EFFECTS OF CULTIVATED *BRACHIARIA* GRASSES ON SOIL AGGREGATION AND STABILITY IN THE SEMI-ARID TROPICS OF KENYA

[EFECTO DE PASTOS BRACHIARA CULTIVADOS EN LA AGREGACIÓN Y ESTABILIDAD DE LOS SUELOS EN EL TRÓPICO SEMI ÁRIDO DE KENIA]

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SUMMARY

Soil aggregation is a key short term indicators of soil quality attributed to changes in land management. A study was conducted to investigate changes in the size distribution and stability of soil aggregates in a structurally unstable sandy loam soil following cultivation of Brachiaria grass in semi-arid region of Kenya. Brachiaria grass cultivars included *Brachiaria decumbens* cv. Basilisk, *B. brizantha* cvs Marandu, MG4, Piata 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 fertilisers application (40 kg P applied at sowing and 50 kg N ha^{-1} in each wet season) and with no fertiliser applications. Aggregate size fractions were isolated using the wet sieving method. Aggregation based on the proportion of small macro-aggregates (250–2000 μm) increased in soils cultivated with all grass types compared to the control and was greatest in soils under *B. hybrid* cv. Mulato II. Aggregate stability in terms of mean weight diameter (MWD) differed among the grasses and was highest in soils under cv. Mulato II and cv. Marandu with MWD of 4.49 and 4.31 mm, respectively. Changes in small macro-aggregates fraction was positively correlated with particulate organic matter (POM) (r=0.9104, P= 0.001), microbial biomass carbon (MBC) (r=0.5474, P= 0.01), soil organic carbon (SOC) (r=0.3654, P= 0.05) and root biomass (r=0.4977, P= 0.01). This indicated that the binding agents were important in the aggregation of soils cultivated with Brachiaria grasses.

Keywords: Macro-aggregates; micro-aggregates; microbial biomass; organic carbon; soil.

RESUMEN

La agregación del suelo es uno de los indicadores a corto plazo de la calidad del suelo atribuido a los cambios en la gestión del suelo. Se realizó un estudio para investigar los cambios en la distribución del tamaño y la estabilidad de los agregados del suelo en un suelo franco arenoso estructuralmente inestable tras el cultivo de pasto Brachiaria en la región semiárida de Kenia. Los cultivares de Brachiaria incluyeron *Brachiaria decumbens* cv. Basilisco, *B. brizantha* cvs Marandú, MG4, Piata 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 suelo desnudo (control negativo). Los tratamientos evaluados fueron pasto con aplicación de fertilizantes (40 kg P aplicado a la siembra y 50 kg N ha^{-1} en cada estación húmeda) y sin la aplicación de fertilizantes. Las fracciones de tamaño de agregados se aislaron utilizando el método de tamizado en húmedo. La agregación basada en la proporción de macro-agregados pequeños (250-2000 m) aumentó en suelos cultivados con todos los tipos de pastos en comparación con el control suelo desnudo y fue mayor en suelos bajo *B. híbrido* cv. Mulato II. La estabilidad de los agregados en términos de peso medio de diámetro (MWD) se diferenció entre los pastos y fue mayor en suelos bajo cv. Mulato II y cv. Marandú con un MWD de 4.49 y 4.31 mm, respectivamente. Los cambios en la fracción de macro agregados pequeños se correlacionó positivamente con la materia orgánica particulada (POM) (r = 0.9104, P = 0.001), el carbono de la biomasa microbiana (CBM) (r = 0.5474, P = 0.01), carbono orgánico del suelo (SOC) (r = 0.3654, P = 0.05) y la biomasa de la raíz (r = 0.4977, P = 0.01). Esto indicó que los agentes

de unión eran importantes en la agregación de los suelos cultivados con pastos Brachiaria.

Palabras clave: Macro-agregados; micro agregados; biomasa microbiana; carbono orgánico; suelo.

INTRODUCTION

Soil aggregation is among the key short term indicators of soil quality associated with changes in land management. The aggregate stability of soils improve under certain crops, notably grasses and these improvements are frequently associated with increases in soil organic carbon (SOC) levels caused by plant residues (Lal *et al*., 2003; Marquez *et al* 2004; Denef *et al.,* 2007). Low SOC, weak soil structural stability and degradation are common attributes of most semi-arid soils of eastern Kenya (Gicheru *et al.,* 2004). Agricultural practices that could improve SOC, coupled with increase in soil surface cover, would significantly increase soil aggregate stability and reduce the soil degradation. Soil aggregate stability, defined as the ability of the aggregates to remain intact when subject to a given stress, is an important soil property that affects the movement and storage of water, aeration, erosion, biological activity and plant growth (Spohn and Giani, 2011; Pohl *et al*., 2012). There exist complex interactions between SOC storage and aggregate stability. Soil organic carbon, can encapsulate within stable aggregates thereby offering protection against microbial processes and enzymatic reaction (Lal *et al.,* 2003; Holeplass *et al.,* 2004).

The size of aggregates and aggregation state can be influenced by different agricultural activities that alter the content of SOC and the biological activity of the soil (Mills and Fey, 2003; Wick *et al*., 2009; Fonte *et al*., 2014). The article by Bronick and Lal (2005) provides overview of chemical compounds that are thought to be involved in the formation of soil aggregates, a list of factors that determine extent of this aggregation, and a description of the influence of soil structure on a wide range of soil processes. Over short periods, the stability of soil aggregates is modified under the influence of different cropping practices, probably being more related to changes in the organic constituents than to the actual total organic matter content (Reid and Goss, 1980; Milne and Haynes, 2004). Reid and Goss (1980) for example, demonstrated that after only 4 weeks growth the living roots of perennial rye grass (*Lolium perenne*) increased the aggregate stability of a sandy loam as measured by turbidimetric and wet sieving analyses which was most strongly associated with the larger aggregates. This effect was probably caused by organic substances released from the roots which either stabilized the aggregates directly or indirectly after microbial colonization (Leifeld, *et al*., 2005; Franchini, *et al*., 2007). However, over long periods of time, the stability of soil aggregates diminishes as the SOC content declines as a result of it being

used as an energy source by the microorganisms in the soil (Mills and Fey, 2003). Loss of SOC will therefore reduce soil fertility, degrade soil structure and water holding capacity and ultimately, leads to land degradation. Grasses, present the greatest effect on the aggregation and aggregate stability due to their extensive root system (Harris *et al*., 1996). Brachiaria grasses have the ability to sequester and accumulate large amounts of SOC through their large and extensive root biomass, reduce emissions of N_2O and CH_4 per unit of livestock production, and survive in dry areas of low soil fertility due to their deep and abundant root system (Fisher *et al*., 2007; Peters *et al.,* 2012). This makes Brachiaria grasses to be drought tolerant and better adapt to poor soils and therefore can offer a better option for livestock feed production and soil improvement.

The resistance of soil aggregates to breakdown from physical forces is a measure of coherence or strength of cementation between or within soil aggregates. Aggregate size is important in determining the dimensions of pore space in cultivated soils. The size of the pores in turn affects the movement and distribution of water and aeration that are major factors affecting plant growth. Soil organic carbon increases aggregate water repellence therefore minimising their disruption and breakdown when wetted through mechanical manipulation such as tillage (Chenu *et al*., 2000). Soil aggregation can be determined by mean weight diameter (MWD), geometric mean weight diameter (GMD) and aggregate stability (AS, %) index, which are obtained by fractioning the soil material into aggregate classes by wet sieving (Kemper and Rosenau, 1986).

Disruption of soil structure is common in semi-arid zones of eastern Kenya, due to the inherent soil type that has weak structure, overgrazing, compaction, and poor land management, which have negative consequences on SOC storage and degradation of the soil structure. There is therefore a need to examine the potential effects of introduced Brachiaria grasses on aggregation and stability of aggregates in these fragile soils. The objective of this study was therefore, to investigate the short-term (2-years) changes in aggregate size distribution and the stability of soil aggregates following cultivation of Brachiaria grasses. We examined linkages between SOC, particulate organic matter (POM), microbial biomass carbon (MBC), and root biomass with aggregation by comparing Brachiaria cultivated soils verses commonly grown Napier and Rhodes fodders and not cultivated weed free soils. We tested the

hypothesis that, cultivation of Brachiaria grasses improves soil aggregation through increased SOC and aggregate associated C resulting from large root biomass. By studying aggregate stability it is possible to quantify whether or not the cultivation of Brachiaria grasses would ameliorate physical conditions of soils in semi-arid eastern Kenya and other areas with similar soil characteristic across the tropics.

MATERIALS AND METHODS

Description of the study site

The experiment was conducted at the Kenya Agricultural and Livestock Research Organization (KALRO), Katumani farm between November 2013 and November 2015. The site is located (37°28'0''E, 1°58'0''S) 75 km south-east 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. The mean temperature is 19.6 °C. The dominant soils are chromic luvisols, which are low in organic C, highly deficient in N and P and to some extent Zinc (NAAIAP, 2014) and generally have poor structure.

Site characterization

Composite soil samples from 12 sampling positions within a plot 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 characterization of the soils. Plant litter on the soil surface was removed before collecting the soil samples. Samples were air-dried, visible plant roots removed, and the samples gently crushed to pass through a 2-mm sieve. The sample was 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 w/w) 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^{2+}, Mg^{2+}, K^+, H^+ and Al^{3+} after extraction with ammonium acetate. Soil bulk density was determined according to Blake and Hartge (1986).

Soils were vertically sampled using stainless steel rings with 10 cm diameter at 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 65° C to a constant weight. All determinations were made in triplicate and expressed on a dry weight basis.

Soil characteristics of the experimental site are shown in Tables 1 and 2. Soil pH was moderately acidic in all the depths (Table 1) and soil organic C content was low and decreased with increasing depth. Similarly N, P and Zn were low.

Physical analysis of soil samples from the test site indicated that the soils were sandy clay loam in the 0–30 cm depth and clay in the lower depths (Table 2). Cation exchange capacity ranged from 20.2 to 27.8 me%, and increased with depth. This is 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 negatively interfere with soil aeration through reduced air-filled pore space (Landon, 1991).

Treatments and experimental design

The treatments consisted of seven Brachiaria grass cultivars *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 including the bare plots were kept weed free throughout the experimental period by hand weeding. The grasses were first harvested 16 weeks after establishment and later, harvestings were done eight times on an 8 weeks interval during the 5 wet seasons.

Table 1. Initial soil chemical characteristics

Properties	Sampling depth (cm)			
	0-15	15-30	30-60	60-100
Soil pH (in 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

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 Percentage (ESP)	0.9	0.7	0.6	0.7
Texture Class	Sandy clay	Sandy clay	Clay	Clay

Root biomass determination

Roots were sampled using the soil-core method (Bolinder, *et al*., 2002). In each plot, four soil cores were randomly taken to a depth of 0–15 and 15–30 cm, two each from the inter-row and intra-row spacing and composited into one sample per plot for each depth. The sampling was carried out using a 5 cm diameter stainless steel auger at least 1m apart from the edge of the plot to avoid edge effects. Sampling was conducted at 24 and 48 weeks after establishment, high root accumulation was expected at these sampling periods. 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 sampling and determination water-stable aggregate distribution, particulate organic matter and microbial biomass carbon

Soil samples for aggregate, POM and microbial biomass carbon 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 a spade, so as to minimize aggregates disruption 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 grass cultivation. Soils from the four sampling positions of a plot were pooled to one sample. The soils were then dried at room temperature (21^0 C) and sieved by gently breaking soil clods along natural planes of weakness, so that they passed through an 8 mm sieve. Soil sub-samples of approximately 400g were taken using the quartering method for further processing and analysis at International Center for Tropical Agriculture (CIAT, Nairobi) as described below.

Water-stable aggregate distribution and particulate organic matter

Four aggregates-size fractions were isolated using triplicate 80 g of air-dry 8 mm sieved soil by the wet sieving method as described by Six *et al.* (1998), and each fraction was named as large macro-aggregates (> 2000 μm), small macro-aggregates (250–2000 μm), micro-aggregates (53–250 μm), and silt + clay fraction (<53 μm). The soil subsamples were spread evenly onto a 2000-μm sieve and slaked for 5 min in distilled water. The soil was then sieved for 2 min by oscillating the sieve 50 times up and down (approximately 3-cm amplitude). Large macro-aggregates retained on the 2000 μm sieve mesh were backwashed into pre-

weighed pans for drying. Large (>2000 μm) floating litter was removed, while soil passing through the 2000 μm sieve was transferred to a 250 μm sieve and the process was repeated to obtain the small macro-aggregates fraction (250–2000 μm). The sieving process was repeated once more using a 53 μm sieve to separate micro-aggregates (53–250 μm) from the silt and clay fraction (<53 μm). All pans and soil solutions were placed in an oven at 65°C until dry and weighed in order to determine the mass of each aggregate size class.

Aggregate fractions > 53 μm were corrected for sand prior to calculation of the proportional weight of aggregates and mean weight diameter (MWD) was determined. However, large macro aggregate > 2000 μm were not used in computing or calculating MWD, because the proportion of aggregates > 2000 μm recovered after the wet sieving were too small in weight to be corrected for sand (sand free fraction). The proportional weight of sand free aggregates (aggregate size distribution) was calculated as follows:

Weight of fractioned aggregate - % sand content
Weight of bulk soil - % sand content

(Eq. 1)

The MWD was then calculated as an index of aggregate stability using the formula of Kemper and Rosenau (1986) as follows:

$$MWD = \frac{\sum w_i\, x_i}{100}$$

(Eq. 2)

Where:
x (μm) = average diameter of the openings of two consecutive sieves, and
w = weight ratio of aggregates remained on the ith sieve.

Particulate organic matter was separated from water-stable aggregate fractions by floatation and decanting after mechanical dispersion of the soil by agitation in water with glass beads. The collected organic size fraction was oven dried at 65 °C for 24 h and their weight determined. The soil POM was expressed in g kg^{-1} after adjusting for soil moisture using the weight loss of sub-samples oven dried at 105 °C to a constant weight.

Soil microbial biomass C

Microbial biomass C was determined on field moist soil (18-23% by weight) by the chloroform fumigation-extraction technique as described in Vance *et al*. (1987) on soils sampled in November 2015 as described above. Briefly, 10 g dry weight equivalent of soil was fumigated with ethanol-free chloroform in a glass desiccator; and another 10 g 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$_2$SO$_4$ for microbial biomass C determination. 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 (Vance *et al*., 1987). All determinations were made in triplicate and expressed on a dry weight basis.

Statistical analysis

Treatment effects on soil aggregate stability and POM were tested using an analysis of variance (ANOVA) as a split-plot with fertilizer NP as the main factor and grass type as the sub-plot factor 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 ≤0.05 were considered significant and mean separation was done using Fischer's protected Least Significant Difference test (lsd). Regression analyses and Pearson correlation coefficient (r) were used to find models best describing the relationships between soil aggregate stability and other soil and plant properties.

RESULTS AND DISCUSSION

Soil aggregate size fractions and stability

The growth of perennial grasses enhance aggregate formation due to the production of large quantities of polysaccharide and phenolic binding agents by the large microbial biomass in the pasture rhizosphere (Milne and Haynes, 2004). Additionally, the fine grass roots and associated fungal hyphae physically enmesh fine soil particles into aggregates (Milne and Haynes, 2004). The results of aggregate size distribution and stability determinations are presented in Table 3. The effects of grass types on the proportion of aggregate size fractions 250-2000, 53-250 and <53μm were significantly (p<0.01) different. The small macro-aggregates (250-2000 μm) comprised the largest proportion, which accounted for 34.1 – 64.2% of the total soil dry weight, and the fraction of micro-aggregates (53-250 μm) was the second largest, being 28.5–48.2% of whole soil dry weight. The large macro-aggregates (>2000 μm) and silt + clay (<53μm) fractions were the least components. The silt + clay fractions accounted for only 8.9-17.6% of whole soil dry weight (Table 3). In contrast, there were no significant differences (p≥0.05) in the distributions of water stable aggregates with aggregates sizes >2000 μm between the grass types and accounted for less than 1% of the bulk soil. Fertilizer NP addition did not influence the aggregate size distribution. More than 99.6% of whole soil dry weight was recovered after wet-sieving, indicating that losses during the fractionation process were negligible.

The effects of grass types on water stable aggregates (Table 3) revealed that soils under *B. hybrid* cv. Mulato II and *B. brizantha* cv. Marandu, significantly increased small macro aggregates (250-2000 μm) fraction. However, *B. brizantha* cv. MG4 and the control bare plots, *Chloris gayana* cv. KAT R3 had the lowest levels of small macro-aggregates (250-2000 μm). The least proportion of silt and clay aggregates (<0.053mm) was recorded in soil planted to *B. brizantha* cv. Xaraes and *B. hybrid* cv. Mulato II. This could have resulted from the effect of soil cementing agents binding primary particles to micro-aggregates and macro-aggregates. Macro-aggregates (diameter >250 mm) are considered as a secondary soil structure associated with pores, microbial habitat, and physical protection of organic matter (Christensen, 2001). In addition significant differences were observed in aggregate stability expressed by MWD among the grass species and were much higher in *B. hybrid* cv. Mulato II, *B. brizantha* cv. Marandu and *B. decumbens* cv. Basilisk (Table 3). Similarly as in the proportion of aggregate fractions, the effects of fertilizer application on MWD were not significantly different.

High macro-aggregates proportion favoured soil aggregate stability as indicated by high MWD values which might have resulted from increased soil cementing by organic compounds (Bronick and Lal, 2005). The soil cementing agents bind micro-aggregates and primary particles to macro-aggregates, and minimize microbial decomposition by promoting physical protection through sorption to clay minerals and encapsulation within soil aggregates (Mikha and Rice, 2004). The increase in macro-aggregates and the decrease in fine size aggregates in the 0–10 cm layer as a result of grass treatments accelerated the integration of fine particles into the coarse elements. The results generally agree with the finding of Sommerfeldt and Chang (1985), that macro-aggregates were increased while micro-aggregates were decreased due to increased organic matter. Macro-aggregates are good predictors of potential C responses to pasture establishment because of their importance in protecting recently deposited, labile, organic matter (Dungait *et al*., 2012). Tisdall and Oades (1982), reported that the water stable micro-aggregates (<250μm) are insensitive to cropping and management, whereas, macro-aggregates are found to be dependent on soil management. According to the conceptual model of Six *et al*. (2002), recent inputs of organic matter induces macro-aggregates formation, while the decomposition of SOC within these macro-aggregates leads to the formation of stable micro-aggregates and organo-mineral complexes. Consequently, macro-aggregates formation leads to longer mean residence time of SOC in soil over time through the formation of smaller, more stable soil fractions with increasingly intimate associations between organic matter and mineral surfaces (Gale *et al*., 2000). In this study, small macro-aggregates were the most prominent aggregates fraction in the 0-10 cm soil layer in the grass treatments whereas 53-250 μm aggregates fraction was most prominent the bare plot soils. This indicates increased aggregation in the grass treatments implying that these poorly structured soils should not be left bare due to risks of erosion and surface sealing/crusting due to their high silt content.

Particulate organic matter and microbial biomass carbon

Both POM and MBC are labile non-humic fraction of organic matter and constitute important pools of nutrients in the soil. Particulate organic matter defined as organic matter that is intermediate in the decay continuum between fresh litter and humified organic matter has high sensitivity to management than total soil organic carbon (Grandy and Robertson, 2006; Todd *et al*., 2015). The POM fraction hosts a large concentration of microorganisms because it provides a substrate for their activities (Zhang *et al*., 2014). The POM and MBC are therefore important in maintaining soil structure in that the microorganisms associated with them in the decomposition process exude mucilaginous carbohydrate material which acts as a glue and helps cement soil aggregates together. For example, MBC has been shown to be positively correlated with aggregate stability, indicating the important role of MBC in aggregation (Milne and Haynes, 2004). The analysis of light organic fractions separated from wet sieved aggregates showed that POM differed between the grass types (Table 3). The POM concentration in 0-10 cm depth ranged from the minimum of 0.16 g kg^{-1} in the bare soil plots to the maximum of 0.93 g kg^{-1} in soil under *B. hybrid* cv. Mulato II (Table 3). The gains in POM within macro-aggregates shown here concur with the results of others that suggest macro-aggregates may be good predictors of potential C responses to changes in agro-ecosystems management (Grandy and Robertson, 2006; Todd *et al*., 2015). This also supports the findings of Six *et al.* (2002) who reported that soil aggregation was enhanced as soil organic matter increased, due to increased production of organic matter derived binding agents resulting from the activity of microbes on deposited residues in soils. While we found that total SOC did not vary among Brachiaria grasses over the short duration of the study (Table 3), changes in below ground C cycling were apparent through aggregates formation. Aggregation increased over the two years of this study in all grass types compared to the bare control treatment, with *B. hybrid* cv. Mulato II and *B. brizantha* cv. Marandu showing the largest proportion of small macro-aggregates and mean weight diameter (Table 3).

Root biomass

The effect of grass types on root biomass was significantly different with higher root biomass recorded in treatments with cvs Xaraes, Marandu, Piatã and cv. Basilisk (Figure 1). The amounts of roots recorded in the fertilized plots in this study were also significantly higher than where fertilizers N and P were not added for cv. Llanero, cvs Piatã, Xaraes and *B. hybrid* cv. Mulato II) and *Pennisetum purpureum cv*. Kakamega 1 (Figure 1). Generally, the composition of POM consists mainly of root fragments (Cambardella and Elliot, 1992) and therefore this affirms that significant differences in the levels of POM observed in this study were due to differences in the amounts of root biomass among grass types. Reid and Goss (1980) for example demonstrated that after only 4 weeks growth the living roots of perennial rye grass (*Lolium perenne*) increased the aggregate stability of a sandy loam soil.
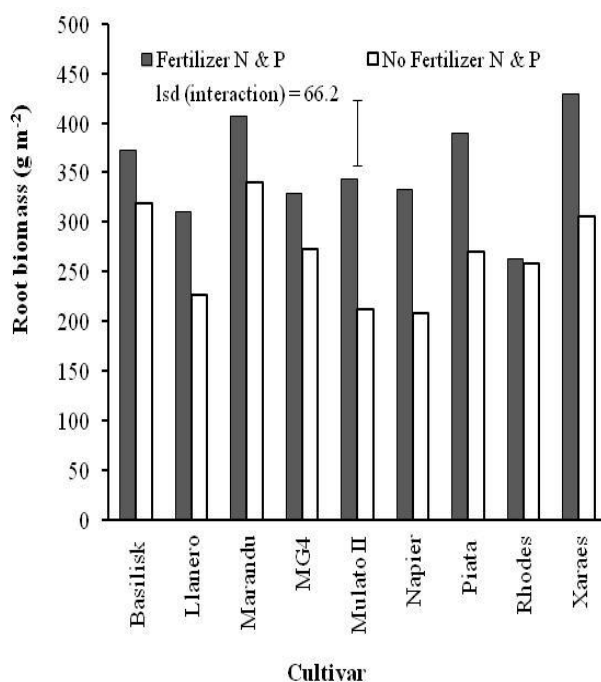

Figure 1 Effects of cultivar and fertilizer N and P on root biomass (0-15 cm depth) 1 year after the grasses were established

Plant roots can increase aggregation by enmeshing small particles into stable macro-aggregates; by supplying organic substrates such as sloughed cells and mucilage and by influencing soil moisture content (Grandy and Robertson, 2006). According to Broersma *et al*. (1997), crops affect soil structure differently because of diverse rooting habits, type of organic matter and the rhizosphere processes. Increasing root biomass influences soil organic matter: i) directly by increasing organic inputs to soil and ii) indirectly by influencing the 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 POM and humus fractions than shoot-derived SOC (Clapperton *et al*., 2003; Walker *et al*., 2003; Hurisso, *et al*., 2013; Zhang *et al*., 2014). Depending on the root turnover rates the amount of C stored in the soil can be determined from the root biomass, plant residue and SOC. Commonly, root mass and plant residue in the soil form between 3,400 (annual crop) and 17,000 (perennial grasses) kg ha^{-1} year^{-1} of the soil biomass (Harwood *et al*., 1998).

Relationships between soil aggregation and stability with binding agents

Particulate organic matter and MBC all act as important binding agents for aggregation (Six *et al*., 2004; Bronick and Lal, 2005). Previous studies have reported that soil aggregate stability was strongly correlated with POM and MBC in different soils (Six *et al*., 2002). Regression of the proportional weights of the 250-2000 μm aggregates fraction and MWD with POM showed that POM explained 79.4% and 81.7% of the variations of the aggregates fraction and MWD, respectively (Figure 2). Similar results were also reported by Ashagrie *et al*. (2007) and Spohn and Giani (2011), who suggested that POM contribute to soil aggregation as it acts as nucleation sites for the formation of macro-aggregates .

The proportional weights of the 250-2000 μm aggregates fraction and MWD in this study was found to be positively but weakly correlated with MBC (Figure 3) which however indicated that soil aggregation and stability increased with increasing levels of MBC in the bulk soil. Overall, POM made the greatest direct contributions to aggregate stability (Figure 2), suggesting that greater POM in Brachiaria cultivated soils enhanced aggregate stability and by extension improved soil structure was comparable to soils under *Pennisetum purpureum cv* KK1 a commonly cultivated fodder in the region. Gartzia-Bengoetxea *et al*. (2009) also found a strong relationship between MWD and POM. Other previous studies using other sources of SOC have reported higher aggregate MWD with increased organic C in soils (Gulde *et al*., 2008; Min *et al*., 2003; Whalen *et al*., 2003). For example, Wortmann and Shapiro (2008) observed higher macro-aggregates formation with composted manure application than unamended control. Similarly, Min *et al*. (2003) observed that livestock manure added at 32.7 Mg C ha^{-1} resulted in 30% higher aggregate stability than an unamended control.
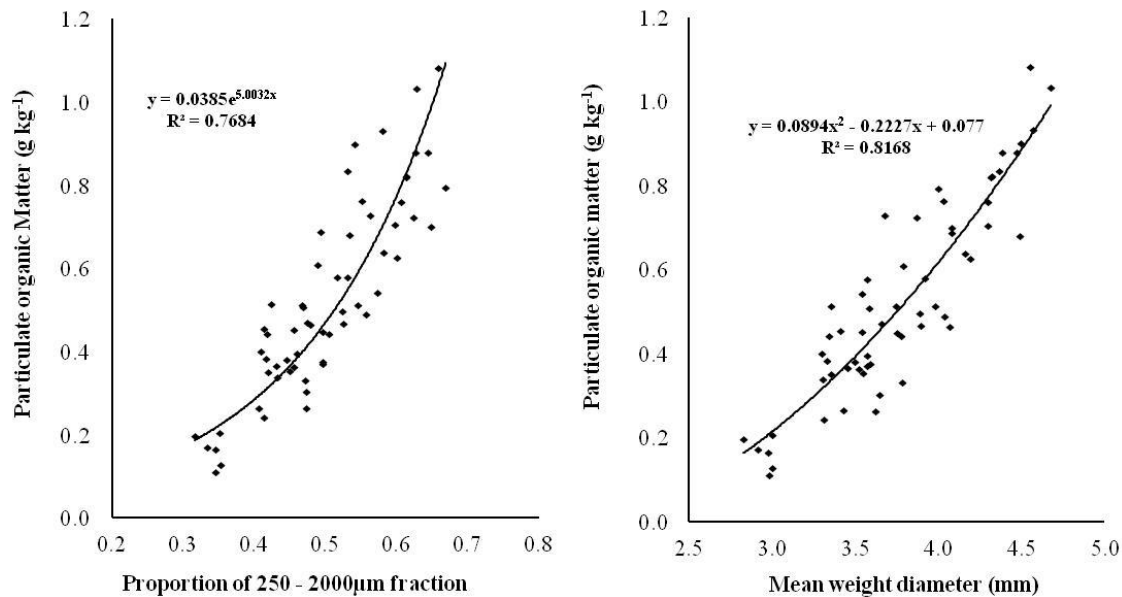
Figure 2. Relationships between particulate organic matter (POM) with a) proportional weights of 250-2000 μm aggregates fraction and b) mean weight diameter (MWD)
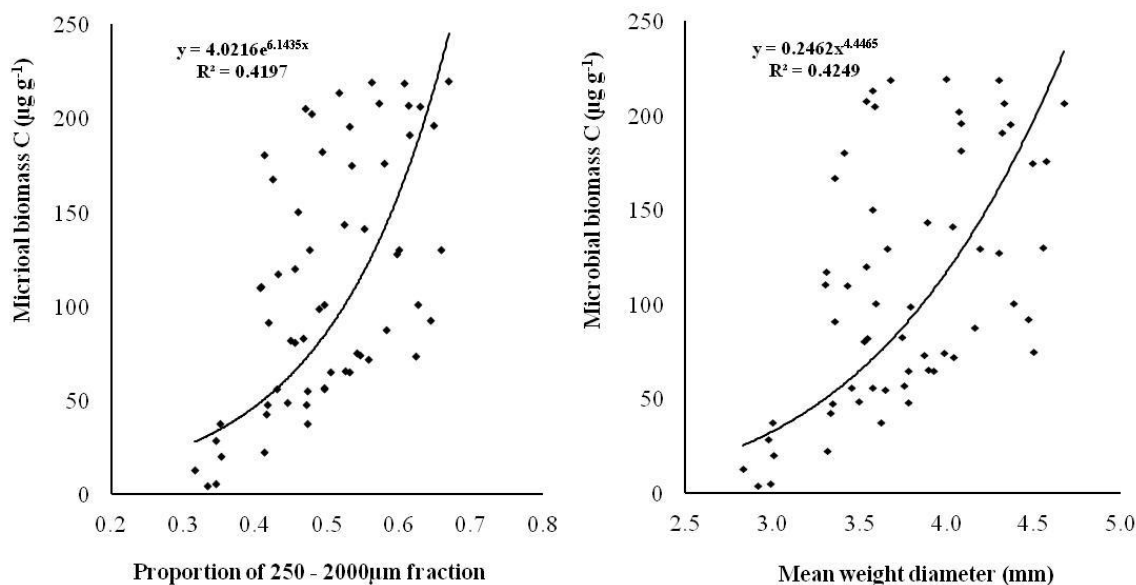

Figure 3. Relationships between microbial biomass carbon (MBC) with a) proportional weights of 250-2000 μm aggregates fraction and b) mean weight diameter (MWD)

Small macro-aggregates fraction were found to be positively correlated with POM (r=0.9104, p=0.001), MBC (r=0.5474, p=0.01), and SOC (r=0.3654, p=0.05) and root biomass (r=0.4977, p=0.01) but other fractions (>2000, 53-250 and <53 μm) were negatively correlated with the binding agents (Table 4). This agrees with other studies that have reported strong correlation of soil aggregate stability with POM in different soils (Six *et al.*, 2002; Franchini *et al.*, 2007) and due to the sensitivity of this parameter, POM has been used in previous studies as an indicator of changes caused

by soil and crop management (Leifeld, *et al.*, 2005; Franchini, *et al.*, 2007).

Recent work elsewhere has also shown strong positive links between root biomass and aggregation suggesting that changes in root biomass alters the structure of soil food webs, changing below ground C cycling and the mean residence time of different SOC pools (Reid *et al.*, 2012). It is generally understood that formation of larger aggregates is enhanced by fine roots and fungal hyphae, while micro-aggregates are stabilized by long-chained organic compounds (e.g.,

polysaccharides) and fungal hyphae. In our study, correlations of root biomass with MWD and the 250-2000 μm size fraction were significant and positive (Table 4) which supports the hypothesis that roots act as temporary binding agents which aid in stabilizing larger aggregates (Tisdall and Oades 1982).

Larger size fractions (>2000 μm), micro-aggregates (53-250 μm) and the silt+ clay (<53 μm) fractions were negatively correlated with root biomass. According to hierarchical theory of soil aggregation, binding of micro-aggregates into macro-aggregates occurs through the entanglement by roots and fungal hyphae, particularly vascular arbuscular mycorrhiza (VAM) hyphae (Tisdall and Oades 1982; Bearden, 2001). The production of mucigel, rhizo-deposition, increases of poly-cations in the rhizosphere, and soil water extraction by plant roots have been implicated in the formation of soil aggregates (Perfect *et al.*, 1990). Root and hyphal growth stimulate microbial activity and simultaneously promote the formation of macro-aggregates (Denef, *et al.*, 2007). Aggregates up to <1000 μm are predominantly assembled by fungal hyphae, mechanically through entanglement of soil particles and chemically with glue-like metabolites (Bearden 2001). Pohl *et al.* (2009) found a positive and significant correlation between root length density and soil aggregate stability. Similarly, Reid *et al.* (2012) have reported strong positive links between root biomass and the abundance of nematodes and several taxa of mesofauna, suggesting that changes in root biomass alters the structure of soil food webs, changing belowground C cycling and the mean residence time of different SOC pools.

The direct influence of roots as the primary C source to soil and particularly to POM is reflected in the significance influence (50.3% relative importance) of root biomass to changes in POM (Table 4). Likewise, root biomass was positively correlated to changes in both MBC (68.3% relative importance) and small micro-aggregates (49.8% relative importance). The greater below ground root biomass of the Brachiaria grasses likely increased microbial activity, stabilizing aggregates through increases in microbially-derived soil binding agents leading to increases in physically protected POM (O'Brien and Jastrow, 2013; Zhang *et al.*, 2013; Zhang *et al.*, 2014). In other studies on soil aggregate stability in agricultural systems, Milne and Haynes (2004), Pohl et al. (2012), Hurisso et al. (2013 and O'Brien and Jastrow (2013) found highest percentages of large aggregates in systems with permanent pasture. The authors showed that the activity of the grass root system, coupled with the absence of soil disturbance, effectively contribute to the formation of stable macro-aggregates. The authors also reported the importance of relations between the MWD and the organic C pools, confirming the statement of Christensen (2001) that, aside from the interactions between minerals, the interaction of MWD with SOC strongly affects the size of water stable aggregates. Soils with higher water stable aggregates are likely to have greater resistance to soil degradation and erosion.

CONCLUSIONS

Aggregate stability in terms of MWD differed among the Brachiaria grasses and was highest in soils under *Mulato II* hybrid and lowest under cv. MG4. This was attributed to the presence of higher POM and MBC in *Mulato II* hybrid cultivated soils. While we found that SOC did not vary among Brachiaria grasses over the short duration of the study, changes in below ground C cycling were apparent through effect on aggregate formation and higher POM and MBC in Brachiaria cultivated soils. By significantly improving soil aggregation and associated C content, the potential of Brachiaria grasses for enhancing C storage was noted.

Acknowledgement

Table 3. Effect of grass types on soil aggregation, MWD, POM, SOC and MBC

Grass type	Proportion of aggregate size fraction				MWD (mm)	POM (gkg^{-1})	SOC (gkg^{-1})	MBC (µgg^{-1})
	>2000 µm	250-2000 µm	53-250 µm	<53µm				
B. decumbens cv. Basilisk	0.002	0.569	0.310	0.103	4.08	0.65	13.97	145.3
B. humidicola cv. Llanero	0.002	0.488	0.416	0.101	3.73	0.46	13.82	122.3
B. brizantha cv. Marandu	0.003	0.604	0.319	0.099	4.31	0.77	14.13	147.8
B. brizantha cv. MG4	0.001	0.432	0.441	0.120	3.43	0.37	13.87	86.5
B. brizantha cv. Xaraes	0.002	0.516	0.354	0.089	3.81	0.66	14.24	134.3
B. brizantha cv. Piatã	0.001	0.461	0.419	0.126	3.58	0.38	13.86	105.8
B. hybrid cv. Mulato II	0.001	0.642	0.285	0.090	4.49	0.93	13.99	154.2
Pennisetum purpureum cv. KK1	0.002	0.519	0.378	0.104	3.87	0.51	13.97	127.4
Chloris gayana cv. KAT R3	0.002	0.422	0.440	0.140	3.38	0.35	14.07	69.3
Bare plot	0.002	0.341	0.482	0.176	2.95	0.16	12.09	18.3
Lsd (p≤0.05)	NS	0.046	0.038	0.014	0.18	0.08	0.96	24.0
CV (%)	-	4.9	6.2	10.4	4.1	12.3	6.0	19.6

MWD= Mean weight diameter; POM= Particulate organic matter, SOC = Soil organic carbon; MBC= Microbial biomass carbon, Lsd= Fischer's protected least significant difference; NS= Not significant, Cv = coefficient of variation

Table 4. Relationships between soil aggregate fractions and MWD with other soil and plant properties

Properties	>2000 µm	250-2000 µm	53-250 µm	<53 µm	MWD mm	POM gkg^{-1}	MBC µgg^{-1}	SOC gkg^{-1}	Total N gkg^{-1}	Root biomass gm^{-2}
>2000 µm	-									
250-2000 µm	0.0069	-								
53-250 µm	-0.0030	-0.8942***	-							
<53 µm	0.0238	-0.7735***	0.6688**	-						
MWD mm	0.0085	0.9950***	-0.8457***	-0.7618***						
POM gkg^{-1}	-0.0167	0.9104***	-0.8355**	-0.7636***	0.8991***	-				
MBC µgg^{-1}	-0.1419	0.5474**	-0.4917**	-0.6415**	0.5371**	0.6469**	-			
SOC gkg^{-1}	0.1243	0.3654*	-0.3229*	-0.4907**	0.3577*	0.3357*	0.2774*	-		
Total N gkg^{-1}	0.1527	0.2861*	-0.2807*	-0.4207*	0.2727*	0.2922*	0.3351*	0.8670***	-	
Root biomass gm^{-2}	-0.0754	0.4977**	-0.4553*	-0.6590**	0.4841**	0.5034**	0.6833**	0.4654*	0.5017**	-

*** p < 0.001, ** p < 0.01 and * p < 0.05, MWD-Mean weight diameter, POM-Particulate organic matter, MBC-microbial biomass carbon, SOC-Soil organic carbon

REFERENCES

Ashagrie, Y., Zech, W., Guggenberger, G., Mamo, T., 2007. Soil aggregation, and total and particulate organic matter following conversion of native forests to continuous cultivation in Ethiopia. Soil Tillage Research 94: 101-108.

Bearden B. M., 2001. Influence of arbuscular mycorrhizal fungi on soil structure and soil water characteristics of vertisols. Plant Soil 229: 245–258.

Blake, G.R., Hartge, K. H., 1986. Bulk density. In: Klute, A. (Ed.), Methods of Soil Analysis. Part 1 Physical and Mineralogical Methods, 2nd edition. America Society of Agronomy, Madison, WI, pp. 363–375.

Bolinder, M. A., Angers, D. A., Bélanger, G., Michaud, R., Laverdière, M. R., 2002. Root biomass and shoot to root ratios of perennial forage crops in eastern Canada. Canadian Journal Plant Science 82: 731–737.

Broersma, K., Robertson, J. A., Chanasyk, D. S., 1997. The effects of diverse cropping systems on aggregation of a Luvisolic soil in the Peace River region. Canadian Journal Soil Science 77: 323–329.

Bronick, C. J., Lal, R., 2005. Soil structure and management: a review. Geoderma 124: 3–22.

Cambardella, C. A., Elliott, E. T., 1992 Particulate soil organic-matter changes across a grassland cultivation sequence. Soil Science Society of American Journal 56: 777–783.

Chenu, C., Le Bissonnais, Y. and Arrouays, D., 2000, Organic matter influence on clay wettability and soil aggregate stability. Soil Science Society of American Journal 64: 1479-1486.

Christensen, B. T., 2001. Physical fractionation of soil and structural and functional complexity in organic matter turnover. European Journal of Soil Science 52: 345–353.

Clapperton, M. J., Chan, K. Y., Larney, F. J. 2003 Managing the soil habitat for enhanced biological fertility; Soil Biological Fertility – a key to sustainable land use in agriculture (Eds. L. K. Abbott and D. V. Murphy); Dordrecht, Netherlands; Kluwer Academic; pp. 203-224.

Denef, K., Zotarelli, 1., Boddey, R. M., Six, J., 2007. Micro-aggregates-associated carbon as a diagnostic fraction for management-induced changes in soil organic carbon in two oxisols. Soil Biology and Biochemistry 39: 1165 – 1172.

Dungait, J. A., Hopkins, D. W., Gregory, A. S., Whitmore, A. P., 2012. Soil organic matter turnover is governed by accessibility not recalcitrance. Global Change and Biology 18: 1781–1796.

Fisher, M. J., Braz, S. P., Dos Santos, R.S.M., Urquiaga, S., Alves, B. J. R., Boddey, R.M., 2007. Another dimension to grazing systems: soil carbon. Tropical Grassland 41: 65-83.

Fonte, S. J., Nesper, M,, Hegglin, D,, Velásquez, J. E., Ramirez, B., Rao, I. M., Bernasconi, S. M., Bünemann, E. K., Frossard, E., Oberson, A., 2014. Pasture degradation impacts soil phosphorus storage via changes to aggregate-associated soil organic matter in highly weathered tropical soils. Soil Biology and Biochemistry 68: 150-157.

Franchini, J. C., Crispino, C. C., Souza, R. A., Torres, E., Hungria, M., 2007. Microbiological parameters as indicators of soil quality under various soil management and crop rotation systems in southern. Brazil Soil Tillage Research 92: 18–29.

Gale, W.J., Cambardella, C.A., Bailey, T. B., 2000. Root-derived carbon and the formation and stabilization of aggregates. Soil Science Society of American Journal 64: 201–207.

Gartzia-Bengoetxea, N., Gonz´alez-Arias, A., Kandeler, E., Mart´inez de, I. A. 2009. Potential indicators of soil quality in temperate forest ecosystems: a case study in the Basque Country. Annals of Forest Science 66: 1-12.

GENSTAT Release 4.24DE 2005. Lawes Agricultural Trust (Rothamsted Experimental Station), UK.

Gicheru, P., Gachene, C., Mbuvi, J., and Mare, E. 2004. Effects of soil management practices and tillage systems on surface soil water conservation and crust formation on a sandy loam in semi-arid Kenya. Soil Tillage Research, 75: 173-184.

Grandy, A. S., Robertson G. P., 2006. Aggregation and organic matter protection following tillage of a previously uncultivated soil. Soil Science Society of American Journal 70: 1398–1406.

Gulde, S., Chung, H., Amelung, W., Chang, C., Six, J., 2008. Soil carbon saturation controls labile and stable carbon pool dynamics. Soil Science Society of America Journal 72: 605–612.

Harris, R.F., Chesters, G., Allen, O.N., 1996. Dynamics of soil aggregation Advances in Agronomy 18: 107-169.

Harwood, R. R., Cavigelli, M. A., Deming, S. R., Frost, L.A., Probyn L. K. (eds.) 1998. Michigan Field Crop Ecology: Managing Biological Processes for Productivity and Environmental Quality. Michigan State University Extension Bulletin E-2646, 92 pp.

Holeplass, H., Singh, B. R., Lal, R., 2004. Carbon sequestration in soil aggregate under different crop rotations and nitrogen fertilization in an Inceptisol in south eastern Norway. Nutrient Cycling in Agroecosystems, 70: 167-177.

Hurisso, T. T., Davis, J. G., Brummer, J. E., Stromberger, M. E., Mikha, M., Haddix, M. L., Booher, M. R., Paul, E. A., 2013. Rapid changes in microbial biomass and aggregate size distribution in response to changes in organic matter management in grass pasture. Geoderma 194: 68-75.

Jones, D. L., Nguyen, C., Finlay, R. D., 2009. Carbon flow in the rhizosphere: carbon trading at the soil–root interface. Plant and Soil 321: 5–33.

Kemper, W. D., Rosenau, R. C., 1986. Aggregate stability and size distribution In: Klute, A. (Ed.), Methods of Soil Analysis Part 1 Physical and Mineralogical Methods 2nd Edition. American Society of Agronomy, Madison, WI, USA. p. 377–382.

Lal, R., Follet, R. F. and Kimble, J. M., 2003. Achieving soil carbon sequestration in the United States: A challenge to policy makers. Soil Science, 168: 827-845.

Landon, J. R., 1991. Booker Tropical Soil Manual: A Handbook of Soil Survey and Agricultural Land Evaluation in the Tropics and Subtropics; Longman Scientific Technical and Booker Tate Limited: London, U.K.

Leifeld, J., Kögel-Knabner, I., 2005. Soil organic matter fractions as early indicators for carbon stock changes under different land-use. Geoderma 124: 143–155.

Marquez C. O, Garcia V. J, Cambardella C. A, Schultz R. C, Isenhart T. M., 2004. Aggregate size stability distribution and soil stability. Soil Science Society of American Journal 68: 725-735.

Mikha, M. M., Rice, C. W., 2004. Tillage and manure effects on soil and aggregate-associated carbon and nitrogen. Soil Science Society of American Journal 68: 809–816.

Mills, A. J., Fey, M. V., 2003. Effects of land use on soil organic matter and surface crusting. South Africa Journal of Science 99: 429-436.

Milne, R. M., Haynes, R. J., 2004. Comparative effects of annual and permanent dairy pastures on soil physical properties in the Tsitsikamma region of South Africa. Soil Use and Management 20: 81–88.

Min, D. H., Islam, K. R., Vough, L. R., Weil, R. R., 2003. Dairy manure effects on soil quality properties and carbon sequestration in alfalfa-orchard grass systems. Communication Soil Science Plant Analysis 34: 781–799.

Murphy, J. and Riley, J. P., 1962. A modified single solution method for determination of phosphate in natural waters. Analitica Chimica Acta, 27: 31-36.

NAAIAP 2014. Soil suitability evaluation for maize production in Kenya. A report by National Accelerated Agricultural Inputs Access Programme (NAAIAP) in collaboration with Kenya Agricultural Research Institute (KARI) Department of Kenya Soil Survey, Kilimo, Nairobi, Kenya.

Nelson, D. W., Sommers, L.E., 1982. Total C, organic C and organic matter. Methods of soil analysis 2, Chemical and Microbiological Properties. Agronomy, 9: 539-579.

O'Brien, S. L., Jastrow, J. D., 2013. Physical and chemical protection in hierarchical soil aggregates regulates carbon and nitrogen recovery in restored perennial grasslands. Soil Biology and Biochemistry 61: 1–13.

Okalebo, J. R., Gathua, K. W., Woomer, P. L. 2002. Laboratory Methods of Soil and Plant Analysis: A Working Manual. TSBF-KARI, SSEA, SACRED Africa, Nairobi, Kenya, pp. 128.

Perfect, E., Kay, B. D., van Loon., W.K.P., Sheard, R.W., Pojasok, T. 1990. Rate of change in soil structural stability under forages and corn. Soil Science Society of American Journal 54: 179-186.

Peters, M., Rao, I., Fisher, M., Subbarao, G., Martens, S., Herrero, M., van der Hoek, R., Schultze, K.R., Miles, J., Castro, A., Graefe, S., Tiemann, T., Ayarza, M. and G. Hyman, G. 2012. Chapter 11.Tropical forage-based systems to mitigate greenhouse gas emissions. In: Hershey, C. (Ed.) Issues in Tropical Agriculture I. Eco-Efficiency: From Vision to Reality.CIAT, Cali, Colombia. http://www.ciat.cgiar.org/publications/eco

Pohl, M., Alig, D., Körner, C., Rixen, C. 2009. Higher plant diversity enhances soil stability in disturbed alpine ecosystems. Plant Soil 324: 91-102.

Pohl, M., Graf, F., Buttler, A., Rixen C. 2012. The relationship between plant species richness and soil aggregate stability can depend on disturbance. Plant Soil 355: 87–102.

Reid, J. B. and Goss, M. J., 1980. Changes in the aggregate stability of a sandy loam effected by growing roots of perennial ryegrass. Journal of the Science of Food and Agriculture 31: 325-328.

Reid, J., Adair, E.C., Hobbie, S.E., Reich, P.B., 2012. Biodiversity, nitrogen deposition and CO_2 affect grassland soil carbon cycling but not storage. Ecosystems 12: 580–590.

Six J, Bossuyt H, Degryze S, Denef K., 2004. A history of research on the link between (micro)aggregates, soil biota, and soil organic matter dynamics. Soil Tillage Research 79: 7–31.

Six J, Elliot E. T, Paustian K, Doran J. W., 1998. Aggregation and soil organic matter accumulation in cultivated and native grassland soils. Soil Science Society of American Journal 63: 1350–1358.

Six, J., Feller, C., Denef, K., Ogle, S. M., Sa, M. J. C., Albrecht, A., 2002. Soil organic matter, biota and aggregation in temperate and tropical soils effects of no tillage. Agronomy 22: 755-775.

Sommerfeldt, T. G., Chang, C. 1985. Changes in soil properties under annual applications of feedlot manure and different tillage practices. Soil Science Society of American Journal 49: 983-987.

Spohn, M., Giani, L., 2011. Impacts of land use change on soil aggregation and aggregate stabilizing compounds as dependent on time. Soil Biology and Biochemistry 43: 1081–1088.

Tisdall, J. M., Oades, J., M., 1982. Organic matter and water stable aggregates in soils. Journal of Soil Science 33: 141-163.

Todd A. O., Cambardella, C. A., Schulte, L. A., Kolka, R. K. 2015. Factors influencing soil aggregation and particulate organic matter responses to bioenergy crops across a topographic gradient. Geoderma 255: 1–11.

Vance, E. D., Brookes, P. C., Jenkinson, D. S., 1987. An extraction method for measuring soil microbial biomass C. Soil Biology and Biochemistry 19: 703–707.

Walker, T. S., Bais, H. P., Grotewold, E., Vivanco, J. M., 2003. Root exudation and rhizosphere biology. Plant Physiology 132: 44–51.

Whalen, J. K., Hu, Q., Lui, A., 2003. Compost applications increase water-stable aggregates in conventional and no-tillage systems. Soil Science Society of America Journal 67: 1842–1847.

Wick A. F., Ingram L. J., Stahl P. D., 2009. Aggregate and organic matter dynamics in reclaimed soils as indicated by stable carbon isotopes. Soil Biology and Biochemistry 41: 201–209.

Wortmann, C. S., Shapiro, C. A., 2008. The effects of manure application on soil aggregation. Nutrient Cycling in Agroecosystems 80: 173–180

Zhang, S., Li, Q., Lu, Y., Zhang, X., Liang, W., 2013. Contributions of soil biota to C sequestration varied with aggregate fractions under different tillage systems. Soil Biology and Biochemistry 62: 147–156.

Zhang, X., Wua, X., Zhang, S., Xing, Y., Wang, R., Liang, W. 2014. Organic amendment effects on aggregate-associated organic C, microbial biomass C and glomalin in agricultural soils. Catena 123: 188–194.