EFFECTS OF BRACHIARIA GRASSES ON SOIL MICROBIAL BIOMASS CARBON, NITROGEN AND PHOSPHORUS IN SOILS OF THE SEMI ARID TROPICS OF KENYA

[EFECTO DE LOS PASTOS BRACHIARIA SOBRE EL CARBONO DE LA BIOMASA MICROBIANA, NITRÓGENO Y FÓSFORO EN SUELOS DE LA REGIÓN SEMI ARIDA TROPICAL DE KENIA]

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SUMMARY

A study was conducted to investigate the changes in microbial biomass carbon (C), nitrogen (N) and phosphorus (P) following cultivation of Brachiaria grasses in semi-arid region of Kenya. The Brachiaria grass cultivars included *Brachiaria decumbens* cv. Basilisk, *B. brizantha* cvs Marandu, MG4, Piatã and Xaraes, *B. humidicola* cv. Llanero and *B.* hybrid cv. Mulato II which were compared with two locally cultivated forage grasses (*Chloris gayana cv.* KAT R3 and *Pennisetum purpureum cv.* Kakamega 1) and a bare plot (negative check). The grass treatments were evaluated with fertilizers application (40 kg P applied at sowing and 50 kg N ha^{-1} in each wet season) and with no fertilizer applications. Microbial biomass C, N and P were determined on field moist rhizosphere soil (18-23% by weight) from a depth of 10 cm. Microbial biomass was influenced ($p < 0.01$) by grass cultivars and N and P fertilizers. Generally, microbial biomass N was higher in plots with grasses than in the bare plots. A significant enrichment of organic matter was noted in the microbial biomass when Brachiaria grasses are grown with N and P fertilizers. Among Brachiaria cultivars, the highest microbial biomass C was recorded in plots with cv. Mulato II and the lowest from the plots with cv. MG4. Brachiaria grasses with fertilizers application accumulated the highest microbial C and N compared to grasses without fertilizers, but no interaction was observed between fertilizer and grass cultivars. The cv. Marandu had the highest microbial biomass N (21.2 mg N kg^{-1}) in fertilizer treatments whereas cv. Mulato II hybrid had the highest microbial N (14.6 mg N kg^{-1}) in no fertilizer treatments.

Keywords: Brachiaria; Carbon sequestration; *Chloris gayana;* microbial biomass; *Pennisetum purpureum*

RESUMEN

Se realizó un estudio para investigar los cambios en el carbono de la biomasa microbiana (C), nitrógeno (N) y fósforo (P) después de cultivo de pastos Brachiaria en la región semiárida de Kenia. Los cultivares de Brachiaria incluyeron *Brachiaria decumbens* cv. Basilisco, *B. brizantha* cvs Marandú, MG4, Piatã y Xaraes, *B. humidicola* cv. Llanero y *B. híbrido* cv. Mulato II, que se comparó con dos gramíneas forrajeras cultivadas localmente (*Chloris gayana* cv. KAT R3 y *Pennisetum purpureum* cv. Kakamega 1) y un suelo desnudo (comtrol negativo). Los tratamientos se evaluaron con la aplicación de fertilizantes (40 kg P aplicado a la siembra y 50 kg de N ha^{-1} en cada estación húmeda) y sin la aplicación de fertilizantes. El C de la biomasa microbial, N y P se determinaron en el suelo húmedo de la rizósfera (18 a 23% en peso) de una profundidad de 10 cm. La biomasa microbiana fue influenciada ($p < 0.01$) por los cultivares de pasto y los fertilizantes de nitrógeno y fósforo. En general, el N de la biomasa microbiana fue mayor en las parcelas con pastos que en las parcelas desnudas. Un enriquecimiento significativo de la materia orgánica se observó en la biomasa microbiana cuando pastos Brachiaria se cultivan con fertilizantes de nitrógeno y fósforo. Entre los cultivares de Brachiaria, la mayor cantidad de C de la

biomasa microbiana se registró en parcelas con cv. Mulato II y la más baja en las parcelas con el cv. MG4. Los pastos Brachiaria con aplicación de fertilizantes acumulan el mayor contenido de C y N microbial en comparación con pastos sin fertilizantes, pero no se observó ninguna interacción entre fertilizantes y cultivares de pasto. El cv. Marandú tuvo la mayor cantidad de N de biomasa microbiana (21.2 mg N kg^{-1}) en tratamientos de fertilización, mientras que el cv. Mulato II híbrido tuvo la mayor cantidad de N microbial N (14.6 mg N kg^{-1}) tratamientos sin fertilización.

Palabras clave: Brachiaria; Secuestro de carbón; *Chloris gayana*; biomasa microbiana; *Pennisetum purpureum*

INTRODUCTION

Soils in the semi-arid regions of Kenya are often low in organic matter content (< 1%) and deficient in plant-available nutrients, especially nitrogen (N) and phosphorus (P). Any practice that increases the production of biomass carbon (C) via photosynthesis and slows down the return of C to the atmosphere increases C reserves in the soils through Carbon sequestration process. This would improve the soil quality and productivity (Smith *et al*., 2007) and make soils more resilient to climate change. One way to achieve this is to increase the amount of biomass C in soil where it is less susceptible to loss. This way the soil becomes a C 'sink' by absorbing atmospheric CO_2 from circulation and locking it in organic C from in the soil. Brachiaria grasses are endaphytic and have a great ability to sequester and accumulate large amounts of organic C, reduce emissions of N_2O and CH_4 due to their deep and abundant root system which also make them drought tolerant and enhance adaptation to poor fertility soils (Clapperton *et al*., 2003; Peters *et al.,* 2012).

Soil organic matter is an important component of soil quality and productivity. However its measurement alone does not adequately reflect the short term changes in soil quality and nutrient status. Measurements of biologically active fractions of organic matter, such as microbial biomass C (MBC), N (MBN) and P (MBP) reflect changes in soil quality due to more recent (1-5yr) management and cultural practices (Hargreaves *et al.,* 2003). These measurements are based on rapidly changing capacity of both C and N forms in the soils. Microbial biomass is part of the active pool of soil organic matter that plays focal roles in decomposition of organic matters, nutrient cycling and biophysical manipulation of soil structure. It is considered to be a labile reservoir of potentially plant- available nutrients, since it acts as a source and sink of plant nutrients (Brookes et al., 1984). Microbial processes are driven by the availability of decomposable organic C, which highlights the importance of sustaining and improving soil organic matter concentrations if large populations of microbes are to be active in the soil. Root exudates are a major source of substrate for soil microorganisms. These compounds can be utilized by microorganisms immediately, increasing significantly the diversity, number and activity of microorganisms in the rhizosphere. It has been accounted that nearly 5 to 21% of all photosynthetically fixed C is transferred to the rhizosphere through root exudates, which range from 20 to 50% of plant biomass (Jones *et al*., 2009).

Organic amendments such as manure, plant residues and root exudates are a major source of organic substrate in the soil. During the process of biomass turnover, the nutrients may be released slowly and taken up by the crop more efficiently (Brookes et al, 1984; Parham *et al.,* 2003; Gichangi *et al.,* 2010). These nutrients can be utilized by microorganisms immediately, increasing significantly the diversity, number and activity of microorganisms in the rhizosphere. This makes microbial biomass a fundamental component of nutrient cycling in agro-ecosystems and critical in determining soil quality (Belay *et al.,* 2002). Soil microbial biomass is an important early indicator of long term changes in soil (Saffigna *et al*., 1989; Romaniuk *et al*., 2011; Lauer *et al*., 2011) and therefore can be used to determine the level of degradation or improvement of the soil following changes in management and cultural practices (Brookes 1995; Sparling 1997). We tested the hypothesis that, cultivation of climate smart Brachiaria grasses improves soil quality through increased organic matter resulting from the large root biomass. The objective of this study was therefore to quantify the amount of soil microbial biomass C, N and P as indicators of improved soil quality resulting from 2-year cultivation of Brachiaria grasses.

MATERIALS AND METHODS

Description of the study site

The experiment was established in November during the short rains of 2013 at the Kenya Agricultural and Livestock Research Organization (KALRO) Katumani. The site is located (37°28'0''E, 1°58'0''S) 75 km southeast of Nairobi at an elevation of 1580 m above sea level. It receives mean annual rainfall of 717 mm in bimodal pattern with the long rains (LR) occurring from March to May and the short rains (SR) from October to December with peaks in April and November, respectively and a mean temperature of

19.6˚C. The dominant soils are chromic Luvisols, which are low in organic C and highly deficient in N and P and to some extent Zinc (NAAIAP, 2014).

Site characterization

Soil samples were collected in November 2013 before establishing the experiment at depths of 0–15 cm, 15–30 cm, 30–60 cm, and 60-100 cm using a bucket auger for initial analysis of the soils at the testing site. Plant litter on the soil surface was removed before collecting the soil samples. A composite soil sample, consisting of 12 cores, was collected in a grid pattern from within the 25 × 10 m blocks. Samples from each block were air-dried, visible plant roots removed, and the samples gently crushed to pass through a 2 mm sieve. The fractions sample <2 mm were used for subsequent chemical and physical analyses. Total soil N, available P (Mehlick III), exchangeable K, Ca, and Mg were estimated following standard methods as described by Okalebo *et al.,* (2002)**.** Cations Ca^{2+}, Mg^{2+}, and K^{+} were determined by atomic absorption spectrometry and soil P was measured as described by Murphy and Riley (1962).

Soil texture was determined by the hydrometer method. Soil pH was measured in water (soil: water ratio of 1: 2.5) using a pH meter and reference calomel electrode (Model pH 330 SET-1, 82362) after the suspensions were shaken for 30 minutes and allowed to stand for 1 hour. Organic carbon, was determined by the modified Walkley and Black procedure (Nelson and Sommers 1982), and ranged from 1.08 to 1.36%. Cation exchange capacity (CEC) was based on the sum of exchangeable Ca, Mg, K, H and Al after extraction with ammonium acetate. Soil bulk density was determined according to Blake and Hartge (1986). Soils were vertically sampled using stainless steel rings (diameter 10 cm) at soil depths of: 0–15 cm, 15–30 cm, 30–60 cm, and 60-100 cm, resulting in undisturbed soil samples for bulk density determination. Soil samples were dried at 65ºC to a constant weight to allow soil bulk density calculation. All determinations were made in triplicate and expressed on a dry weight basis.

Tables 1 and 2 below show the initial main soil characteristics of the experimental site. Soil pH was moderately acidic in all the depths (Table 1) and organic C content was low and decreased with depth. Similarly, N, P and Zn were low. Calcium, K and Fe levels in the soil were adequate (Table 1).

Physical analysis of soil samples from the test site indicated that the soils were sandy clay loam in the 0 - 30cm depth and clay in the lower depths (Table 2). Cation exchange capacity ranged from 20.2 to 27.8 me%, increasing with depth. This was expected as the clay content also increased with depth resulting to increased number of exchange sites (Table 2). Bulk density ranged from 1.32 to 1.45 g cm^{-3} and was greater than the ideal range of 1.1-1.3 g cm^{-3} for non-restricted plant root growth. Soil bulk density exceeding 1.46 g cm^{-3} for such soils would restrict root growth and could negatively interfere with soil aeration through reduced air-filled pore space (Landon, 1991).

Table 1 Initial soil chemical characteristics

Properties	Sampling depth (cm)			
	0-15	15-30	30-60	60-100
Soil pH (water)	5.88	5.76	5.81	6.10
Total nitrogen %	0.12	0.12	0.07	0.05
Organic carbon %	1.16	1.15	0.65	0.49
Phosphorus ppm	10	12	10	15
Potassium me%	0.29	1.01	0.52	0.32
Calcium me%	3.1	3.4	2.2	2.4
Magnesium me%	5.72	5.99	5.96	6.31
Iron ppm	17.0	17.4	18.8	18.3
Zinc ppm	1.78	1.44	0.97	0.64

Treatments and experimental design

The treatments consisted of seven Brachiaria grasses: *Brachiaria decumbens* cv. Basilisk, *B. humidicola* cv. Llanero, *B. brizantha* cvs. Marandu, MG4, Piatã, Xaraes and *B. hybrid* cv. Mulato II, two commonly cultivated local grasses [(*Chloris gayana* cv. KAT R3 and *Pennisetum purpureum cv*. Kakamega 1 (KK1) as local check)] and a bare plot (as negative control). These treatments were evaluated in the plots with fertilizer (40 kg P ha^{-1} applied at sowing and 50 kg N top-dressed in each wet season) and without fertilizer application. The treatments were laid out in a randomized complete block design in a split plot arrangement (fertilizer treatments as main plots and the grass treatments as sub plots) in three replications. The grasses were sown in November 2013 during the short rains. All the plots were kept free of weeds throughout the experimental period by hand weeding. The grasses were first harvested 16 weeks after establishment and later, on an 8-week interval harvestings during the wet seasons.

Table 2 Initial soil physical characteristics

Properties	Sampling depth (cm)			
	0-15	15-30	30-60	60-100
Bulk density g/cm^3	1.32	1.35	1.41	1.45
Sand %	50.7	48.7	44.0	40.0
Silt %	6.0	8.0	5.3	7.3
Clay %	43.3	43.3	50.7	52.7
Cation exchange capacity me%	20.2	21.3	26.9	27.8
Base saturation %	92.4	85.7	78.9	64.2
Exchangeable Sodium Potential (ESP)	0.9	0.7	0.6	0.7
Texture Class	Sandy clay	Sandy clay	Clay	Clay

Above-ground and root biomass determination

Data for aboveground plant biomass was collected eight times on an 8 weeks interval after plants were well established. The establishment period was considered as 16 weeks after seedling emergence. Harvesting of plant shoots was conducted from 2 m x 2 m net plots at a cutting height of 5 cm above ground. Samples of fresh shoot biomass were recorded, and approximately 500g subsamples were dried at 65°C to constant weight in forced-air drier for determination of dry matter. Roots were sampled using the soil-core method (Bolinder, *et al*., 2002). In each plot, four soil cores were randomly taken with a 5 cm diameter stainless steel auger to a depth of 0–15 and 15–30 cm from the inter-row and intra-row positions and composited into one sample per plot for each depth. The sampling was carried out at least 1m apart from the edge of the plot to avoid edge effects. Sampling was conducted at 24 and 48 weeks of plants establishment. The roots from each soil layer were washed separately by hand with a 2.8 mm and a 2 mm soil sieve under running tap water. Root samples integrating both living and dead roots were then dried at 65°C to constant weight and root dry weights were recorded.

Soil microbial biomass C, N, P

Soil samples for microbial biomass carbon, nitrogen and phosphorus analysis were collected in November 2015 twenty four months after the grasses had established. Four soil samples were carefully collected from a depth of 0-10 cm using an auger in each pasture plot and from the bare plot controls. In this study, only the top 10 cm soil was sampled which was assumed to contain the highest biological activity and most likely exhibit short-term changes in response to Brachiaria grasses cultivation. Soils from the four sampling positions of a plot were pooled to one sample and used in the subsequent analysis as described below.

Microbial biomass C, N and P were determined on field moist soil (18-23% by weight) by the chloroform fumigation-extraction technique as described in Brookes *et al*. (1984, 1985) and Vance *et al*. (1987). Briefly, 10 g dry weight equivalent of soil was fumigated with ethanol-free chloroform in a glass desiccator; and another 10g was incubated without fumigation at the same moisture content, time period and temperature for 24 h at 25°C. Both sets were extracted with 0.5 M K_2SO_4 (for C and N) or 0.5 M $NaHCO_3$ (for P). Soil microbial biomass element content was calculated as the difference between the fumigated and un-fumigated samples using conversion factors of 0.45 for C (Wu *et al*., 1990), 0.45 for N (Jenkinson *et al*., 2004) and 0.40 for P (Hedley *et al*., 1982). All determinations were made in triplicate and expressed on a dry weight basis.

Statistical analysis

The concentrations of the microbial biomass C, N and P were compared by 2-way analysis of variance (ANOVA) using GenStat statistical software (GenStat Release 4.24DE, 2005). This was evaluated by running a full model (20 treatments, 19 df), which was further split into a fertilizer effect (1 df), cultivar effect (9 df), fertilizer*cultivar effect (9 df). Differences at $p \leq 0.05$ were considered significant and mean separation was done using Fischer's protected least significant difference (LSD). Regression analyses and Pearson correlation coefficient (r) were used to find models best describing the relationships between soil microbial biomass and other soil and plant properties.

RESULTS AND DISCUSSION

Microbial biomass carbon

Although it is only a small part of soil organic C, the soil microbial biomass is regarded as one of the most sensitive indicators of ecosystem function. The effects of the different Brachiaria cultivars and fertilizer application on microbial biomass were significant ($p \leq 0.05$). Microbial biomass C was significantly higher ($p < 0.01$) in grass vegetated soils compared to bare plots (Table 3).

The MBC ranged from 23.9 to 200.5 mg C kg^{-1} of soil and 12.9 to 107.9 mg C kg^{-1} of soil in the fertilized and non-fertilized treatments, respectively. The highest MBC in cultivated Brachiaria soils was recorded under Mulato II hybrid and lowest under MG4 (Table 3). The effects of the different treatments on MBC followed the order Mulato II hybrid > Basilisk > Marandu > Xaraes > Llanero > KK1 > Piata > MG4 > KAT R3 > bare for plots that received N and P fertilizer. Microbial biomass C increased in the grass plots probably due to rhizo-deposition (Benizri *et al*., 2007). Rhizo-deposition may occur by root exudation and root cell sloughing (Rasse *et al*., 2005). These compounds represent a source of labile C in soil, which is rapidly consumed by micro-organisms (Jones *et al*., 2009), thereby stimulating microbial biomass production (Benizri *et al*., 2007). Similarly, MBC had a higher range of values in fertilized plots. This small component of the soil organic matter has been shown to be more responsive to cultural treatments than is total soil organic matter (Jenkinson *et al*., 2004).

Microbial biomass nitrogen

There was a highly significant interaction effect ($p < 0.01$) of grass cultivars by fertilizer application on MBN (Table 4). Among the N and P fertilized treatments, the soils under cv. Marandu had the highest MBN (21.2 mg N kg^{-1} soil) whereas cv. Mulato II hybrid had the highest MBN (14.6 mg N kg^{-1} soil) in non-fertilized treatments (Table 4). However, the amounts of MBN of soils under cv. Marandu in the N and P treatments were statistically similar to those recorded from soils under cultivars Mulato II, Basilisk, KK1, Xaraes, Llanero, and Piata (Table 4). The bare plot treatment had the least amount of microbial N (1.5 mg N kg^{-1} soil) and (1.1 mg N kg^{-1} soil) in the N and P fertilized and un-fertilized treatments respectively. This indicates that the cultivated grasses had greater contribution to the amounts of MBN recorded. The increases in MBN in the grasses plots due to N addition may be attributed to increased N availability to soil microorganisms. Gama Rodrigues *et al.* (2005) reported that only 40 to 60% fertilizer N is absorbed by plants, while 20-50% of the applied N is incorporated into the soil as organic N which contributes to the microbial biomass.

Table 3 Effects of cultivar and fertilizer N and P on microbial biomass C

Grass type	Microbial biomass C (mg C kg^{-1} soil)	
	No-Fertilizer NP	Fertilizer NP
B. decumbens cv. Basilisk	91.3	199.3
B. humidicola cv. Llanero	65.3	179.2
B. brizantha cv. Marandu	97.6	198.0
B. brizantha cv. MG4	47.5	125.4
B. brizantha cv. Piata	61.3	150.3
B. brizantha cv. Xaraes	79.1	189.5
B. hybrid cv. Mulato II	107.9	200.5
Pennisetum purpureum cv KK1	77.3	177.5
Chloris gayana cv. KAT R3	37.5	101.0
Control bare plot	12.9	23.6
LSD (interaction)	33.9	

LSD= Fischer's protected least significant difference

Table 4 Effects of cultivar and fertilizer N and P on microbial biomass N

Grass type	Microbial biomass N (mg N kg^{-1} soil)	
	No-Fertilizer	Fertilizer
B. decumbens cv. Basilisk	12.2	19.9
B. humidicola cv. Llanero	9.2	17.9
B. brizantha cv. Marandu	13.7	21.2
B. brizantha cv. MG4	4.3	16.6
B. brizantha cv. Piata	5.6	17.9
B. brizantha cv. Xaraes	7.7	18.7
B. hybrid cv. Mulato II	14.6	20.0
Pennisetum purpureum cv KK1	11.6	19.7
Chloris gayana cv. KAT R3	3.6	15.4
Control bare plot	1.1	1.5
LSD (interaction)	3.9	

LSD= Fischer's protected least significant difference

Studies on the effects of fertilizer application on soil microbial biomass remain equivocal. For instance, Zhang *et al*. (2005) measured significant increase of soil microbial biomass after two year of N fertilization in deteriorated grassland in China. However, Sarathchandra *et al*. (2001) reported significant decrease of soil microbial biomass in a perennial pasture of New Zealand due N fertilization. Meanwhile, Johnson *et al*. (2005) found that no effect of N applications on soil microbial biomass in upland grassland in Scotland. The mechanisms behind the variations may depend on other soil features, such as soil moisture, soil organic matter, total N, pH, the rate of N addition etc. (Compton *et al*., 2004; Heinze, *et al*., 2010), but the specific drivers are still not completely identified (Zhang and Zak, 1998). As an important component in regulating below ground ecological processes, the soil microbial populations are facilitators of nutrient availability, particularly soil N availability (Coleman and Crossley, 1996). Thus, any changes in the availability of soil N may in turn, affect the soil microbial community, and hence obviates their role in the turnover of soil organic matter. Additionally, changes in soil microbial function and community composition may trigger a series of responses, such as impacting litter and organic matter decomposition rates (Carreiro *et al*., (2000), humus formation (Magill and Aber, 1998), nutrient transformation and cycling (Fisk and Fahey, 2001), and then alter the interaction between soil microbes and plant communities.

Microbial biomass phosphorus

Microbial biomass P was significantly higher ($p < 0.01$) in treatments that received N and P fertilizer and the amounts were even much higher in soils under grasses than in bare plots (Table 5). The values of P in the microbial biomass were larger in the presence of Brachiaria grasses with significant ($p < 0.01$) interaction effect with N and P addition. Among the Brachiaria grasses cvs. Piata and MG4 had the lowest MBP in treatments without N and P application but in fertilizer treatments the MBP levels in these soils increased by 60.9% and 69.9%, respectively. Brachiaria cultivar MG4 had the highest response to fertilizer with an increase of 69.9% in fertilized soils compared to no fertilizer treatments. Basilisk, Marandu and Mulato II cultivars contributed to the highest MBP (4.4 – 5.4 mg P kg^{-1}soil) in no fertilizer treatments and (10.4 – 12.0 mg P kg^{-1}soil) under fertilizer treatments (Table 5).

Table 5 Effects of cultivar and fertilizer N and P on microbial biomass phosphorus

Grass type	Microbial biomass P (mg P kg^{-1} soil)	
	No-Fertilizer	Fertilizer
B. decumbens cv. Basilisk	4.4	10.4
B. humidicola cv. Llanero	3.7	10.5
B. brizantha cv. Marandu	5.1	11.7
B. brizantha cv. MG4	2.3	7.7
B. brizantha cv. Piata	3.5	8.9
B. brizantha cv. Xaraes	4.4	11.9
B. hybrid cv. Mulato II	5.4	12.0
Pennisetum purpureum cv KK1	5.0	11.2
Chloris gayana cv. KAT R3	2.0	6.0
Control bare plot	1.1	2.0
LSD (interaction)	1.8	

LSD= Fischer's protected least significant difference

Since microbial biomass acts as an important source of P in soils, cultivation of Basilisk, Marandu and Mulato II could contribute to increased available P levels in these P deficient soils of eastern Kenya (Table 1). The Brachiaria cultivars gave higher MBP than the commonly grown *Chloris gayana* cv. KAT R3 making them a good alternative for soil amelioration and animal feeds in the drylands of Kenya. However, the MBP in Brachiaria grasses were comparable to the widely grown Napier grass (cv. KK1). During growth, Brachiaria grasses may also

have encouraged higher microbial populations within their rhizosphere, which contributed to the increased MBP. Phosphate immobilization by microorganisms is an important sink, which contributes to microbial P pool. Addition of N and P fertilizers doubled the MBP in all the grass cultivars. The P concentration in the microbial biomass in this study falls within the range reported earlier (Brookes *et al*., 1984; Singh, 2007). It has been reported that incorporation of P into the soil microbial biomass is a mechanism that significantly increases the availability of P to plants and forms a significant pool of plant nutrients (Nziguheba *et al.*, 1998). This pool play a key role in P dynamics in soils by immobilizing inorganic P which is later mineralized (Parham *et al.*, 2003; Gichangi *et al*., 2009, Gichangi *et al*., 2010). For example, Nziguheba *et al.* (1998) reported increased soil MBP and decreased P sorption following incorporation of wild sunflower (*Tithonia diversifolia*) as green manure in an acid soil in western Kenya.

A high microbial biomass may indicate greater accumulations of C, N and P in the organic pool, and could represent either a sink or a source of plant-available nutrients, depending on the soil management. The higher C, N and P in the soil microbial biomass under Brachiaria grasses in this study may be due to a higher capacity of nutrient immobilization by the microbes from the decomposing litter fall and root residues in addition to the root exudates released which serves as substrate for microbial growth in the soil. Root exudates are a major source of substrate for soil microorganisms. These compounds can be utilized by microorganisms immediately, increasing significantly the diversity, number and activity of microorganisms in the rhizosphere. It has been accounted that nearly 5 to 21% of all photosynthetically fixed C is transferred to the rhizosphere through root exudates, which range from 20 to 50% of plant biomass (Jones *et al*., 2009). Due to its highly dynamic character, the microbial biomass responds more rapidly to soil fertility than the physical/chemical properties, which change relatively slowly (Sparling, 1997) and this might explain measurable changes in microbial biomass in the N and P fertilized plots in this study. The amounts of roots in the plots with fertilizer were significantly higher than no fertilize plots (Figure 1). Increasing root biomass influences soil organic matter: i) directly by increasing organic inputs to soil and ii) indirectly by influencing the location of roots and production of root exudates that may stimulate mineralization (Jones *et al*., 2009). Root exudates and other by-products are also more readily absorbed and protected by soil aggregates and where concentrated are more likely to persist in the particulate organic matter and humus fractions than shoot-derived soil organic C (Clapperton *et al*., 2003; Walker *et al*., 2003; Zhang *et al*., 2005).

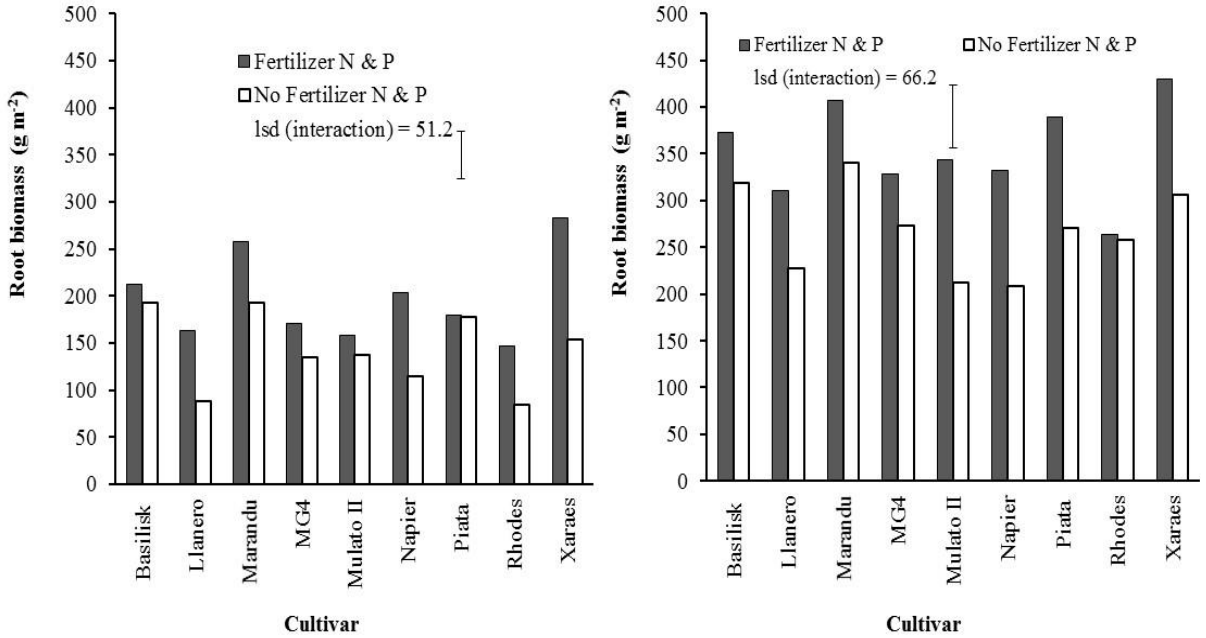

Figure 1. Effects of cultivar and fertilizer NP on root biomass (0-15cm) a) 24 weeks and b) 48 weeks after plants had established

Relationships between microbial biomass carbon, nitrogen and phosphorus

There were a number of significant correlations between microbial biomass and soil and plant properties (Figure 2; Table 6). Microbial biomass N and P showed a significant positive correlation with microbial biomass C (Figure 2). Multiple regression across all treatments showed that there were high coefficients of determination between MBC and MBN ($r^2 = 0.83$, $p < 0.01$, Figure 2a), MBC and MBP ($r^2 = 0.85$, $p < 0.01$, Figure 2b) as well between MBN and MBP ($r^2 = 0.82$, $p < 0.01$, Figure 2c). This results show evidence that soil C, N and P cycles are intimately related through the processes of mineralization and immobilization, suggesting a strong relationship that may exist between soil N and P transformations and soil C.

Microbial biomass was significantly and positively correlated to soil organic carbon ($r = 0.3139$, $p < 0.05$); ($r = 0.4596$, $p = 0.01$) and ($r = 0.2583$, $p = 0.05$) for MBC, MBN and MBP, respectively and total N ($r = 0.356$, $p < 0.05$); ($r = 0.5029$, $p = 0.01$) and ($r = 0.3521$, $p = 0.05$) for MBC, MBN and MBP, respectively (Table 6). This indicates that microbial biomass is highly influenced by the concentration of soil nutrients. Positive relationship between microbial biomass C, N and P and soil organic C and total N in grassland has been reported elsewhere by Moore *et al*. (2000). Similarly, microbial biomass correlated, significantly and positively, to root biomass measured at various stages of growth 24 and 48 weeks after plants had established (Table 6). However a stronger relationship was recorded for root biomass measured 24th week (Table 6). Roots are major C source in soil and can also stimulate SOM mineralization (Jones *et al*., 2009). Root exudates and other by-products are more readily absorbed and protected by soil aggregates and where concentrated are more likely to persist in the particulate organic matter and humus fractions than shoot-derived soil organic C (Fornara and Tilman, 2008). The capacity to generate roots, in part explains why perennials and pastures are sometimes associated with increasing soil organic matter compared to annuals.

CONCLUSION

Soil microbial biomass C, N and P in this study varied in the different grass types and was higher in the N and P fertilizer treatments and were therefore useful parameters to elucidate the changes of organic matter in the different grass type's soils. The results showed a significant enrichment of the microbial biomass component of organic matter due to the cultivation of the grasses and which was further enhanced by applications of N and P fertilizer.

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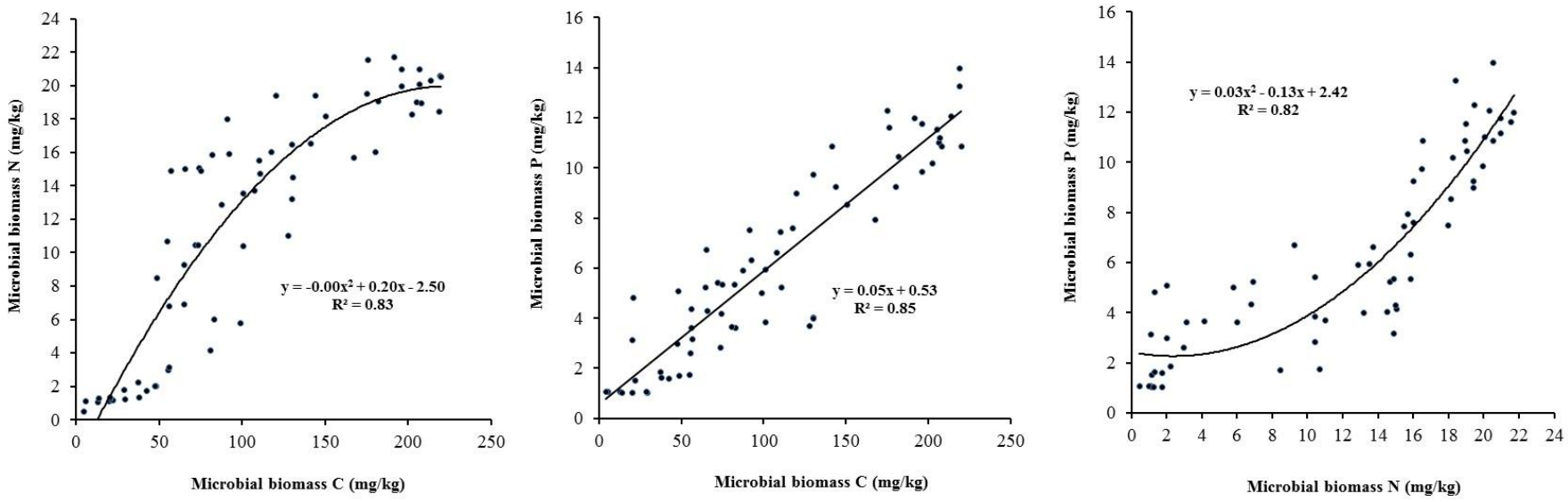

Figure 2 Relationships between a) MBC with MBN and b) MBC with MBP c) MBN with MBP

Table 6 Relationships between microbial biomass carbon, nitrogen and phosphorus with soil and plant properties

Properties	MBC	MBN	MBP	NH4-N	NO3-N	Root biomass-Week 24	Root biomass-Week 48	Total C	Total N	Above ground biomass	pH
MBC	1.0000										
MBN	0.8759**	1.0000									
MBP	0.9210**	0.8633**	1.0000								
NH4-N	0.1047	0.0998	0.1711	1.0000							
NO3-N	0.1121	0.0848	0.1750	0.9202**	1.0000						
Root biomass-Week 24	0.7172**	0.6904**	0.6905**	-0.1467	-0.2096	1.0000					
Root biomass-Week 48	0.6225**	0.6290**	0.6445**	-0.0335	-0.1143	0.8210**	1.0000				
Total C	0.3139*	0.4596**	0.2583*	-0.1984	-0.1747	0.4527**	0.4059**	1.0000			
Total N	0.3568*	0.5029**	0.3521*	0.0219	0.0480	0.4502**	0.4734**	0.8403**	1.0000		
Above ground biomass	0.6363**	0.6502**	0.6366**	0.0830	0.0227	0.6802**	0.6992**	0.4287**	0.4777**	1.0000	
pH	0.1561	0.1701	0.0576	-0.5637**	-0.6536**	0.3076*	0.2310	0.2568*	0.1101	0.3206*	1.0000

** $p < 0.01$ and *$p < 0.05$, MBC-microbial biomass carbon, MBN-microbial biomass nitrogen, MBP-microbial biomass phosphorus

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