ensure homogeneity after which it was put in a polythene paper and taken to ICRAF laboratory and stored at 4^{o} C prior to analysis.

3.6 Maize harvesting

Maize was harvested at maturity stage (dry ears) from the standing crop and separated as cob and stover. One guard row on each side and two outer plants on each row were left out unharvested to eliminate edge effects. The harvest (i.e. 6 ears and 6 stovers) was sorted and classified according to size as small, medium and large. From each class, a subsample of six maize plants (ears and stovers) was picked and the fresh weight recorded. The subsamples (ears and stovers) were then oven dried at 60^{o} C until a constant weight was attained. Hand threshing of maize grains was done and their weights recorded for yield analysis.

3.7 Laboratory procedures and analysis

Soil and plant analysis was done at ICRAF laboratory as follows.

3.7.1 Soil moisture determination

Prior to any chemical analysis, field soil moisture content was determined by drying about 30 g of soil sub sample at 105° C for 24 hours. The moisture content was calculated using the following formula:

$$\% \text{ Moisture content} = \frac{\text{Sample fresh weight} - \text{Sample dry weight} \times 100\%}{\text{Sample dry weight}}$$

3.7.2 Pre-incubation preparation

In order to activate soil microorganisms, about 200 g of soil sub samples were weighed and the water holding capacity adjusted to 45% field capacity. The soil sub samples were then incubated at 25° C for 7 days in the dark to permit uniform rewetting and to allow microbial activity to equilibrate after initial disturbance.

3.7.3 Laboratory fumigation

Fumigation for both chloroform fumigation and extraction (CFE) and chloroform fumigation and incubation (CFI) methods was carried out according to the procedure of Jenkison and Powlson (1997a, b). This was done by weighing in duplicates 25 g of previously pre-incubated soil into a 50 ml beaker. The samples were placed in two separate desiccators, one with a 100 ml beaker containing 25 ml of alcohol-free chloroform and other free from chloroform to serve as non-fumigated control. The aim of the alcohol-free chloroform was to kill the soil microbes while the

free alcohol would eliminate the possibility any other source of C apart from the lysed microbes in the soil. The desiccators were tightly closed after adding some chips to assist in volatilization of the chloroform and stored under dark conditions for 24 hours at room temperature. The samples were then removed after evacuating the desiccators repeatedly using a vacuum pump. Thereafter CFE and CFI methods were carried out to determine soil microbial biomass carbon and nitrogen.

3.7.4 Soil extraction for chloroform fumigation extraction method

Chloroform fumigation extraction method (CFE) was done by transferring 25 g (dry weight basis) of the fumigated and non-fumigated (control) soil in Section 3.6.3 into 125 ml shaking bottles and adding 10 ml of 0.5*M* K_2SO_4. The samples were then placed on a shaker and agitated for an hour at 150 reciprocations per minute and subsequently filtered gravimetrically using pre-washed Whatman No. 5 filter paper to give an extract for microbial C and N determination.

3.7.5 Determination of total soluble carbon and soil microbial biomass carbon by chloroform fumigation extraction method

Determination of soil microbial biomass carbon (Cmic) by CFE was done by adding 2.0 ml of 0.16*M* $K_4S_2O_8$ and 10 ml of concentrated H_2SO_4 into 10 ml of the sample extract obtained in section 3.6.4. The resultant contents were mixed using a vortex mixer and then heated in the digestion block for 30 minutes at 150° C. Total soluble C (soil available carbon) was determined colorimetrically at 600 nm and was calciluated as: Total soluble C= Carbon $_{\text{non-fumigated}}$ whereas soil microbial biomass carbon (Cmic) was calculated as:

Microbial biomass C (Cmic) = Microbial biomass $C_{\text{fumigated}}$ – Microbial biomass$C_{\text{non-fumigated}}$

3.7.6 Determination of soil microbial biomass nitrogen by chloroform fumigation extraction method

Determination of soil microbial biomass nitrogen by CFE was done by adding boric acid as the oxidizing reagent to 10 ml sample of soil extracted in section 3.6.4. Both organic-N and ammonium-N was oxidized to nitrate and total N was then determined by adding 1.0 ml of 5% salicylic acid and 10 ml of 16% NaOH while mixing. After cooling, nitrogen was colorimentically read at 410 nm and soil microbial biomass nitrogen (Nmic) was calculated as

Microbial biomass N (Nmic) = Microbial biomass $N_{fumigated}$ – Microbial biomass $N_{non\text{-}fumigated}$

3.7.7 Chloroform fumigation incubation method (CFI)

In chloroform fumigation incubation method, 25 g (dry weight basis) of the previously pre-incubated soil in Section 3.6.2 was fumigated as described in 3.6.3 to kill and lyse microbial cells. A separate set of samples was left un-fumigated to act as a control. Thereafter, the samples were inoculated with 1 g of fresh soil and sealed in a jar. The microorganisms from the fresh soil were expected to grow vigorously using the killed cells of the microbes as substrate. The inoculated samples were placed in 250 ml gas jar together with 10 ml of 1*M* NaOH in a small glass vial before sealing with plastic lids. The jars were incubated at 25° C for 10 days in a dark room. Three 250 ml jars without soil were also incubated similarly to act as blanks. The flushes of CO_2 and extractable N during a 10-day incubation period were assumed to be directly proportional to the amount of C and N in the microbial biomass of the original sample.

3.7.8 Determination of soil inorganic nitrogen (nitrate and ammonium-N)

Analysis of nitrate and ammonium ions in soils was done as described in ICRAF (1995) and by Anderson and Ingram (1993). Ten grams of previously incubated soil in section 3.6.7 was weighed and extracted with 50 ml of 2*N* KCl by shaking for 1 hour at 150 reciprocations per minute and filtered using pre-washed Whatman No. 5 filter paper. Ammonium ions were determined by salicylate-hypochlorite and colorimetrically read at 655 nm while nitrate was determined by cadmium reduction method and read at 525 nm absorbance/concentration.

3.7.9 Calculation of soil microbial biomass nitrogen by chloroform fumigation incubation method

Soil microbial biomass N was calculated as follows:

Microbial biomass N = $(F_N - UF_N)/k_c$ (Horwath et al. 1994)

Where:

$F_N = NO_3 + NH_4$ flush from the fumigated

$UF_N = NO_3 + NH_4$ flush from the non-fumigated

k_c = Constant (Proportion of biomass N mineralized)

3.7.10 Determination of Soil microbial biomass carbon by chloroform fumigation incubation method

Soil microbial biomass carbon was determined by analyzing the CO_2 trapped in 5 ml of NaOH after a 10 days incubation period as described in Section 3.6.7. The CO_2 trapped in 5 ml of NaOH was analyzed by back titration with 1*N* HCl after addition of excess 3*N* $BaCl_2$ as a CO_2

precipitator. The amount of CO_2 respired from fumigated and non-fumigated soil was used to calculate soil microbial biomass as in the equation:

Biomass C = $(F_C - UF_C)/k_c$ (Horwath et al. 1994)

Where:

F_C = CO_2 flush from the fumigated

UF_C = CO_2 flush from the non-fumigated

k_c = Constant (Proportion of biomass N mineralized)

3.7.11 Calculation of basal respiration rate

Basal respiration rate (qCO_2), represents the C flow through microbial biomass; that is, the energy needed to support a given biomass and was calculated by dividing the carbon dioxide evolved with soil microbial biomass carbon in section 3.10.

3.8 Statistical analyses

Data was subjected to analyses of variance (ANOVA) using Genstat for Windows version 6 to determine the effects of the treatments on soil microbial biomass C and N. Least significance differences at a probability of 0.05 was used to determine the significant differences between and within the treatments. Correlation and regression analysis was carried out in order to establish the relationships between microbial biomass and the measured variables.

CHAPTER FOUR: RESULTS AND DISCUSSION

4.1 The scope of the results presented

This chapter presents the research findings and discussion on the effect of organic and inorganic amendments on soil microbial biomass studied in the 2003 long and short rain seasons at Kabete, Kenya. The results on soil microbial biomass carbon (Cmic), soil microbial biomass nitrogen (Nmic), N mineralization, soil respiration, basal respiration, their ratios and comparison of methods are presented in this chapter. The analytical data in form of tables and figures are also summarized in this chapter.

4.2 Maize grain yield in 2003 long and short rain season at Kabete, Kenya

Tithonia+urea and sole tithonia treatments gave significantly ($P \leq 0.05$) higher maize yield than all other treatments in 2003 long rain season (LRS). Control and sole calliandra treatments resulted in significantly ($P \leq 0.05$) lower maize yield than all other treatments (Table 3). Combining organic inputs and urea significantly increased maize for calliandra +urea while no difference was observed for all other treatments.

All treatments yielded higher maize yield in the 2003 LRS compared to the 2003 SRS (Table 3). Control and sole calliandra treatments yields were lower than those for other treatments. The control and senna+urea treatments gave the lowest and highest maize yield respectively in this season and the control treatment was found to be significantly ($P \leq 0.05$) lower than all the treatments with respect to maize grain yields.

Unlike in the previous season (2003 LRS) where only calliandra+urea outyielded it's corresponding sole organic treatment, in the 2003 short rain season (SRS), the intergrated treatments had lower yields except for calliandra treatment, which gave significantly higher maize yield for calliandra+urea than sole calliandra treatment.

Table 3: Treatment effects on maize grain yield at Kabete, Kenya

Treatment	2003 LRS	2003 SRS
	Tons/ha	
Control	0.5	2.6
Tithonia+urea	2.7	4.9
Sole tithonia	2.7	5.2
Sole urea	2.0	5.0
Senna+urea	2.0	4.9
Sole senna	2.0	5.3
Calliandra+urea	1.6	4.3
Sole calliandra	0.9	3.7
SED	0.3	0.4

Yields observed in the two seasons for all the treatments were below the average yield (6 to 12 tons ha^{-1}) for this area which can be attributed to low and poor rainfall distribution experienced in the two seasons. However, maize yield from treatment-imposed soils was higher compared to the control treatment indicating the importance of soil amendments for high crop productivity. This could possibly be attributed to the positive effects of the organic materials on soil physical and chemical properties, which may include improved moisture retention and addition of micronutrients (Kimetu, 2004; Murwira et al. 2002; Lehmann et al. 1999; Wallace, 1996). Lower maize yield for calliandra treatments compared to tithonia and senna treatments could be

attributed to N immobilization or reduced N release as was also noted by Mwale et al. (2000a) in their study at Chalimbana, Zambia. Other researchers have also observed that a large portion of N from a slowly decomposing biomass may be incorporated into soil organic matter fractions (Lehmann et al. 1999) or immobilized into forms not readily available to annual crops (Mugendi et al. 1999). As indicated by Palm (1995), nutrient release patterns from organic materials are partly determined by their chemical composition or quality. These results therefore suggest that tithonia and senna green biomass may be recommended for direct application for soil fertility improvement (Kimetu, 2004) while callianrda may not be not be recommended due to the high polyphenol content and lignin as shown in Table 2. The maize yield recorded in 2003 LRS was lower than that of 2003 SRS suggesting the effect of other factors on maize yield. For example, rainfall distribution and magnitude could have greatly influenced the yield. The long rain season received a total of 440 mm rainfall which was a poorly distributed compared to 750 mm in the 2003 SRS and this could have resulted in reduced nutrient availability to the maize plants (Appendix 1). Further, this could be due to the residue effect of organic and inorganic amendments from the previous season as was also observed by Kimetu et al. (2004) and Obaga et al. (2001).

The combined application of organic resources and mineral N is hypothesized to yield added benefits in terms of extra yield compared to the sum of the responses in the treatments with a sole application of organic resources and mineral N (Vanlauwe at al., 2001). This phenomenon has been observed by many authors, (example, Okalebo et al., 2002; Mucheru et al., 2002 and Nhamo, 2001) and could be explained by the fact that N from an inorganic source is readily available (from both the added urea and increased decomposition of organic materials due to

addition of the same) for plant uptake at an early stage of plant growth as compared to the sole organic inputs (Kimetu, 2002). But the findings of this study did not show a consistent agreement to this hypothesis in the two seasons. In the 2003 LRS, organic+urea and sole organic treatments resulted in equal maize yields except for calliandra+urea treatment that gave higher maize yield than its corresponding sole organic treatment. The increase in maize yield for calliandra+urea can be explained by available N from the added urea to a low quality material and therefore leading to readily available nutrients due to increased decomposition However, in 2003 SRS all treatment receiving sole organic inputs resulted in higher maize yield than their corresponding organic+urea treatments except for calliandra treatment. Further, all organic+urea treatments did not out yield sole urea treatment as hypothesed. In 2003 SRS, only tithonia+urea and senna+urea resulted in higher maize yield than with sole urea treatment while in 2003 SRS all integrated treatments gave lower maize yield than sole urea treatment. The results of this study suggest that other factors (e.g. moisture, temperature) could influence the maize yield even in the events that soil has received inputs.

4.3 Effect of organic, inorganic inputs and their combinations on soil mineral N in the 2003 long and short rains at Kabete

All treatments receiving sole organic and organic+urea inputs recorded higher mineral N than sole urea and control treatments throughout the 2003-LRS except in October-2003 (end of the season) when sole calliandra resulted in lower mineral N than sole urea treatment (Figure 1). However, there was no consistency in the amount of mineral N recorded in the 2003-LRS.

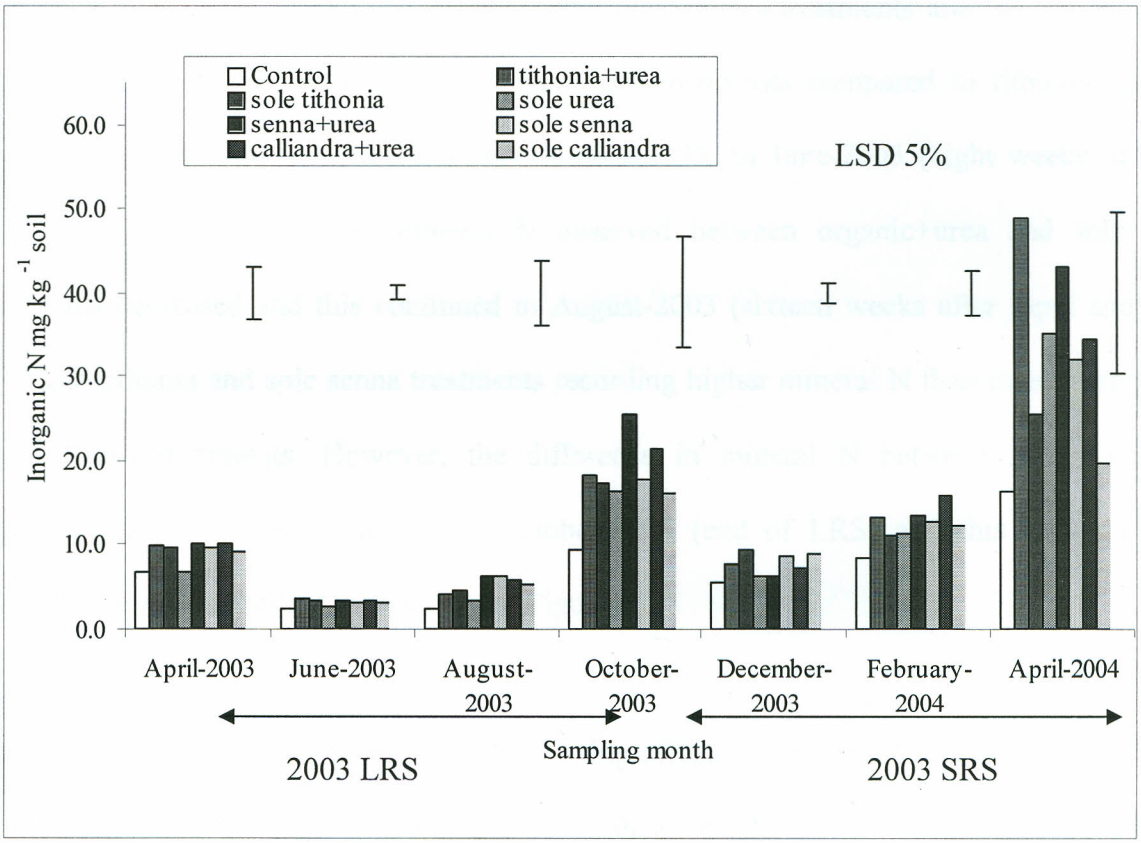

Figure 1: Treatment effects on soil mineral N in 2003 LRS and SRS at Kabete, Kenya

Senna+urea treatment gave higher mineral N content than all other treatments in April-2003 and October-2003. In June-2003 and August-2003, tithonia+urea and senna+urea respectively resulted

in higher mineral N content than all the other treatments, and the latter was found to be significantly ($P \leq 0.05$) higher than the control treatment.

Higher mineral N was observed for organic+urea treatments compared to their corresponding sole organic treatments in 2003-LRS except in August-2003 when sole tithonia and sole senna treatments gave higher mineral N than their organic+urea treatments counterparts (Figure 1). The difference in mineral N observed between the organic+urea treatments and their corresponding sole organic treatments was higher for calliandra treatments compared to tithonia and senna treatments in April-2003, June-2003 and October-2003. In June-2003 (eight weeks after input application) the difference in mineral N observed between organic+urea and sole organic treatments decreased and this continued in August-2003 (sixteen weeks after input application) with sole tithonia and sole senna treatments recording higher mineral N than their corresponding organic+urea treatments. However, the difference in mineral N between sole organic and organic+urea determined increased in October-2003 (end of LRS) and this also marked the highest difference observed in this season (Figure 1).

During the 2003 LRS, the treatments had varying effects on mineral N content within different months. In June-2003 (eight weeks after input application), the content of mineral N recorded was found to be lower than that observed in April-2003 (beginning of LRS). The decrease in mineral N content between months was higher for organic+urea than sole organic treatments. Senna+urea (7.0 mg N kg^{-1}) and sole tithonia (4.1 mg N kg^{-1} soil) treatments recorded highest and lowest decrease in mineral N content between April-2003 and June-2003 respectively. However, an increase in mineral N content was observed thereafter in August-2003 (sixteen weeks after inputs

application) and in October-2003 (end of LRS) (Figure 1). The change in mineral N from one month to another was found to be higher for organic+urea treatments than sole organic treatments except in August-2003 and October-2003 when sole calliandra and sole tithonia resulted in higher increase in mineral N than their corresponding organic+urea treatments respectively.

In 2003-SRS, the control treatment gave lowest mineral N content throughout the season and was found to be significantly lower than all the treatments in February-2004 (Figure 1). In December-2003 (eight weeks after input application), sole tithonia resulted in higher soil mineral N than all the other treatments while calliandra+urea and tithonia+urea gave the highest mineral N in February-2004 and April-2004 respectively. At the end of the 2003-SRS (April-2004), tithonia+urea gave significantly higher mineral N content than the control treatment. The difference in mineral N calculated between organic+urea and sole organic treatments within months in 2003-SRS was similar to that observed during the 2003-LRS. Organic+urea treatments resulted in lower mineral N than their corresponding sole organic treatments in December-2003 (eight weeks after input application) (Figure 1). However, this trend was reversed in February-2004 and April-2004 as organic+urea treatments gave higher mineral N content than their corresponding sole organic treatments (Figure 1). The highest difference observed between organic+urea treatments and their corresponding sole organic treatments was in April-2004 (end of SRS) where tithonia+urea and sole tithonia gave a difference of 23.4 mg kg^{-1} soil (Figure 1).

The trend observed in the changes in soil mineral N during the 2003-SRS was similar to that observed during the 2003-LRS. A decrease in mineral N content was observed in December-2003 (eight weeks after inputs application) but an increase was noted thereafter in February-2004 and

in April-2004 (end of 2003 SRS). The highest mineral N difference between the months was recorded in February-2004 and April-2004. Organic+urea treatments resulted in higher mineral N increases within the season (between months) compared to their corresponding sole organic treatments. Calliandra+urea (8.7 mg N kg^{-1} soil) and senna+urea (29.6 mg N kg^{-1} soil) treatment gave the highest mineral N difference between December-2003, February-2004 and April-2004 respectively.

The results of this study indicate that addition of organic materials and urea contributed to mineralization as the observed mineral N was higher for all the treatments over the control. This could be attributed to readily available mineral N from the organic materials added to the soil. Many authors have observed this phenomenon referred to as the priming effect, which arises either immediately or very shortly after addition of specific substances to the soil (Pascual et al., 1998; Dalenberg and Jager, 1989, 1981; Kudeyarov, 1988). Further, mineral N seemed to be related to the quality of the organic materials. Sole tithonia gave higher mineral N than sole senna and sole calliandra treatments eight weeks after inputs application (June-2003 and December-2003) (Figure 1), which can be attributed to its high decomposition rate and subsequent higher N release (Gachengo et al., 1999; Lehmann et al., 1999). Sole calliandra tended to give lower values of mineral N in the two seasons apart from the months of August-2003, December-2003 and February-2004. This may be attributed to it's polyphenol content, which is more than double the critical value (4.0%) for release of N to occur (Palm et al. 2001; Delve et al., 2000) and hence decomposing relatively slowly compared to the tithonia and senna. The quality of organic residues, as reflected in the C: N ratio has been shown to be of primary importance in regulating the magnitude of the two opposing processes of mineralization and immobilization (Nunan et al.,

2000). Similar results were observed by Mugendi et al. (2000) in a study done at the sub humid highlands of Embu in Kenya.

Organic+urea treatments gave relatively higher mineral N as compared to their corresponding sole organic treatments. The difference in mineral N between the organic+urea and sole organic treatments seemed to be related to the quality of the organic material added. The findings could be attributed to the readily available mineral N from the added urea to the soil. A positive impact on N mineralization has been reported after addition of exogenous source of inorganic N, particularly in the presence of high C: N ratio organic residues (Sakala et al., 2000; Henriksen and Breland, 1999; Recous et al., 1995). The results agree with what Kimetu et al. (2004) reported in a study on fertilizer equivalency values for organic materials of contrasting qualities. The differences in the chemical properties of the organic materials as indicated in Table 2 seemed to influence mineral N content differently. Sole tithonia and tithonia+urea gave relatively lower mineral N differences than senna and calliandra over the two maize cropping seasons. It has been postulated that soil N mineralization is mediated by two distinct microbial populations (opportunistic and generalistic) (Bonde and Rosswall, 1987), and that changes in net N mineralization are due to changes in both amount and quality of the substrate available to the mineralizing biomass (Nunan et al., 2000). However, the difference in mineral observed among the treatments was not significant and this could be attributed to rapid mineralization of the organic residues due to increased aeration as a result of tillage, higher soil temperatures leading to higher decomposition rates, lower litter inputs and the shorter duration of organic residue application (Nandwa, 2001).

Both seasons gave a decrease in mineral N in 2003 and December-2003 (i.e., eight weeks after inputs application). This coincides with the peak demand for mineral N by the plants suggesting that most of the N released could have been taken up by plants and hence the low level of mineral N determined in the soil (Kimetu, 2002). Mineral N recorded at the end of the 2003-SRS was also found to be significantly ($P \leq 0.05$) higher than in 2003-LRS and this could be attributed to seasonal differences in rainfall patterns for the two seasons. The 2003-LRS experienced a high down pour four weeks after planting (see appendix 1) that could have resulted in N leaching and this may explain the lower level of mineral N determined in the soil. The months of April-2003, October-2003 and April 2004 were characterized by increased mineral N that could be associated with the high rainfall and therefore favoring N mineralization. An increase in mineral N was observed in the dry spell between the month of December 2003 and February 2004. Results of a study carried out by Kelting et al. 2002 at North Carolina indicated a generally higher monthly net N mineralization in warm summer months than cool winter months for all treatments suggesting increased temperatures may stimulate net N mineralization. Strong positive relationships between temperature and net N mineralization were also shown by Goncalves and Carlyle (1994) and Sierra (1997),

4.4 Effects of organic, inorganic inputs and their combinations on microbial biomass nitrogen in 2003 long and short rains at Kabete, Kenya

During the 2003-LRS, the control treatment gave the lowest level of soil microbial biomass nitrogen (Nmic) (Figure 2). In April-2003 (beginning of the LRS and before inputs application), all treatments with sole organic inputs resulted in higher levels of Nmic than their corresponding organic+urea treatments with sole calliandra treatment (45.0 mg N kg^{-1} soil) giving the highest (Figure 2). No particular trend in Nmic was observed within the 2003-LRS. In June-2003 (eight weeks after inputs application), tithonia+urea treatment gave the highest Nmic whereas senna+urea treatments gave the highest Nmic in August-2003 and October-2003 (Figure 2). Organic+urea treatments recorded relatively higher levels of Nmic than sole organic treatments throughout the 2003-LRS season apart from the month of June-2003 and October-2003 when calliandra+urea and tithonia+urea respectively gave lower Nmic than their corresponding sole organic treatments (Figure 2). The highest Nmic difference observed between sole organic and organic+urea treatments was in August-2003 with calliandra and tithonia treatments recording highest (4.2 mg N kg^{-1} soil) and lowest (1.9 mg N kg^{-1}) values respectively. However, the difference in Nmic from the two treatments was not significant.

Eight weeks after inputs application (June-2003), a decrease in Nmic was observed and this continued upto August-2003. The decrease in Nmic was higher for sole organic treatments compared to their organic+urea counterparts except in August when sole senna recorded lower Nmic than senna+urea treatment. Sole calliandra (19.4 mg N kg^{-1} soil) and senna+urea (3.2 mg N kg^{-1} soil) respectively gave the highest and lowest decrease in Nmic between April-2003 and June-2003. Between June-2003 and August-2003, sole tithonia (9.4 mg N kg^{-1} soil) and

calliandra+urea (0.6 mg N kg^{-1} soil) treatments respectively gave the highest and lowest decreases in Nmic. However, an increase in Nmic was noted between August-2003 and October-2003 with sole calliandra (24.8 mg N kg^{-1} soil) and tithonia+urea treatments (17 mg N kg^{-1} soil) giving the highest and lowest increase respectively (Figure 2).

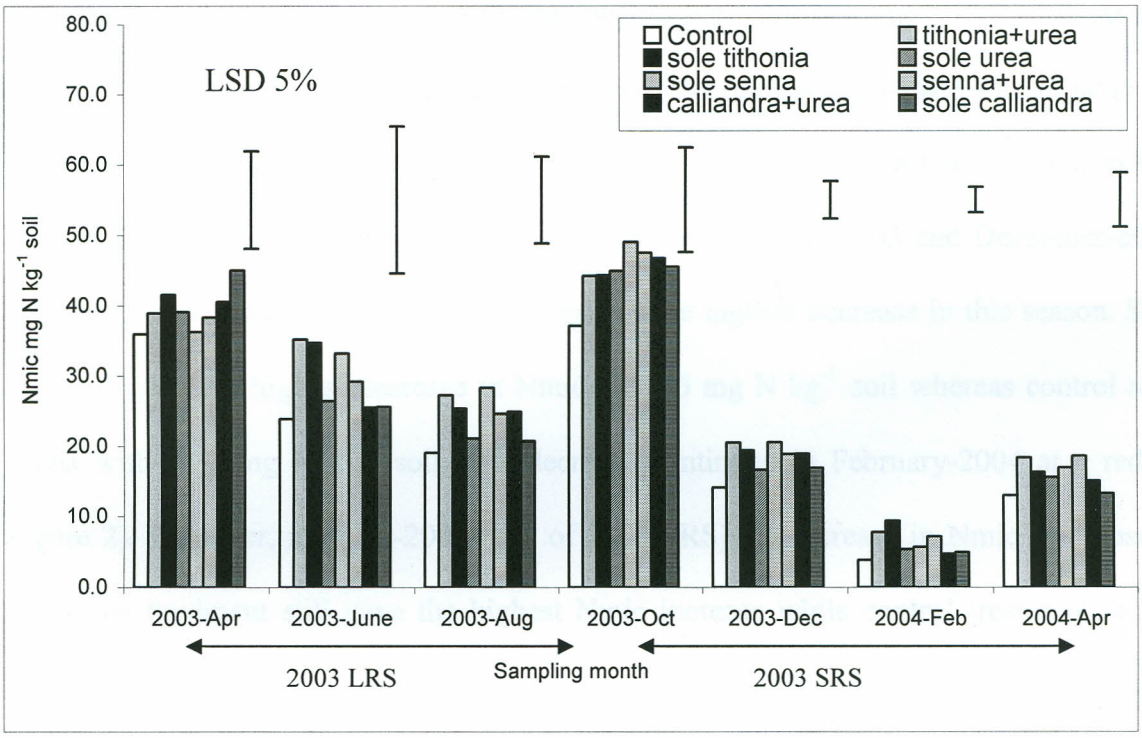

Figure 2: Effect Treatments on soil microbial biomass nitrogen in 2003 LRS and SRS at Kabete, Kenya

There was no consistent trend in the treatment effects on Nmic in 2003-SRS. Senna+urea, sole tithonia and sole senna resulted in highest Nmic in December-2003, February-2004 and April-2004 respectively (Figure 2). The control treatment gave the lowest Nmic throughout the season and was found to be significantly ($P \leq 0.05$) lower in Nmic than tithonia+urea and senna+urea in December-2003 (Figure 2). Organic+urea treatments also recorded higher Nmic than their corresponding sole organic treatments in the same month. The difference in Nmic between sole calliandra and calliandra+urea (2.0 mg N kg^{-1} soil) was higher than that calculated between sole

senna and senna+urea (1.6 mg N kg^{-1} soil) and, sole tithonia and tithonia+urea (1.1 mg N kg^{-1} soil) treatments. However, in February-2004, this trend was reversed as organic+urea treatments gave lower Nmic than their corresponding sole organic treatments (Figure 2). The highest difference in Nmic was observed between sole calliandra and calliandra+urea (2.0 mg N kg^{-1} soil) while the lowest was between sole tithonia and tithonia+urea (0.3 mg N kg^{-1} soil). At the end of the 2003-SRS only senna+urea gave lower Nmic than its corresponding sole organic treatment (Figure 2). The treatment effect on Nmic between months in 2003 SRS was similar to that in the 2003-LRS, a decrease in Nmic was observed between October-2003 and December-2003 (eight weeks after inputs addition) and this also marked the highest decrease in this season. Sole senna treatment gave the highest decrease in Nmic of 28.6 mg N kg^{-1} soil whereas control resulted in lowest with 23.0 mg N kg^{-1} soil. The decrease continued in February-2004 at a reduced rate (Figure 2). However, in April-2004 (end of 2003-SRS), an increase in Nmic was observed and sole senna treatment still gave the highest Nmic increase while control treatment recorded the lowest.

The results of this study indicate that Nmic was increased by addition of inputs as the control treatment gave lower Nmic than all the treatment imposed plots. The increase suggests that the turnover of Nmic is dependent on application of inputs. This could be attributed to stimulated suitable conditions for microbial growth as a result of readily available decomposable material. Similar results were reported by El-gharmry (2001) in a study on the effect of organic residues on soil microbial biomass. The results of this study also indicate that Nmic is dependent on the quality of the organic resources. The tendency of sole tithonia to record a high level of Nmic could be attributed to its high decomposition and high N content and hence releasing mineral N

for microbial growth. The low Nmic observed for sole calliandra treatment could be explained by its low quality (low N content, slow mineralization and N release); suggesting that less mineral N is available for microbial growth.

The results observed for organic+urea treatments disagree with one of the hypothesis of this study that combining organic and inorganic inputs will decrease soil microbial biomass above their corresponding sole organic inputs. Higher Nmic observed in organic+urea treatments could be attributed to readily available N from the urea applied. Application of urea would have reduced the competition for mineral N between the soil microbes and plant. The results concur with what was reported by Wang and Bakken (1997) that improving N availability under N-limited (C-rich) conditions allows increased microbial activity and growth. Similar trend of results were also reported independently by Moore et al. (2000), Omay et al. (1997) and Fraser et al. (1994).

In both seasons however, a decrease in Nmic was noted eight weeks after addition of the inputs, i.e., in June-2003 and December-2003 respectively (Figure 2). This decrease continued in August-2003 and February-2004 (16 weeks after addition of inputs) in 2003 LRS and 2003 SRS respectively. The decrease in Nmic recorded at eight and sixteen weeks after input application could be explained by the peak demand for N by the growing crop, indicating a plant-microbes competition. Competition for mineral N by plants and microbes in the soil has been reported elsewhere (Kaye and Hart, 1997; Schimel et al., 1989). However, a flush in Nmic was noted at the end of each season (April-2003, October-2003 and April-2004) (Figure 2), which could be explained by reduced competition for mineral N as the crop gets to maturity as well are high amount of rainfall that favors microbial growth (Appendix 1). The Nmic observed at the end of

the 2003-LRS was also found to be significantly ($P \leq 0.05$) higher than that obtained at the end of 2003-SRS (Figure 2). The high Nmic in the 2003 LRS could partly be explained by the low N competition for mineral N between the plant and microbes as the crop performed poorly due to low and poor rainfall distribution (Appendix 1). This could mean that more mineral N was available for microbial growth in 2003-SRS. Unless the mineralized N is removed (e.g. by the roots), rapid N uptake by microorganisms under N limited conditions takes place (competition theory) (Hunt et. al. 1997; Cheng, 1999), and this could explain the higher Nmic determined in 2003-LRS.

4.5 Relationship between soil microbial biomass nitrogen and moisture, total nitrogen, maize yield and net-N mineralization

Soil microbial biomass N was found to be positively related to most of the measured variables in this study (Table 4). Moisture was found to positively influence Nmic indicating a high dependency of the microbes on the moisture status of the soil. The positive relationship can be explained by the fact that N is available for microbial utilization in solution as NO_3^-. Conversion of organic N to available mineral forms ($NH_4^+ + NO_3^-$) through biochemical transformations is mediated by microorganisms and is influenced by those factors that affect microbial activity, e.g., temperature, moisture, pH, organic matter content and rate of residue application and lignin content He et al. 2003). Water is one of the key factors affecting microbial biomass, as it is the media through which all nutrients are dissolved and made available for plant and microbes uptake. Soil microbial biomass N increased with increase in total N (Table 4). When total soil N is increased, competition for N between microbes and plants is probably reduced since adequate N is available for microbial and plant growth. The results of this study also agree with what was

reported by Moore et al. (2000) and Benjamin and Douglas (1998) that Nmic is positively related total N.

Table 4: Relationship between Nmic and measured variables in the 2003 long and short rains at Kabete.Kenya

Variables	**Linear Regression equation**	**R^2value**
moisture	Y=2.38x-31.17	0.63
% total N	Y=166.4x+11.8	0.56
maize total dry matter	Y=0.05x+9.92	-0.21
N mineralization	Y=0.09x+0.75	0.73

Soil microbial biomass N decreased with increase in maize total dry matter (Table 4) and this may partly be attributed to plant-microbe competition for mineral N. It also suggests that a competitive balance between plants and microbes may not yet have been reached and therefore increasing microbial N immobilization may result in reduced plant uptake. Low crop yield would also mean low N uptake by plants and consequently more N available for microbial growth. The results of this study concur with the observations made by Kaye and Hart (1997) and Schimel et al. (1989) that the relationship between plant N and Nmic may indicate a potential competitive interaction for mineral N. Linear regression between Nmic and N-mineralization gave a positive correlation

of $R^2 = 0.73$ (Table 4) meaning that net N-mineralization was influenced by the size of the Nmic. Generally, it would be expected that a large microbial biomass would result in high N-mineralization especially if the substrate is not limiting.

4.6 Total soluble carbon dynamics in the 2003 long rains

Total soluble carbon (water soluble organic C) was determined for only one season (2003 LRS) since carbon tends to be more stable than any other nutrients in the soil. Sole senna treatment recorded the highest total soluble carbon in April-2003, June-2003 and August-2003 while control treatment gave the lowest total soluble carbon in August and October-2003 (Figure 3). In April-2003 and June-2003, sole tithonia and calliandra+urea treatments resulted in lowest total soluble carbon respectively. Total soluble carbon observed for the control treatment was found to be significantly ($P \leq 0.05$) lower than that of all other treatments except sole urea treatment in August-2003 (Figure 3). Sole senna treatment gave significantly ($P \leq 0.05$) higher soluble carbon than sole urea in August-2003. In October-2003, all treatments resulted in significantly higher total soluble carbon than the control except sole tithonia and calliandra treatments. In the same month urea treatment gave significantly higher total carbon than sole tithonia and calliandra treatments. Tithonia+urea and calliandra+urea treatments resulted in higher total soluble carbon than their corresponding sole organic treatments throughout the season except in June-2003 when sole calliandra gave higher than calliandra+urea treatment (Figure 3). Senna+urea (309.0 mg C kg^{-1}) treatment gave higher total soluble carbon than sole senna (295.0 mg C kg^{-1}) in October-2003 only. Tithonia treatment resulted in highest difference of total soluble carbon between organic+urea and sole organic treatment in April-2003, August-2003 and October-2003. A significant difference ($P \leq 0.05$) was observed between sole tithonia and tithonia+urea treatments as the latter resulted in higher total carbon than the former in October-2003.

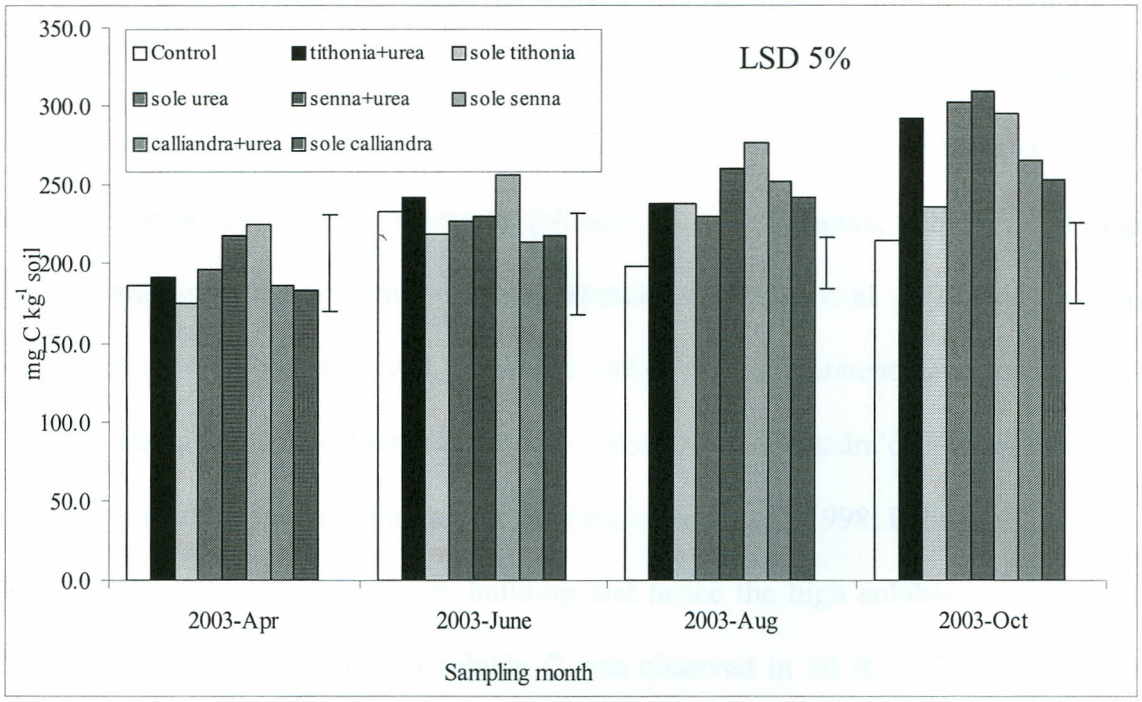

Figure 3: Dynamics of soluble carbon in 2003 LRS at Kabete, Kenya

However, in June-2003, calliandra treatment gave the highest difference in total soluble carbon. Total soluble carbon was found to increase with time within the season. The lowest total soluble carbon was observed in April-2003 (beginning of the season) while the highest was in October-2003 (end of season) (Figure 3). The increase in total soluble carbon was found to be greater with organic+urea than sole organic treatments within the season.

The results indicate an increase in soil soluble C with organic inputs amendments. Sole tithonia treatment recorded lower soluble carbon compared to the other sole organic treatments throughout the season while sole senna treatment recorded the highest soluble carbon throughout the season except in October-2003. The results suggest that it is not only the C content of a material that is

important, but also it's quality. Since the substrate contains a lot of different complex compounds, the entire C may not be easily accessible to the decomposers. The quality is a measure of how accessible the C is to the microbes and the more accessible the higher the quality and the more easily decomposable is the substrate (Nilsson, 2004). Tithonia, which is of high quality, decomposes faster than the other organic materials (Gachengo et al. 1999) and this may explains the low soluble carbon observed. Generally, sole tithonia treatment gave lower soluble carbon than sole senna and sole calliandra treatments. Senna and calliandra decompose fairly slowly due to their high polyphenol and lignin contents (Mafongoya et al. 1998; Palm and Sanchez 1990) and this leads high soil organic matter build-up and hence the high soluble carbon determined for these treatments. An increase in soluble C was observed in all the treatments after combining organic and inorganic inputs and this could be attributed to increased decomposition due to the readily available N from urea, as has been explained in section 4.3. However, combining organic and inorganic inputs had no significant effect on soil soluble carbon.

4.7 Effects of organic, inorganic inputs and their combinations on soil microbial biomass carbon

The control treatment resulted in lowest soil microbial biomass C (Cmic) throughout the 2003-LRS and was found to be significantly ($P \leq 0.05$) lower than sole senna (285.1 mg C kg^{-1}) and sole calliandra (238.4 mg C kg^{-1}) treatments in April-2003 (Figure 4). However, no significant difference in Cmic was noted in the rest of the months within this season. Sole senna and sole calliandra treatments gave the highest Cmic in April-2003 and June-2003 respectively while calliandra+urea treatment recorded highest Cmic in August-2003 and October-2003 and was found to be significantly ($P \leq 0.05$) higher in Cmic than all the other treatments except sole

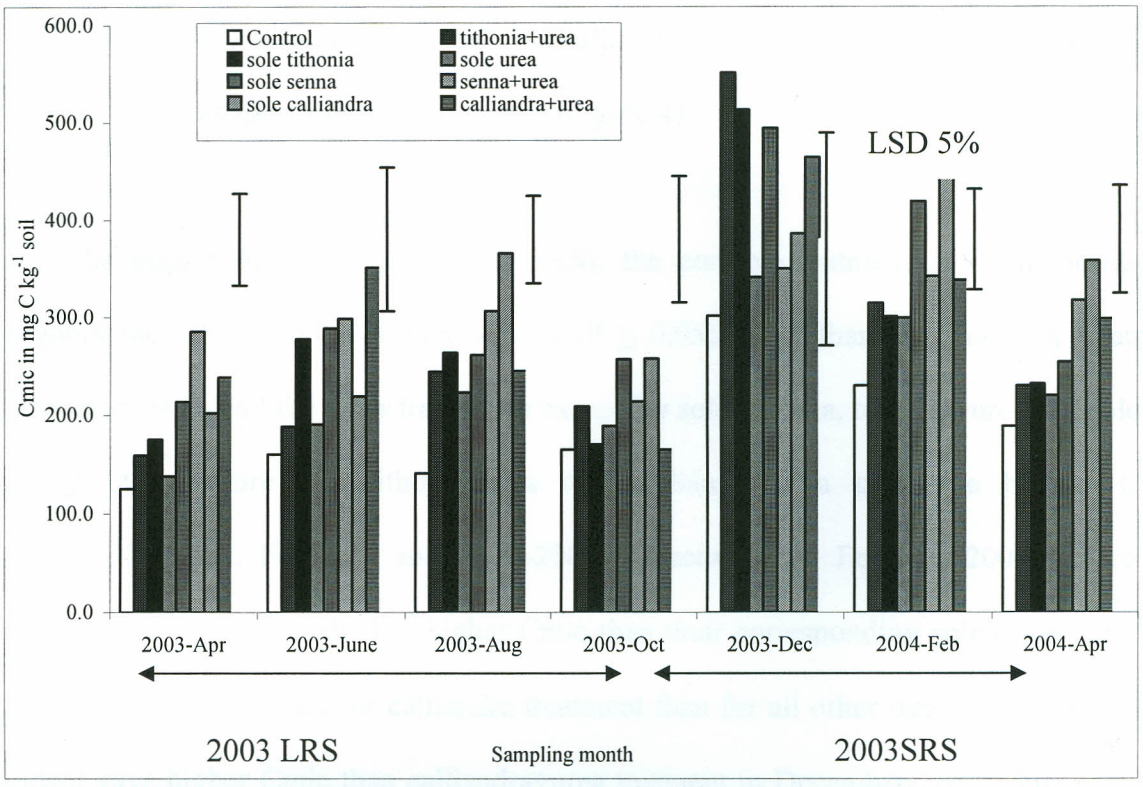

Figure 4: Treatment effects on soil microbial carbon in 2003 LRS and SRS at Kabete, Kenya

calliandra and sole senna in the former month. Organic+urea treatments did not show a consistent effect on Cmic within the season. In April and June-2003, all treatments receiving organic+urea gave lower Cmic than their counterpart sole organic treatments (Figure 4). In August-2003 calliandra+urea treatment resulted in higher Cmic than sole calliandra treament. However, in October-2003, all organic+urea treatments recorded higher Cmic than their corresponding sole organic treatments. Calliandra+urea and sole calliandra treatment gave the highest difference in Cmic in August-2003 and October-2003 with 120.0 mg C kg^{-1} and 92.3 mg C kg^{-1} respectively. In June-2003 (eight weeks after input additions), an increase in Cmic was observed compared to April-2003 for all the treatments with sole calliandra (12.7 mg C kg^{-1}) and calliandra+urea (16.9 mg C kg^{-1}) recording the highest and lowest increases, respectively. This also marked the peak increase in Cmic during this season. The increase continued in August-2003 for all treatments

except for sole tithonia, senna+urea and sole calliandra. However, in October-2003 (end of 2003 LRS), all treatments gave a decrease in Cmic (Figure 4).

During the season that followed (2003 SRS), the control treatment gave the lowest Cmic throughout the season and was significantly ($P \leq 0.05$) lower than tithonia+urea treatment in December-2004 and all the other treatments except for sole tithonia, tithonia+urea and sole urea in February-2004 (Figure 4). Tithonia+urea and calliandra+urea gave the highest Cmic in December-2003 and, February and April-2004 respectively. In February-2004, all treatments receiving organic+urea resulted in higher Cmic than their corresponding sole organic treatments and the increase was higher for calliandra treatment than for all other treatments. Sole calliandra treatment gave higher Cmic than calliandra+urea treament in December-2003 while sole tithonia and sole senna resulted in higher Cmic than their corresponding organic+urea treatments in April-2004. The treatment effects on Cmic over time in 2003-SRS was similar to that observed in 2003-LRS. An increase in Cmic was observed in December-2003 (eight weeks after input application) (Figure 4). The increase was found to be higher for sole organic treatments than organic+urea treatments. However, unlike in the 2003-LRS, the increase in Cmic did not continue in February-2004 (sixteen weeks after input application) but rather a decrease was noted and this continued in April-2004 (end 2003-SRS) (Figure 4).

The results indicate that Cmic is highly influenced by addition of inputs as the control recorded lower Cmic than all the treatments. Further to this, treatments with organic inputs recorded higher levels of Cmic than urea treatment. This supports one of the hypotheses of this study that organic inputs will increase soil microbial biomass. The increase in Cmic could be attributed to readily

available C for microbial growth after addition of easily decomposable inputs. The results of this study concur with the findings of Leita et al. (1999), Smith et al. (1993) and Tunlid and White (1981) where the control and fertilizer treatments recorded lower values than organically treated soils. Generally, the growth and functioning of microbial biomass is limited by C (Singh and Singh, 1993; Smith and Paul, 1990). When flushes of C are supplied to the soil in the form of residues, the microbial biomass increases in size until the substrate is depleted. Coleman et al. (2000) stated that soil organisms are strongly limited by available energy sources and are in a state of starvation much of the time. The increased supply of organic materials may possibly eliminate this state, in turn allowing the microorganisms to subsequently increase in number hence increase in biomass. Further, the results may also suggest that equilibrium in C supply and microbial biomass has not yet been attained with the current organic residue management regime since the experiment was only started in 1999.

Organic+urea treatments did not show a consistent trend in their effect on Cmic compared to sole treatments. An increase in Cmic was observed for the organic+urea in the month of October-2003 (beginning of SRS) and February-2004 short rain season above the sole organic treatments. The increase in Cmic may partly be attributed to the readily available N in the urea leading to higher decomposition and therefore, more C available as substrate for microbial growth. The increased Cmic observed in the study agrees with what has been reported in some studies. Thus, Singh and Singh (1993) reported a 77% increase in Cmic under straw+fertilizer treatment while Hossain et al. (1995) described a stimulatory effect of fertilizer application on Cmic increase, though the magnitude of response appeared to be dependent on the type of fertilizer added. The results of a study reported by Grierson et al. (1999) also indicated a significant increase in Cmic, 14 days after fertilizer N application.

Although the quantity of microbial biomass is mainly related to C inputs, other mitigating factors can regulate the growth and activity of the native micro flora (Smith and Paul, 1990). The inconsistency observed in the present study suggests that the temporal variations in Cmic may have been driven by climatic (e.g. temperature and moisture) factors other than fertilizer as also observed by Kasia et al. (1999) in a study carried out on arable soils of Eastern plains of Columbia.. This reflects the sensitivity of microbial biomass to varying seasonal climatic conditions. Changes also in Cmic within the season seemed to depend on the stage of plant growth. Thus, soil microbial biomass C measured was high eight weeks after inputs applications in both seasons and this also coincided with stage of rapid plant growth. These results concur with the findings of Kaiser and Heinemeyer (1993), Fraser et al. (1988), McGill et al. (1986) and Lynch and Panting (1982), that crop growth often stimulates an increase in the size of microbial biomass during the growing season. Similar results were also reported by Kasia et al. (1999) in a study carried out on temporal changes in microbial biomass C and cellulolytic enzyme activity in arable soils of Eastern plains of Columbia.

4.8 Relationship between microbial biomass carbon and microbial biomass nitrogen, moisture, soil total carbon and maize yield

Linear regression analysis gave a poor relationship between Cmic (soil microbial biomass carbon) and Nmic (soil microbial biomass nitrogen) in both seasons (Table 5). A negative relationship was observed between Cmic and soil moisture. However, there was a reasonably close linear regression and positive relationship noted for Cmic and percent total C and, Cmic and maize grain yield (Table 5). The poor relationship observed for Cmic and Nmic may be attributed to the

fluctuating rainfall patterns in both seasons (Appendix 1) that may have resulted in reduced decomposition of organic resources in the periods of moisture stress.

Table 5: Relationship between soil microbial biomass carbon and measured variables at Kabete, Kenya

Variables	**Linear Regression equation**	**R^2**
Cmic:Nmic (2003 LRS)	Y= 0.06x + 30.7	0.06
Cmic:Nmic (2003 SRS)	Y=-0.001x-11.3	0
Cmic: moisture	Y= -10.73-20.2	-0.26
Cmic: total C	Y= 1120x-18.99	0.65
Cmic: maize yield	Y= 57.13x+397.9	0.54

The reduced decomposition rate may imply low level of nutrients available for microbial growth and especially N, which is only available for plant and microbes uptake in solution form. Temporal changes in soil moisture, soil temperature, and C input from crop roots, rhizosphere and crop residues can have a large effect on soil microbial biomass and activity (Ross, 1987). A poor correlation (r = 0.38) between Cmic and Nmic was also reported for soils at North Research Center in Iowa, USA sampled in 1997 in a study on soil microbial biomass carbon and nitrogen as affected by cropping systems (Moore et al., 2000).

The negative relationship observed in the present study between Cmic and moisture in soils is similar to the findings of Ross (1987), who noted a negative relationship between moisture and

soil microbial biomass for New Zealand soils under tussock grasslands. Increase in soil moisture leads to poor aeration in moist soils as water displaces oxygen. The deficit aeration condition with increased soil moisture could lead to low microbial growth and this might explain the negative relationship between soil microbial biomass and moisture.

Soil microbial biomass C was positively correlated with total C and this can be explained by the fact that more carbon is available as food for the microbes with high total carbon (Table 5). An approximately linear relationship between Cmic and total C in soils has been shown in this study although the ratio is subject to change with soil type, soil management and environmental conditions. Anderson and Domsch (1989) stated that the ratio of Cmic to organic C is a good indicator of changes in microbial performance caused by environmental conditions. The Cmic: total C can therefore provide an effective early warning of an improvement or deterioration in soil quality (Polwson, 1994) and its sole measurement could be used to show whether different soil management practices are increasing or losing organic matter. Linear regression analysis indicated that Cmic was positively correlated to maize total yield (Table: 5). The results suggest that Cmic could be used to predict crop yield and agrees with what many authors have indicated (Benjamin and Douglas et al., 1998; Frank et al., 1994; Myrold et al., 1989), that Cmic accurately predicated microbial biomass across sites. Insam et al. (1991) reported similar correlation between Cmic and the yields of sorghum, rye and corn.

4.9 Effects of organic, inorganic inputs and their combinations on carbon dioxide evolution

The control treatment evolved lower CO_2 than all the other treatments throughout the 2003 LRS (Figure 5) indicating lower microbial activity or lower microbial biomass. Carbon dioxide (CO_2) evolved by urea treatment was lower than all the treatments in this season except in June-2003 when it gave higher than sole calliandra treatment.

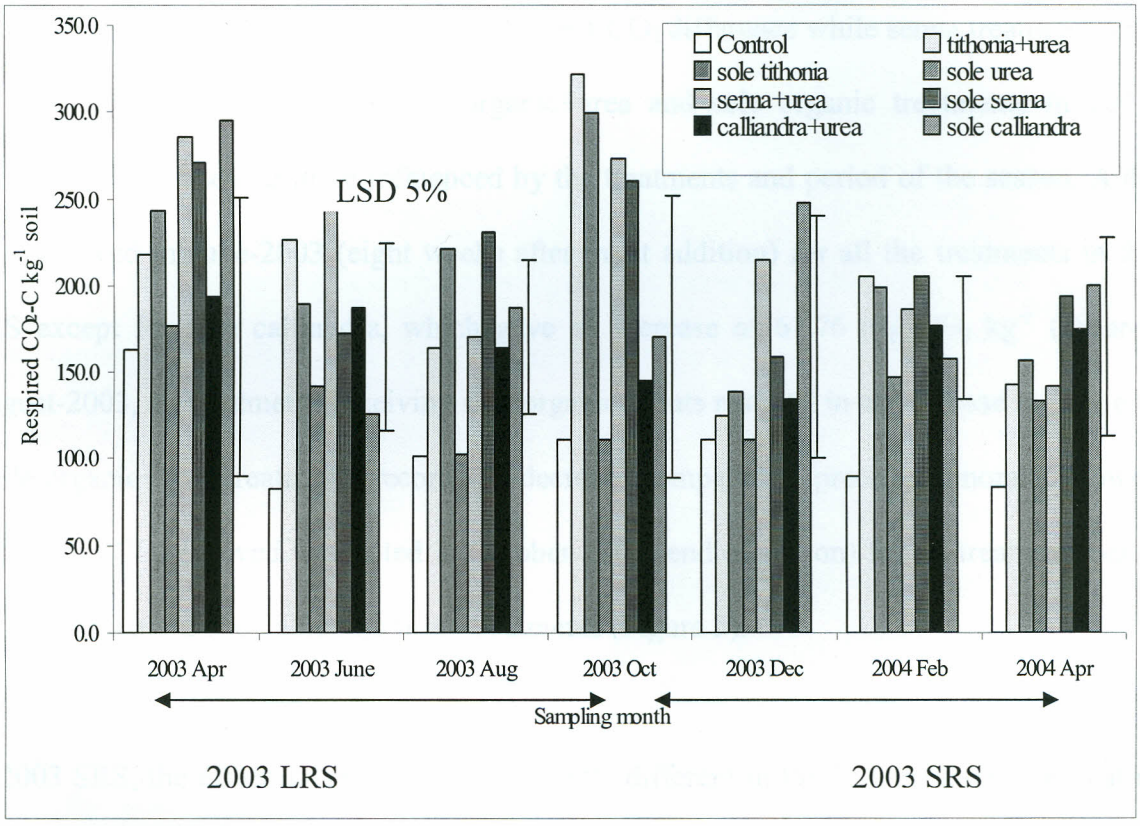

Figure 5: Treatment effects on soil respiration in 2003 LRS and SRS at Kabete, Kenya

In the same month (June-2003), control, sole urea and sole calliandra treatments evolved significantly ($P \leq 0.05$) lower CO_2 than senna+urea treatment. In October-2003, sole tithonia (299.4 mg CO_2 kg^{-1}) and tithonia+fertilizer (321.0 mg CO_2 kg^{-1}) treatments also evolved significantly ($P \leq 0.05$) higher CO_2 than the control (111.2 mg CO_2 kg^{-1}) and sole urea (111.3 mg

CO_2 kg^{-1}) treatments (Figure 5). Organic+urea treatments influenced CO_2 evolution differently at different times within the season.

In June-2003 (eight weeks after input application) all the organic+urea treatments evolved higher CO_2 than their corresponding sole organic treatments (Figure 5). However, this trend was reversed in August-2003 as all treatments receiving sole organics evolved higher CO_2 than organic+urea treatments. Callaindra treatment gave the highest CO_2 difference while senna treatment resulted in the lowest CO_2 difference between organic+urea and sole organic treatments in 2003 LRS. Changes in CO_2 seemed to be influenced by the treatments and period of the season. A decrease was observed in June-2003 (eight weeks after input addition) for all the treatments in the 2003 LRS except for sole calliandra, which gave an increase of 61.76 mg CO_2 kg^{-1} (Figure 5). In August-2003, all treatments receiving sole organic inputs resulted in an increase in CO_2 evolution while organic+urea treatments recorded a decrease compared to preceding months. However, an increase in CO_2 evolved was noted in October-2003 (end of season) for all treatments except for the sole calliandra and calliandra+urea treatments (Figure 5).

In 2003 SRS, the effect of treatments was slightly different in the CO_2 evolved from that in 2003 LRS. Carbon dioxide contents measured for control and urea treatments were lower than for all other treatments in this season. In Februay-2004, control treatment resulted in significantly lower CO_2 contents than sole tithonia, tithonia+urea and sole senna treatments (Figure 5). Sole calliandra treatment gave the highest CO_2 evolved in December-2003 and April-2004 while tithonia+urea and sole senna gave the highest CO_2 in February-2004. Unlike in the 2003 LRS, organic+urea treatments resulted in lower CO_2 evolution for all the treatments in the 2003-SRS

except for the sole senna and sole calliandra which gave higher CO_2 than their corresponding organic+urea treatments in December-2003 and February-2004 respectively. The change in CO_2 evolution between the different months within the season was similar to that observed in the 2003-LRS. A decrease in CO_2 was observed in December-2003 (eight weeks after input addition) (Figure 5) except for sole calliandra treatment. In February-2004, all treatments recorded an increase in CO_2 evolved except sole calliandra. However, at the end of the season (April-2004), there was a decrease in CO_2 evolved for all treatments except for tithonia+urea and sole calliandra.

The results indicate that organic material increased microbial respiration as more CO_2 was observed in all the treatments amended with organic materials. The findings are consistent with what was reported by John et al., 1999 that soil microbes are carbon-limited and that the additions of organic carbon can drastically increase microbial biomass in the soil. The effect on CO_2 evolution with sole organic and organic+urea material seemed to be long-term, as the CO_2 measured at the end of each season was higher compared to control and urea treatments which may be attributed to more available substrate as C for the microbes and therefore continued microbial activity. Sole tithonia and sole senna treatments tended to give high CO_2 evolution among the sole treatments which could be attributed to their higher decomposition rate and therefore releasing nutrients for microbial growth faster than calliandra. Short-term increase in the C mineralization in the soil following physical treatments is believed to have a biological origin (Azam et al., 2003) and this may explain why the organic treatments recorded higher CO_2 evolution.

Organic+urea treatments gave higher CO_2 evolution than their corresponding sole organic treatments in June-2003 (eight weeks after inputs application) of the 2003 LRS and February-2004 for calliandra and tithonia treatments (Figure 5). The increase in CO_2 evolution could be associated with readily available N for the microbes and hence the increased activity. This agrees with what was reported by Sakala et al. (2000), Henriksen and Breland (1999) and Recous et al. (1995) that addition of exogenous source of inorganic N has a positive impact on C mineralization particularly in the presence of high C: N ratio organic residues and N-depleted soils. However, organic+fertilizer did not significantly affect CO_2 evolution and this could partly be attributed to the quantity of the inputs added as well as the time between the sampling periods. Microbial activity is very dynamic and a close sampling interval may mean a lot of changes might have not been detected. The current study may also have been too short to allow this effect to be noted.

A decrease in CO_2 evolution was observed in June-2003 and December-2004 (eight weeks after addition of inputs) for the 2003 LRS and SRS respectively in all treatments except for sole calliandra treatment (December-2003) (Figure 5). This decrease could be attributed to drastic changes in soil moisture due to the fluctuating rainfall pattern. The months of June-2003 and December-2004 received considerably very low amounts of rainfall unevenly distributed (Appendix I) and this could explain the reduced microbial activity observed in these months. Initial high rates of C mineralization have been attributed to the rapid decomposition of the easily decomposable components of applied organic matter. However, Reinertsen et al. (1984) postulated that fresh organic matter decomposition in the early stages was largely dependent on the sizes of water-soluble C pool, and of an intermediately available C pool.

4.10 Effect of organic and inorganic resources on basal respiration rate

There was no specific trend observed in basal respiration rate among the treatments during the 2003-LRS (Figure 6). Sole tithonia treatment resulted in the highest basal respiration rate while sole senna treatment gave the lowest in April-2003 (beginning of season). In June-2003, sole calliandra and tithonia+urea treatments resulted in highest and lowest basal respiration respectively. However, sole tithonia gave the lowest basal respiration rate in the month of August and October-2003. Further to this, sole tithonia resulted in significantly lower basal respiration rate than control and sole calliandra treatments in August-2003 as well as than all other treatments except for control, sole calliandra and calliandra +urea in October 2003 (Figure 6). Tithonia+urea and calliandra+urea treatments gave lower basal respiration rate than their corresponding sole organic treatments in April and June-2003 except for senna treatment. In August-2003 tithonia+urea resulted in higher basal respiration rate than sole tithonia treatment while all treatments receiving organic+urea gave higher basal respiration rate than their corresponding sole organic treatments at the end of this season. The changes in basal respiration observed between the months in 2003-LRS were not consistent. In June-2003 (eight weeks after application), senna+urea, sole senna and calliandra+urea treatments gave an increase in basal respiration rate. Basal respiration rate decreased in August-2003 for all treatments expect for control and tithonia+urea treatments. However, at the end of this season all treatments gave an increase in basal respiration rate except for control sole tithonia and sole calliandra treatments (Figure 6)

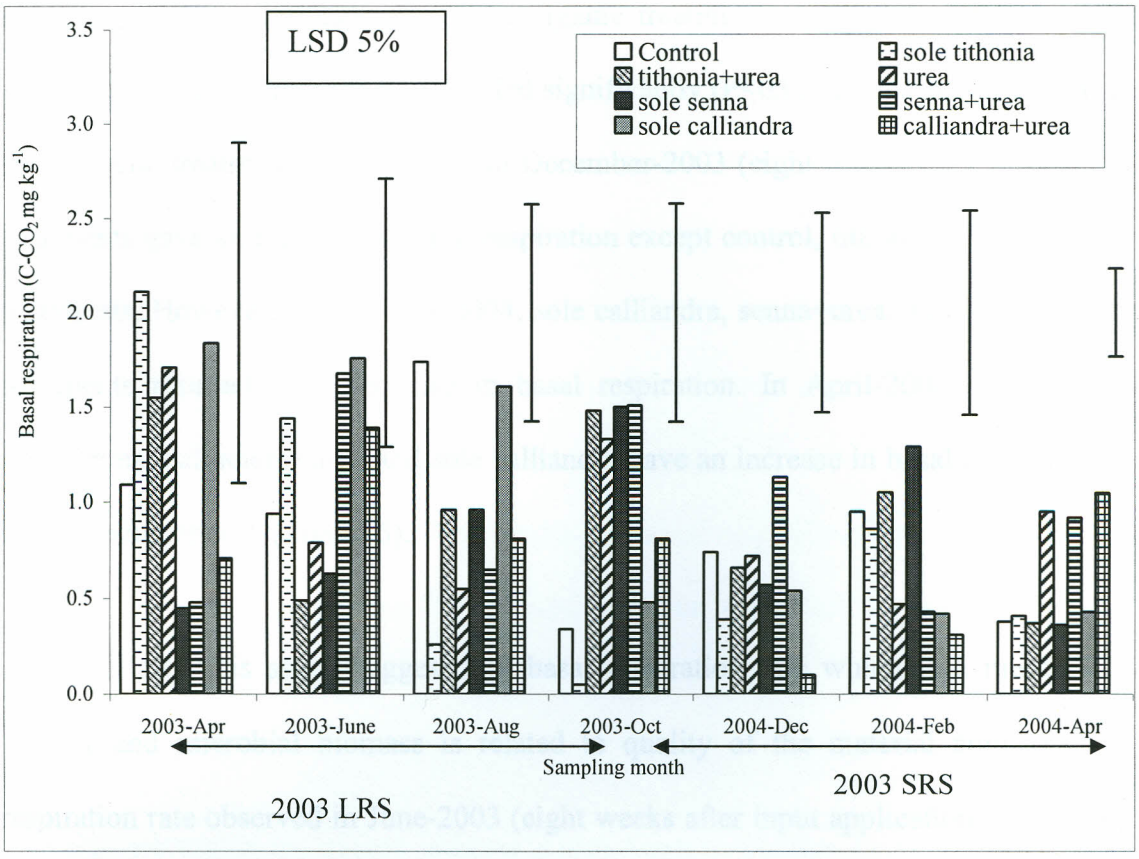

Figure 6: Effects of treatments on basal respiration at Kabete, Kenya

During 2003-SRS, basal respiration determined for different treatments did not show any consistency Thus, in December-2003, senna+urea and calliandra+urea treatments respectively gave highest and lowest basal respiration rate while in February-2004 sole urea and calliandra+urea treatments resulted in the highest and lowest basal respiration rate respectively (Figure 6). At the end of this season (April-2004), calliandra+urea gave highest basal respiration rate and tithonia+urea treatment resulted in lowest basal respiration. Combining organic and urea inputs did not show a specific trend on basal respiration rate (Figure 6). In December-2003, tithonia+urea and senna+urea treatments gave higher basal respiration than their corresponding sole organic treatments. In February-2004, only tithonia+urea treatment gave higher basal

respiration than its corresponding sole organic treatment while in April-2004 (end of season), senna+urea and calliandra+urea recorded significantly ($P \leq 0.05$) higher basal respiration than their sole organic treatment counterparts. In December-2003 (eight weeks after input application), all treatments gave an increase in basal respiration except control, tithonia+urea and calliandra+urea treatments. However, in February-2004, sole calliandra, senna+urea, Tithonia+urea and sole urea treatments resulted in a decrease in basal respiration. In April-2004 (end of season), urea, senna+urea, calliandra+urea and sole calliandra gave an increase in basal respiration compared to the preceding month (Figure 6).

The results of this study suggest that basal respiration rate which is a measure of microbial activity and microbial biomass is related to quality of the material applied. The low basal respiration rate observed in June-2003 (eight weeks after input application) for tithonia+urea and sole tithonia treatments in August and October-2003 could be attributed to its high decomposition rate (as explained in section 4.2) and therefore releasing a pool of available carbon for the microbes hence a less stressed microbial community (Figure 6). However, the high basal respiration noted for senna and calliandra treatments could be explained by their low decomposition rates and therefore leading to low amounts of soil soluble carbon available for the microbes. Further to this, the high basal may be associated to a small microbial community with high activity as evidenced by the high CO_2 production and therefore suggesting a more stressed microbial community. Conservative management and organic farming has been shown to lead to decreased basal respiration rate indicating reduced stress on soil microbial community (Liebig and Doran, 1999; Islam and Weil, 2000 a,b) whereas natural forest brought under cultivation caused increased basal respiration rate, implying increased stress on soil microbial communities

(Islam and Weil, 2000 a,b). In 2003-SRS, calliandra treatment resulted in relatively lower basal respiration rate in December-2003 and February-2004. The low basal respiration rate observed for calliandra could be attributed to the slow decomposition rate for calliandra, suggesting a residual effect of this material from the first season and hence meaning that there is still soluble carbon available in the soil. The results also indicate a lower basal respiration with organic+urea than sole organic treatments though the effect was not consistent and significant. In general, fertilizer additions are capable of either reducing or enhancing a soil's basal respiration, depending on soil management (Leita et al.1999). The increase in basal respiration may be attributed to increased decomposition due to the readily available N from the added urea suggesting more carbon available for the microbes.

The changes within the season indicate a high basal respiration in April-2003 (beginning of the season) suggesting a microbial biomass with high-energy requirement. However, the decrease in basal respiration rate observed for most of the treatments under study in June-2003 and in August-2003 (Figure 6) could mean a less stressed community for the treated soils and can be explained by the fact that decomposition has taken place and therefore a pool of carbon is available. A high basal respiration is common in communities in the initial stages of development and in communities with a large ratio of active to dormant biomass (Anderson, 1994). It may also reflect the presence of a growing microbial community, a community under stress, or a greater proportion of active to dormant microbial biomass (Carpenter-Boggs, 2000).

4.11 Relationship between basal respiration and maize yield

Basal respiration rate is a measure of microbial activity and microbial biomass (Nanipieri and Grego, 1990; Brookes, 1995). Basal respiration rate was negatively correlated to maize total dry matter yield (Figure 7), which suggests that crop yield will increase with increased microbial efficiency.

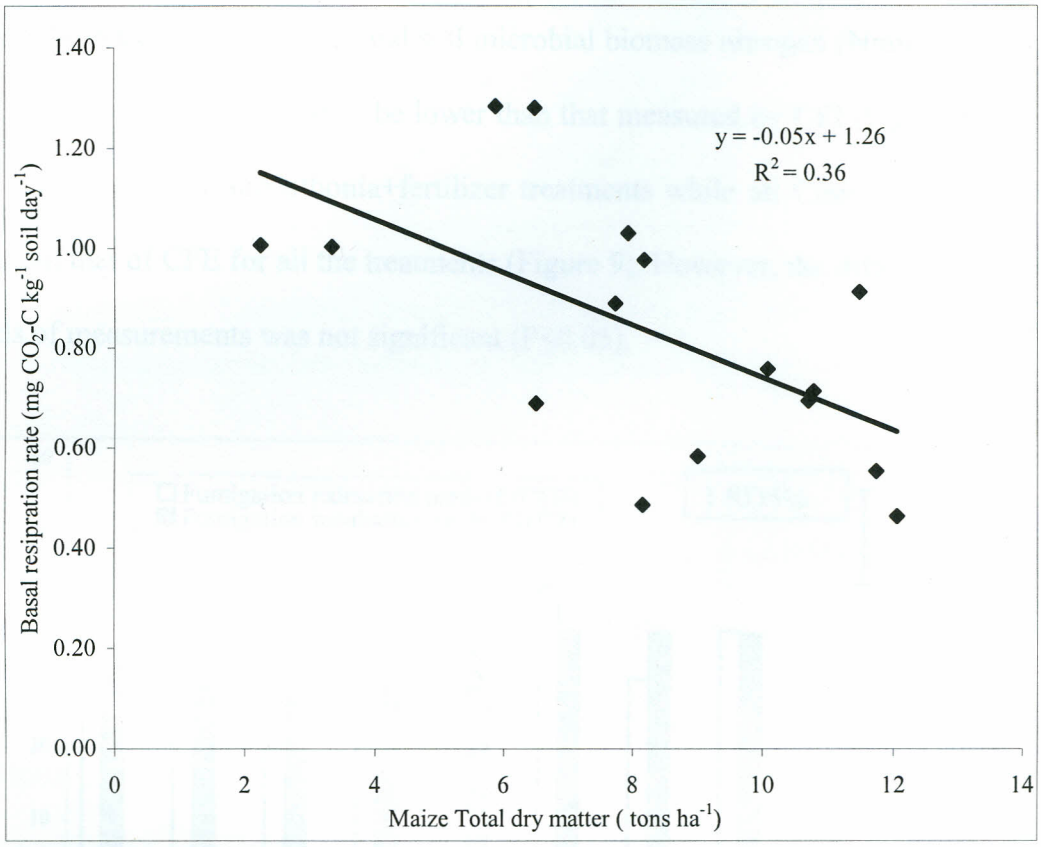

Figure 7: Relationship between basal respiration and maize total dry matter at Kabete, Kenya

This implies that if more C is lost by microbial respiration with less C input, more care must be taken to maintain organic C contents. In the present study, basal respiration observed in 2003 SRS was found to be lower than that in 2003-LRS (Figure 6), yet the maize yield recorded was higher in the 2003 SRS compared to the 2003 SRS (Table 1). Insam et al. (1991) observed a similar

correlation between soybean yield and basal respiration. Since basal respiration depends very much on organic C, the aim should therefore be to maintain organic C contents that will sustain an efficient microbial biomass for increased production.

4.12 Comparison of methods (Fumigation-extraction and fumigation-incubation methods)

In the present study both chloroform fumigation incubation (CFI) and chloroform fumigation extraction (CFE) methods were used to estimate the effect of organic and inorganic inputs on soil microbial biomass carbon (Cmic) and soil microbial biomass nitrogen (Nmic). Microbial biomass N measured by CFE was found to be lower than that measured by CFI (Figure 8) except for sole senna, senna+fertilizer and tithonia+fertilizer treatments while all Cmic determined by CFI was higher than that of CFE for all the treatments (Figure 9). However, the difference between the two methods of measurements was not significant ($P \leq 0.05$).

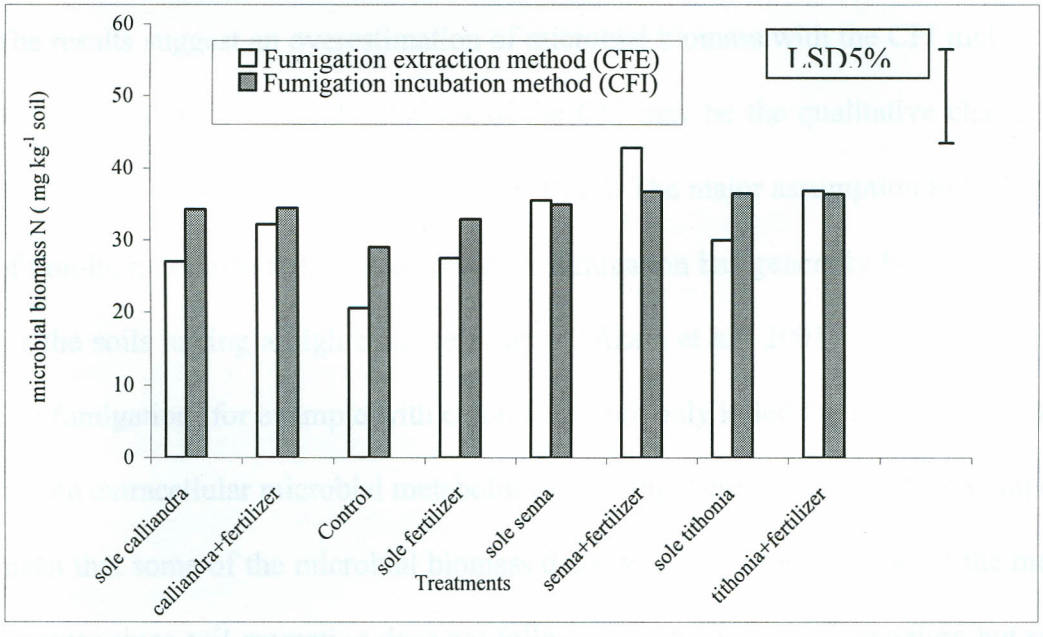

Figure 8: Comparison of microbial biomass N measured by fumigation-extraction in 2003 LRS and SRS on fumigation-incubation methods at Kabete, Kenya

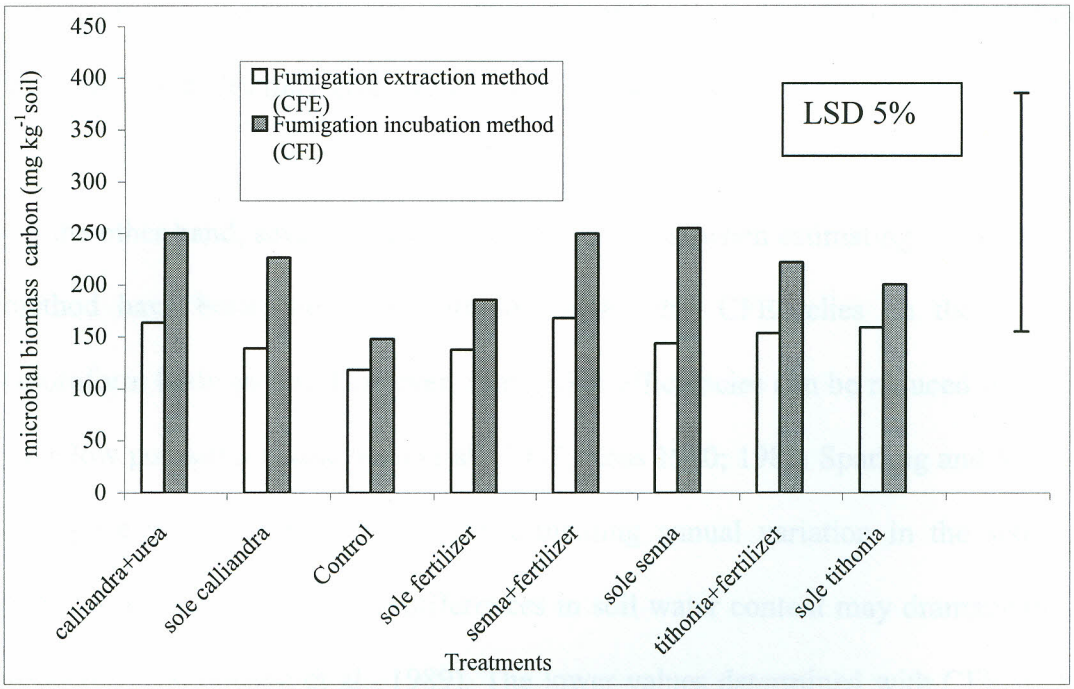

Figure 9: Comparison of microbial biomass C measured by fumigation-extraction and fumigation-incubation methods in 2003-LRS and SRS Kabete, Kenya

The results suggest an overestimation of microbial biomass with the CFI method compared to the CFE method. One of the limitations of the CFI may be the qualitative change in the microbial population following fumigation (Martin, 1963). The major assumption in CFI that mineralization of non-biomass materials is unaffected by fumigation has generally been found invalid especially for the soils having a high basal respiration (Azam et al., 2003). Shields et al. (1974) suggested that fumigation (for example with chloroform) not only killed the microorganisms but also altered certain extracellular microbial metabolites rendering them susceptible to decomposition. This may mean that some of the microbial biomass determined could not be part of the main soil microbial biomass since soil extraction does not follow immediately after fumigation but rather is incubated during which other materials could continue to decompose and therefore increasing the microbial biomass determined by this method. Using ^{15}N isotope methodology, Azam et al. (1989) observed

significant increase in the extractability of non-biomass N (unlabelled N) following fumigation suggesting that CFI may give overestimates of biomass.

On the other hand, several potential sources of errors when estimating Cmic and Nmic using CFE method have been cited. One problem being that CFE relies on the gaseous diffusion of chloroform through soil. However, fumigation efficiencies can be reduced in soils that are wet or have low porosities (Badalucco et al., 1997; Ross 1990; 1989; Sparling and West, 1989), and this may pose particular problems when estimating annual variation in the size of the microbial biomass pool, where seasonal differences in soil water content may dramatically alter extraction efficiencies (Davidson et al., 1989). The lower values determined with CFE in the present study could be associated with the seasonal variations within and between the seasons.

Another problem with CFE is the potential for microbial activity or enzymes to cause changes in extractable C and N during the relatively long fumigation period. One assumption of this method is that the extractable C and N levels in the unfumigated "control'' samples are the same as those in the fumigated sample at the end of the fumigation, less the "flush'' of the microbial biomass C and N released by the chloroform. However, chloroform fumigation does not stop all microbial or enzymatic activity (Badalucco et al., 1997; Davidson et al., 1989) and a substantial portion of microbial biomass may survive fumigation (Dickens and Anderson, 1999; Toyota et al., 1996; Ingham and Horton, 1987). Further, for reliable values measured by CFE, it is recommended that an extraction coefficient (k_{ec}) which is used to convert C flushes to Cmic be used in estimation of the microbial biomass. Values measured by use of automated analyzer (AA) have also been found to be superior to those recorded by determining dissolved organic C in K_2SO_4 extractant using

oxidation-titration (OT). In analyzing the microbial biomass, no k_{ec} was used since it has not been established at this site. Dissolved organic C was also determined by oxidation-titration methods and these factors could have contributed to the differences in observed microbial biomass values.

However, the difference between the two methods was not significant as linear regression analysis gave a positive correlation coefficient of $R^2 = 0.54$ and $R^2 = 0.69$ for Cmic and Nmic respectively (Figure 10 and 11). This could mean that CFE which is faster and less time consuming can be used instead of the traditional CFI. However, to get more reliable data, an extraction factor (k_{ec}) should be established for the soils under study as CFI appears superior to CFE where it has not been established. Considering that CFI procedure is subject to limitation with soils at pH values below 5, use of CFE could solve this problem, as it is applicable to a wide range of soil types and pH below 5 (Vance et al., 1987b; Martikainen and Palojarvi, 1990) and can be used for soils with newly added substrate and for submerged soils (Inubushi et al., 1991; Bremer and Van Kessel, 1990).

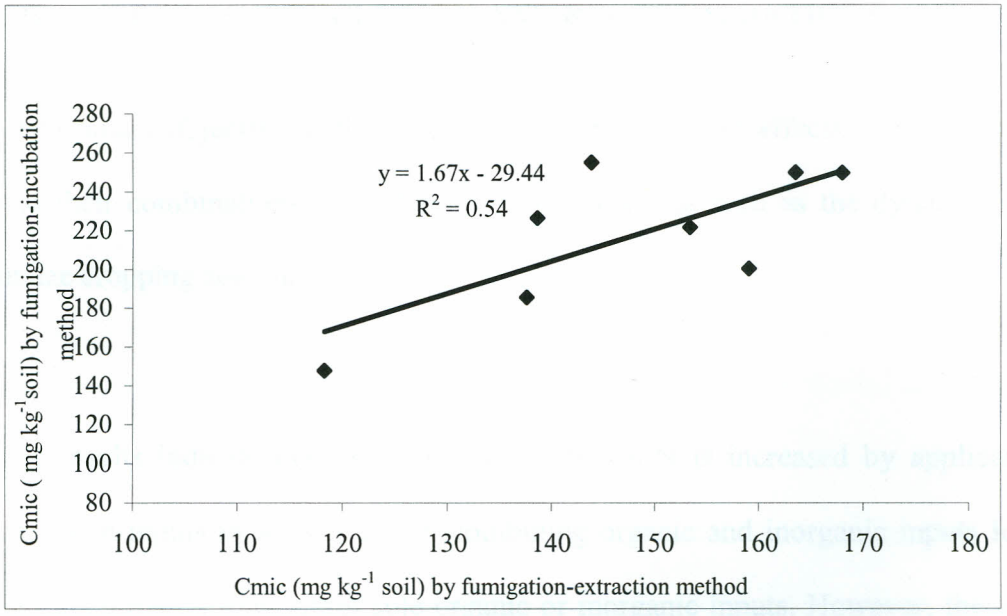

Figure 10: Relationship between Cmic measured by fumigation-extraction and fumigation-incubation methods at Kabete, Kenya

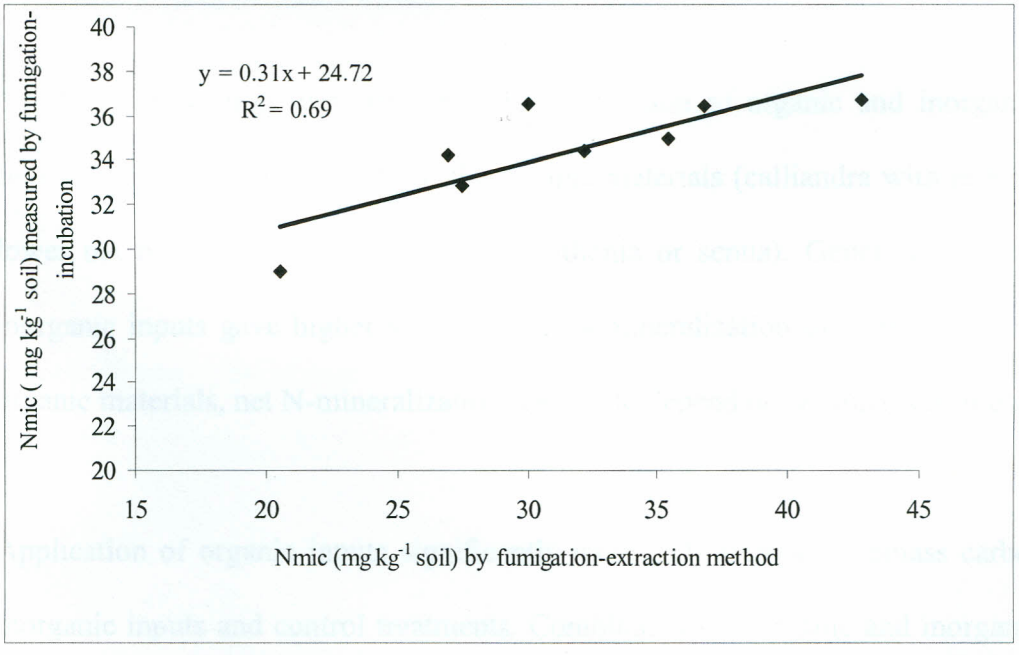

Figure 11: Relationship between Nmic measured by fumigation-extraction and fumigation-incubation method at Kabete, Kenya

CHAPTER FIVE: CONCLUSION AND RECOMMEDATIONS

The primary objective of this study was to determine the effects of organic and inorganic inputs and their combinations on soil microbial biomass as well as the dynamics of the same in two maize cropping seasons.

The results indicate that soil microbial biomass N is increased by application of organic and inorganic inputs to soils. Further, combining organic and inorganic inputs led to larger sizes of soil microbial N than either sole organic or inorganic inputs. However, the size of the microbial biomass was dependent on the stage of plant growth and seasonal fluctuations. For example, the lowest level of microbial biomass N was observed at the peak of plant growth as well as in the short rain season.

Net N-mineralization was stimulated by application of organic and inorganic inputs, and was found to depend on the quality of the organic materials (calliandra with rather poor quality gave lower net N- mineralization than either tithonia or senna). Generally, combining organic and inorganic inputs gave higher values of net N-mineralization than the organic materials. For the organic materials, net N-mineralization seemed to depend on seasonal variations.

Application of organic inputs significantly increased microbial biomass carbon as compared to inorganic inputs and control treatments. Combinations of organic and inorganic inputs increased microbial biomass carbon above the sole organic materials though the increase was not significant. However, the increase seemed to depend on the quality of the organic material with

calliandra with lowest quality treatment giving a higher increase than tithonia and senna. Microbial biomass carbon seemed to relate to stage of plant growth as the lowest values were observed at harvesting. Microbial biomass carbon was also found to appropriately predict total soil organic and plant yield.

The results on soil microbial biomass carbon (Cmic) and soil microbial biomass nitrogen (Nmic) ratio indicated that soil microbial biomass is highly dynamic and depends on time and season and these are very important parameters in soil fertility as they can give an early estimate of SOM.

Soil respiration results indicated a larger microbial biomass for soils imposed with organic materials than sole urea and control treatments. However, the respiration seemed to depend on the quality of the material. Treatments that received calliandra residues (low quality) were associated with less carbon dioxide compared to tithonia (high quality) suggesting that the efficiency of microbial biomass depended on the inputs added.

Chloroform fumigation extraction method gave lower values of microbial carbon and nitrogen than chloroform fumigation incubation method. However, the two methods were positively correlated and could be used interchangeably.

Based on the findings of this study, the following recommendations are made:

- Soil microbial biomass could be used as an effective early warning of an improvement or deterioration in soil quality and can appropriately predict crop yield.
- Farmers may be advised to apply organic or organic/inorganic materials in order to obtain a large and efficient microbial biomass for a long-term productive and sustainable ecosystem.
- For long-term increase in microbial biomass, low quality organic materials could be appropriate as they supply food for microbes for a long time compared to the high quality organic residues
- Chloroform fumigation extraction method, which is faster and less involving, could be used instead of chloroform fumigation incubation method, provided an extraction factor is established for the soils under study.

However, more research should be done in different agroecosystems as well as at different application rates in order to make comparisons and come-up with research findings that can lead to conclusive remarks on changes in soil microbial biomass. It would also be important to set a similar experiment in the greenhouse with soil amendments in order to compare the results from the field and greenhouse conditions. Moreover, short-term observations can be misleading with respect to both the magnitude and direction of long-term changes in soil microbial biomass and related variables and therefore sampling intervals and duration of the study should be considered for future research.

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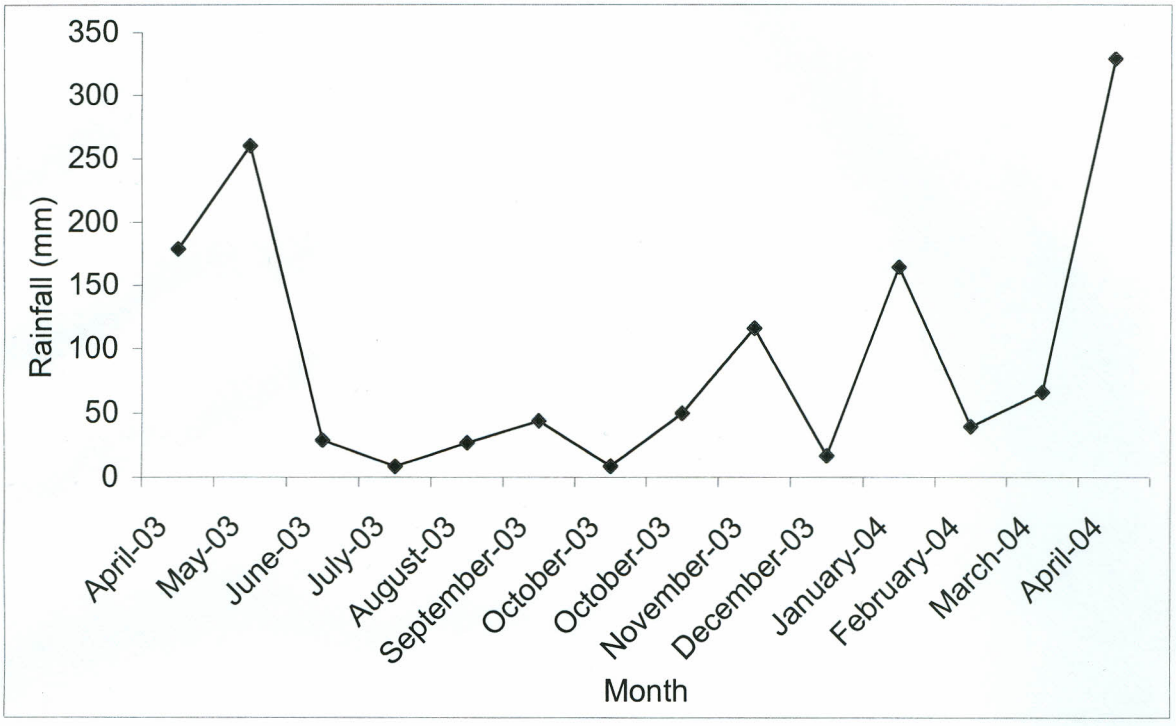
APPENDIX



Appendix 1: Rainfall amount and distribution in 2003 long and short rain season at Kabete, Kenya