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Title: Fertilizer and residue quality effects on organic matter stabilization in soil aggregates

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ABSTRACT

This study examined the influence of organic residue quality and N fertilizer on aggregate-associated SOM in maize cropping systems of southern Ghana. Six residue treatments of differing quality (*Crotalaria juncea*, *Leucaena leucocephala*, maize stover, sawdust, cattle manure and a control with no residues added) were applied at 4 Mg C ha^{-1} yr^{-1} both with and without fertilizer N additions (120 kg N ha^{-1} $season^{-1}$). Soils (0-15cm) were sampled three years after study implementation and wet sieved into four aggregate size classes (8000–2000, 2000–250, 250–53, and <53 μm). Small macroaggregates (2000–250 μm) were further separated into coarse particulate organic matter (>250 μm), microaggregates within macroaggregates (53-250 μm), and macroaggregate occluded silt and clay (<53 μm). N fertilizer additions reduced aggregate stability as was evident from a 40% increase in the weight of the silt and clay fraction ($P = 0.014$) as well as a decrease in microaggregates across all residue types ($P = 0.019$). Fertilizer similarly affected C and N storage within these aggregate fractions, while effects of residue quality were largely insignificant. Our results suggest that fertilizer effects on soil aggregation may have important implications for long-term SOM dynamics.

ABBREVIATIONS: SOM, Soil Organic Matter

INTRODUCTION

Food insecurity and declining soil fertility across much of sub-Saharan Africa in recent decades have lead to the pursuit of alternative nutrient management strategies for both improving crop yields and the restoration of degraded soils (Sanchez, 2002). These new strategies seek to address the limited availability of inorganic N (fertilizer) inputs and rely more upon the management of organic resources that are available to farmers (Palm et al., 2001). The combined use of inorganic and organic nutrient sources in particular has been put forth as means to improve crop yields (Vanlauwe et al., 2001; Kramer et al., 2002), via improved synchronization of nutrient availability (controlled by fertilizer input and residue nutrient release) and plant uptake, and to reduce soil organic matter (SOM) depletion (Vanlauwe et al., 2001; Bationo et al., 2007). As a key component of agricultural sustainability, SOM contributes greatly to improving soil structure, fertility and water relations (Tiessen et al., 1994; Craswell and Lefroy, 2001), and plays a central role in greenhouse gas mitigation efforts (Paustian, 2002; Lal, 2004). The maintenance of SOM may be of particular importance for tropical agroecosystems, where the predominance of 1:1 clays (and the associated low cation exchange capacity) increases the reliance on SOM for both the retention and supply of plant available nutrients by the soil (Craswell and Lefroy, 2001; Oorts et al., 2003). Although abundant research has focused on how plant litter quality affects decomposition and nutrient mineralization (Palm and Sanchez, 1991; Constantinides and Fownes, 1994; Vanlauwe et al., 2005), little is known about how the quality of residue inputs influences the formation of stable SOM pools or how residues might interact with inorganic fertilizer to affect long-term SOM dynamics.

Along with biochemical recalcitrance and the formation of organo-mineral complexes, the physical protection of SOM within soil aggregates is considered a fundamental process governing SOM turnover (Tisdall and Oades, 1982; Six et al., 2002a). Although the linkages between SOM and soil structure may be stronger in temperate soils, aggregation remains an important mechanism for SOM stabilization in tropical soils as well (Feller and Beare, 1997; Six et al., 2002b). A number of studies have identified organic matter incorporation into microaggregates and more specifically the formation of microaggregates (53-250 μm) within macroaggregates (>250 μm) as a key pathway for the long-term stabilization of SOM (Angers et al., 1997; Six et al., 2000). A model put forth by Six et al. (2000) suggests that by accelerating the turnover of macroaggregates, tillage disrupts microaggregate formation and SOM stabilization associated with this soil fraction. In accordance with these concepts, the microaggregate within macroaggregate fraction has been offered as a sensitive, early indicator of management effects on SOM dynamics (Kong et al., 2005; Denef et al., 2007). Similar to the role of tillage, several studies have indicated that residue quality and nutrient additions may influence aggregate dynamics as well (Harris et al., 1963; Bossuyt et al., 2001; Six et al., 2001). For example Six et al. (2001) attributed the faster turnover of macroaggregates under the N-fixing *Tifolium repens* (versus *Lolium perenne* with higher C:N ratio) to differences in plant residue quality. Similarly, both Harris et al. (1963) and Bossuyt et al. (2001) found inorganic N additions to decrease aggregate stability in incubation experiments. Based on these preliminary findings it seems that aggregate turnover (and associated SOM dynamics) may be governed to some extent by soil N availability. We therefore hypothesized that labile N sources (i.e., fertilizer and high quality residues) would accelerate aggregate turnover by increasing the decay rate of organic aggregate binding agents, potentially leading to a decrease in aggregate associated SOM stabilization.

The objective of this study was to investigate how both residue quality and N fertilizer inputs affect aggregate dynamics and C stabilization in low-input agricultural systems (i.e., systems with low chemical inputs and minimal tillage). Furthermore, we aimed to examine interactions between residue and fertilizer for providing best management options to resource-poor farmers in the region.

MATERIALS AND METHODS

Site description, experimental design and trial management

The experiment was established in 2002 on a cleared semi-deciduous forest site at the Soil Research Institute at Kwadaso, Ghana, ($6^{0}40^{/}$ N, 1^{0} $40^{/}$W). The soil is developed on phyllite parent material and is classified as a Ferri-Plinthic Acrisol according to the World Reference Base (FAO, 1998) or a Lithic Hapludult under the U.S. taxonomic system (Soil Survey Staff, 2006). This soil is considered a loam (sand = 46%, silt = 40%, clay = 14%) with an organic C content of 1.2% and pH of 5.4. The experimental site has a bimodal rainfall pattern (1200 mm yr^{-1}) with the major season occurring between April and July and the minor season between September and December. The trial follows a split-plot design with six residue treatments at two levels of synthetic N fertilization each present in one of three replicate blocks. The residues encompass a wide range of litter quality (Table 1) and represent potential organic amendments available in the region. The treatments consist of one of five residues (*Crotalaria juncea* L., *Leucaena leucocephala* Lam., maize stover - *Zea mays* L., sawdust of *Afzelia africana* Sm. and manure derived from maize-fed cattle) added at a rate of 4 Mg C ha^{-1} yr^{-1}, plus a control with no residues applied. Each of these plots is then divided into two fertilizer sub-plots receiving 0 or 120 kg N ha^{-1} $season^{-1}$ as urea. All plots receive an annual application of 30 kg P ha^{-1} yr^{-1} in the form of triple super phosphate and 60 kg K ha^{-1} yr^{-1} as muriate of potash, based on local recommendations for maize. Two crops of maize are grown each year (major and minor seasons) within each 6.4 m x 3.2 m subplot. Organic inputs are surface applied and incorporated minimally with a hoe to a depth of roughly 10 cm, while above ground crop biomass is removed at the end of each growing season.

Soil sampling and aggregate fractionation

Soils from each plot were sampled in late March of 2006 prior to the fourth residue application from a depth of 0-15 cm in order to include the entire zone of influence associated with residue incorporation. Five soil cores (5-cm diameter) were collected from each plot and combined. Field moist samples were passed through an 8 mm sieve by gently breaking soil clods along natural planes of fracture, and then air dried for subsequent analyses.

A subsample (80g) of air dried soil from each plot was then fractionated by wet-sieving according to Elliot (1986). These subsamples were spread evenly onto a 2000 µm sieve and slaked for 5 minutes in distilled water. The soil was then sieved for 2 minutes by oscillating the sieve 50 times up and down (approximately 3 cm amplitude). Large macroaggregates 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 repeated to obtain the small macroaggregate fraction (250–2000 µm). The sieving process was repeated once more using a 53 µm sieve to separate microaggregates (53–250 µm) from the silt and clay fraction (< 53 µm). All pans and soil solutions were placed in an oven at 60 ºC until dry.

Small macroaggregates were fractionated into three components based on Six et al. (2000). In short, subsamples (10 g) of oven-dried macroaggregates were submerged in distilled water for 20 minutes to induce slaking. Fifty glass beads (4mm dia.) were placed on a 250 µm sieve-mesh affixed to a reciprocal shaker and a slow continuous flow of water was introduced in order to submerge the mesh and beads in roughly 1cm of water. The slaked soil sample was then added to the glass beads on the 250 µm mesh and shaken at low speed. The beads function to break up the macroaggregates, while the flowing water flushes the released macroaggregate components through the 250 µm mesh, thus avoiding further disruption of freed microaggregates. Water and soil (<250 µm) passed to a 53 µm sieve below the reciprocal shaker in order to capture the released microaggregates. Shaking continued until water flowing onto the 53 µm sieve was clear and no aggregates remained on top of the 250 µm mesh. Material left on the 250 µm sieve was rinsed into a pan for drying, while soil transferred to the 53 µm sieve was sieved for 2 minutes as described above for the free aggregate fractions. Material remaining above the 250 µm mesh is classified as coarse particulate organic matter (cPOM), while soil passing through the 250 um mesh was separated into microaggregates-within-macroaggregates (> 53 µm; mM) and macroaggregate associated silt and clay (< 53 µm; s+cM).

Plant and soil analyses

Subsamples of all oven-dried soil fractions were weighed, ground and then analyzed for total C and N concentrations using a PDZ Europa Integra C-N isotope ratio mass spectrometer (Integra, Germany). Total C in all fractions was considered to be organic C, since no carbonates are present in this soil. Similarly, total N effectively represents to organic N, since the relative quantity of mineral N in soils is generally considered to be minimal. Plant residues were collected prior to application, dried at 60 ºC and analyzed for total C, N, lignin and soluble polyphenols (Anderson and Ingram, 1993).

Statistical analyses

Differences in aggregation and C content within each fraction were compared across the six residue treatments and two N application rates using a mixed model ANOVA approach to a randomized split-plot design. Residue, fertilizer and the residue x fertilizer interaction were considered fixed effects, while block and block x residue were treated as random. Natural log transformations were applied as necessary in order to meet the assumptions of ANOVA. Individual comparisons between residue treatments were carried out using Tukey's honest significant difference. All analyses were conducted using JMP 7.0 statistical software (SAS Institute Inc, 2007).

RESULTS

Influence of N fertilizer on aggregation and aggregate associated C

N fertilizer (inorganic N) yielded the most notable effects in this study. Although no significant influence of fertilizer or residue class was observed for total soil C (Fig. 1) or N, there were clear effects of fertilizer on both soil aggregation (Fig. 2) and aggregate associated C (Fig. 3) and N pools. Fertilizer increased the percentage of total soil accounted for by the silt and clay fraction across all residue treatments (P = 0.014), from 14.3 % in the 0 kg N ha^{-1} plots to 20.0% of total soil when fertilizer was applied (Fig. 2). This trend corresponded with a decrease in the contribution of microaggregates (from 40.3% to 37% of total soil; P = 0.019) and a consistent, yet non-significant (P > 0.1) tendency of small macroaggregates to decrease with fertilizer application as well (Fig. 2). The large macroaggregate contribution to the overall soil mass was relatively small (5.7% on average) and exhibited high variability.

Fertilizer induced changes in aggregation also resulted in altered distributions of C and N within soil fractions. The quantity of C found in the free silt and clay fraction increased by over 40% on average with fertilizer addition ($P = 0.006$), from 1.82 to 2.73 mg g^{-1} total soil (Fig. 3), while N increased by over 70% ($P < 0.001$) in this fraction (data not shown). At the same time, fertilizer additions reduced N in the microaggregate fraction from 0.44 to 0.39 mg g^{-1} soil ($P = 0.036$; data not shown). Synthetic fertilizer also increased C and N contained within the cPOM fraction ($P = 0.006$ & $P = 0.002$, respectively) through enrichment of the C and N concentrations of this fraction (Fig. 4; N data not shown).

Residue quality effects on aggregate associated C

Residue quality effects on aggregation and SOM dynamics were generally absent compared to those observed for fertilizer additions. Residue quality affected the contribution of free microaggregates to total soil N, with this fraction containing 0.53 mg N g^{-1} soil under *C. juncea* vs. 0.36 mg N g^{-1} with sawdust (significant at the $P = 0.058$ level). However, no significant influence of residue quality was observed for any of the other soil fractions. Furthermore, no interactive effects between fertilizer and residue quality were apparent for any aggregate fraction.

DISCUSSION

The addition of mineral fertilizers and/or plant residue is vital for sustaining long-term productivity in most agricultural systems; however, further evaluation regarding the influence of these inputs on SOM dynamics is needed to better assess the overall sustainability of such practices. In this study the addition of N fertilizer yielded the most consistent effects on aggregate-associated SOM pools. Although the effects were not drastic, an apparent reduction in aggregate stability associated with fertilizer addition causes some concern for long-term soil C dynamics in this system. Reduced aggregation (most evident from the significant increase in the silt and clay fraction) indicates a decrease in aggregate stability, thus suggesting an increase in aggregate turnover rates and a potential decrease in SOM stabilization. These results essentially support our hypothesis and suggest that synthetic N fertilizer plays a less pronounced, yet similar role to that of tillage as outlined by Six et al. (2000). However, alterations to aggregate turnover would, here, be attributed to the accelerated breakdown of organic binding agents within aggregates, rather than the mechanical forces of tillage.

The apparent decrease in aggregate stability observed with N fertilization in this experiment contrasts with other studies where aggregation was found to increase following fertilizer additions (Hati et al., 2006). These studies, however, suggest that an increase in organic matter inputs (due to increased crop growth with fertilizer application) fostered an increase in aggregation, while in the present study aboveground crop residues were removed, thus eliminating a large part of the potential differences in biomass inputs between fertilized and unfertilized plots. Sarakar et al. (2003), on the other hand, found fertilizer additions to decrease aggregate stability in a field trial, despite increases in aboveground production and subsequent residue inputs. Findings of Bossuyt et al. (2001) from an incubation experiment also agree with results from this study, in that they found aggregation to decrease where wheat straw was added to soil along with inorganic N versus soil and wheat straw alone. In another incubation study, Harris et al. (1963) found inorganic N additions to induce aggregate breakdown in a soil with low initial N content (0.06% N), but not in a high N soil (0.24% N). In addition to differences in soil N availability, they suggest factors such as mineralogy, soil texture and organic substrate quality may govern the effect of fertilizer N additions on soil aggregate dynamics (Harris et al., 1963).

Although not significant ($P > 0.1$), a general decrease in total soil C across all residue treatments, with the exception of the control (Fig. 1), may indicate that the influence of fertilizer on aggregate stability observed in this study could eventually lead to reductions in SOM stores and thus have long-term consequences for agroecosystem sustainability. Many studies suggest that fertilizer additions may increase SOM stores due to an increase in organic matter inputs (crop residues) associated with improved crop growth (Paustian et al., 1997; Follett, 2001). Thus, the mineral N induced destabilization and loss of SOM indicated in this study may simply result from the removal of aboveground crop residues at the field site and a subsequent lack of large differences in residue inputs between fertilizer treatments. Such a situation would be relevant since removal of aboveground biomass is a common practice in this region as well as in other parts of sub-Saharan Africa where this material is used to feed livestock. However, a recent study by Khan et al. (2007) reports fertilizer induced losses of SOM across a large number of field trials, regardless of alterations to residue inputs. Management induced changes to total soil C often take many years to become apparent, while the treatments compared in this study represent only three years of differing nutrient inputs. Thus, we might expect losses of soil C associated with fertilizer application to become significant in the years to come at this site.

Effects of residue quality on aggregation and aggregate-associated C and N pools in this study were small and largely insignificant despite large associated differences in N inputs. The absence of differences along with the lack of an interaction between fertilizer and residue quality treatments suggests that residue quality is not a major determinant of aggregate dynamics at this site. Although a few studies indicate that organic resource quality might be important for aggregation (Harris et al., 1963; Six et al., 2001), the evidence is far from conclusive. The findings presented here generally agree with those of Gentile et al. (2008) from two experiments of similar design in Kenya, in that they found little effect of residue quality. Likewise, residue quality effects on SOM fractions reported by Mapfumo et al. (2007) were largely attributed to difference in the size of the organic material added to the soil, but not an effect of residue 'quality', or chemical recalcitrance, as discussed by many authors (Palm et al., 2001; Vanlauwe et al., 2005). Furthermore, Bossuyt et al. (2001) found no difference in aggregate stability for soils incubated with either low or high quality plant residues. Thus, it seems that although residue quality may hold some significance for aggregation processes, other management factors (i.e., tillage, fertilizer application, and irrigation) are probably of greater importance.

CONCLUSIONS

This study provides useful information towards the development of responsible nutrient management strategies in low input cropping systems. Although the influence of residue quality demonstrated here seems to be relatively weak, conclusive results are still lacking. Of greater consequence, N fertilizer appears to decrease aggregate stability and the capacity of this soil to stabilize SOM in soil aggregates, thus highlighting the potential for fertilizer applications to impact long term SOM storage.

Table 1: Residue quality parameters from plant derived materials collected at Kwadaso, Kumasi, Ghana, 2005.

Residue Type	Residue Quality Class[†]	C	N	Lignin	Polyphenols	C:N ratio
		g kg^{-1}				
C. juncea (leaves)	I	407	33.4	47.8	22.2	12.2
L. leucocephala (leaves)	II	458	51.0	51.3	88.7	9.0
Z. mays stover	III	355	13.0	72.0	13.0	27.3
Cattle manure	III	394	6.0	61.0	0.4	65.7
A. Africana sawdust	IV	489	2.5	255.8	46.0	195.6

[†] Residue quality classes (I = high quality, II and III = intermediate quality and IV = low quality) defined by Palm et al. (2001).

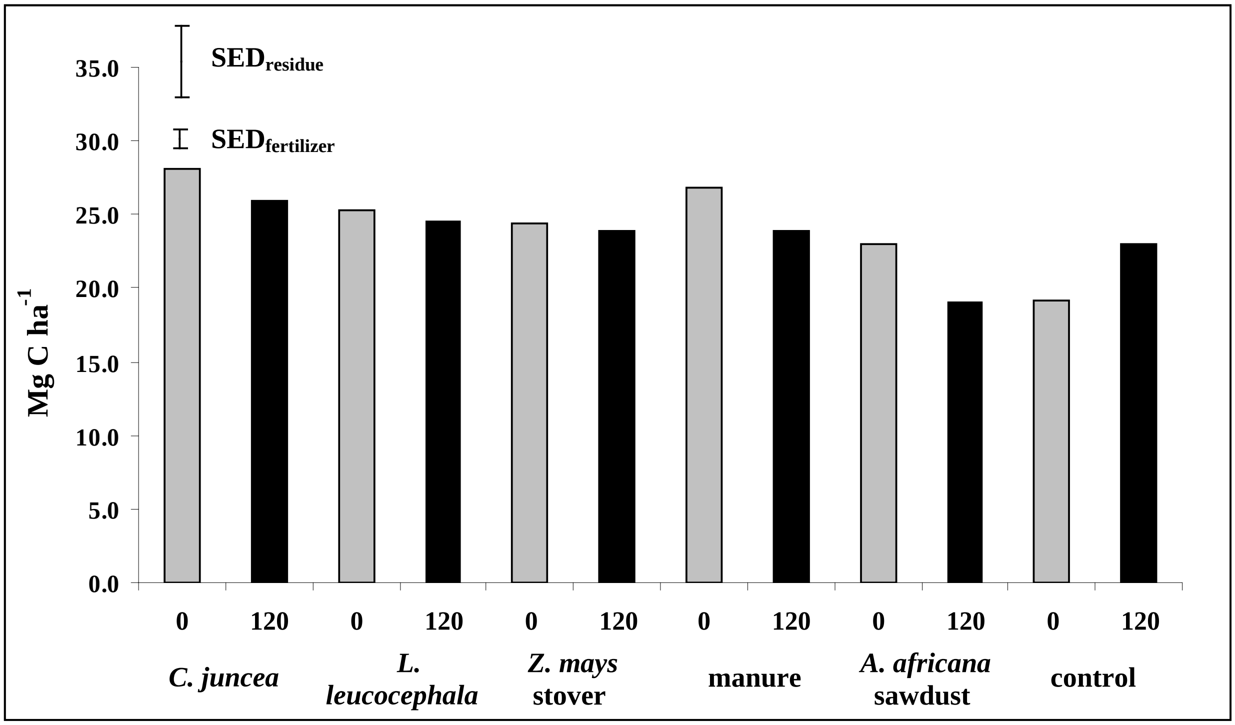

Figure 1: Total soil C (0-15 cm) across residue and fertilizer treatments in March 2006 at Kwadaso, Ghana. 0 and 120 refer to levels of N fertilization in kg N ha^{-1}. Error bars represent standard error of the difference between means.

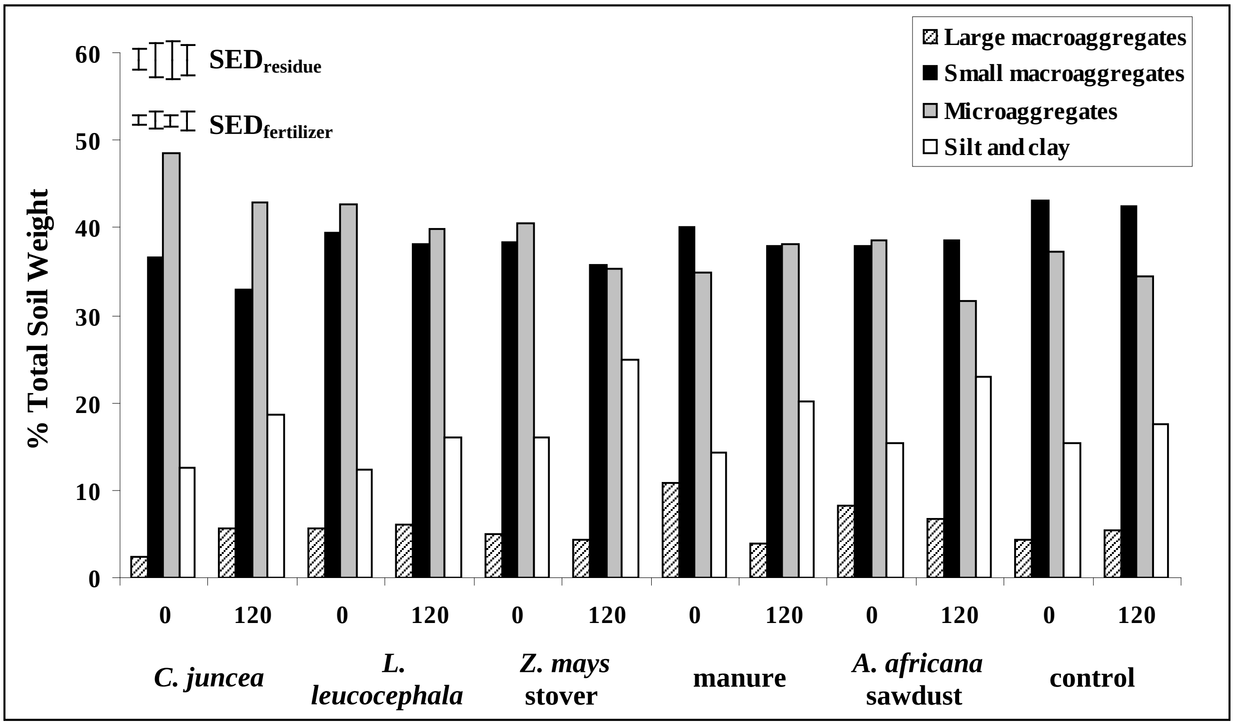

Figure 2: Distribution of free aggregate fractions from soil (0-15 cm) across residue and fertilizer treatments sampled in March 2006 at Kwadaso, Ghana. 0 and 120 refer to levels of N fertilization in kg N ha^{-1}. Error bars represent standard error of the difference between means.

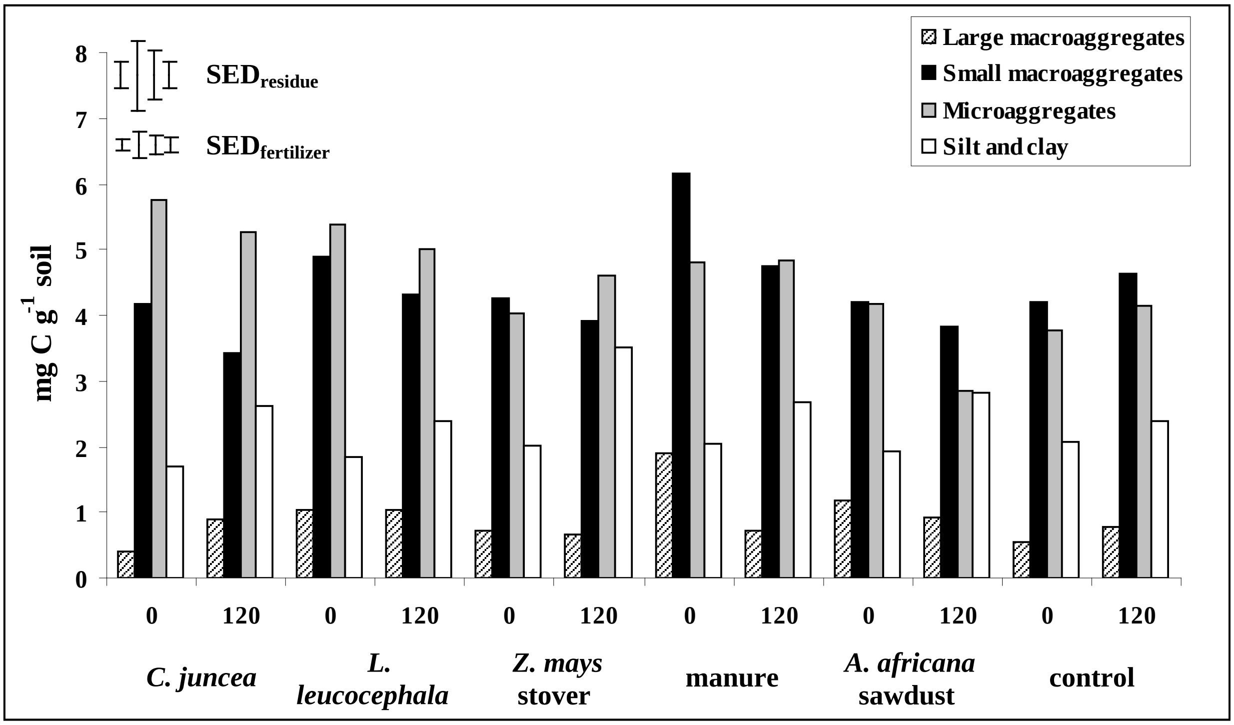

Figure 3: Carbon distribution in free aggregate fractions from soil (0-15 cm) across residue and fertilizer treatments sampled in March 2006 at Kwadaso, Ghana. 0 and 120 refer to levels of N fertilization in kg N ha^{-1}. Error bars represent standard error of the difference between means.

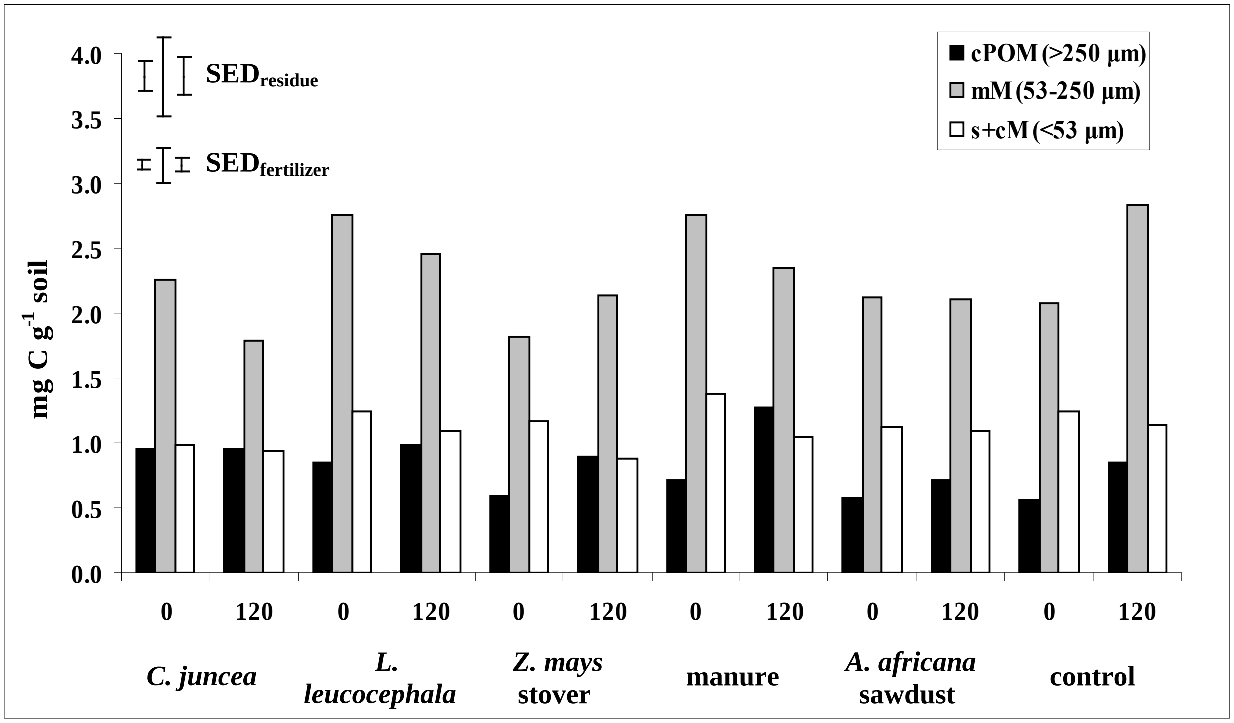

Figure 4: Carbon distribution in small macroaggregate occluded fractions from soil (0-15 cm) across residue and fertilizer treatments sampled in March 2006 at Kwadaso, Ghana. 0 and 120 refer to levels of N fertilization in kg N ha^{-1}. Error bars represent standard error of the difference between means.

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