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## Molecular turnover time of soil organic matter in particle size fractions and soil organic matter alteration velocity of an arable soil through conservation agriculture: A review

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### Abstract

While soil organic carbon (SOC) accumulation and stabilization has been increasingly the focus of ecosystem properties, how it could be linked to soil biological activity enhancement has been poorly assessed. This paper reviewed the soil properties determined and to investigate niche specialization of different soil particle fractions in response to SOM alteration velocity. Conservation Agriculture (CA) can supply SOM, little is known about the temporal resolution of this change. Conventional tillage practices triggered SOM molecular size increases in both aggregates and the fine fraction, whereas switching to CA restored the molecular SOM size of the fine fraction only. Therefore, this fraction can be changed, even in short periods. Soil aggregates were fractionated into different sizes of coarse sand (200-2000  $\mu\text{m}$ ), fine sand (20-200  $\mu\text{m}$ ), silt (2-20  $\mu\text{m}$ ) and clay (< 2  $\mu\text{m}$ ), using separation with a low-energy dispersion protocol. Conservation Agriculture (CA) paradigm that is able to regenerate degraded land, minimize soil erosion, and harness the flow of ecosystem services.

The contribution of each aggregate class to soil C and N stocks changed with depth. Aggregates > 250  $\mu\text{m}$  accounted for a similar percentage of C and N stocks in each layer: 64%, 59% and 50% in the 0-60, 60-200, and 200-400 kg soil  $\text{m}^{-2}$ , respectively. Mid-size aggregates and small aggregates contributed increasingly to C and N stocks with increasing depth, accounting for 29%, 32% and 40% of the C and N stocks as mid-size aggregates and 7%, 9% and 10% as small size aggregates in the 0-60, 60-200, and 200-400 kg soil  $\text{m}^{-2}$  layers, respectively. The DOC concentration is considerably lower than those of other labile C fractions, generally not more than 200  $\text{mg kg}^{-1}$ , but it is the most mobile fraction of SOC. It controls the turnover of nutrient and organic matter by affecting the development of microbial populations. A positive relationship was observed between the  $^{13}\text{C}$  concentration in aggregates and the aggregate turnover rate.

The mass proportion both of coarse-sand (2000-200  $\mu\text{m}$ ) and clay (< 2  $\mu\text{m}$ ) fractions increased with prolonged cereal based cultivation, but the aggregate size fractions were dominated by fine-sand (200-20  $\mu\text{m}$ ) and silt (20-2  $\mu\text{m}$ ) fractions across the chronosequence. SOC was highly enriched in coarse-sand fractions (40-60  $\text{g kg}^{-1}$ ) and moderately in clay fractions (20-25  $\text{g kg}^{-1}$ ), but was depleted in silt fractions (~ 10  $\text{g kg}^{-1}$ ). The recalcitrant carbon pool was higher (33-40% of SOC) in both coarse-sand and clay fractions than in fine-sand and silt fractions (20-29% of SOC). However, the ratio of labile organic carbon (LOC) to SOC showed a weakly decreasing trend with decreasing size of aggregate fractions. Only in the coarse-sand fraction was both microbial gene abundance and enzyme activity well correlated to SOC and LOC content, although the chemical stability and respiratory of SOC were similar between coarse-sand and clay fractions. The finding here provides a mechanistic understanding of soil organic carbon turnover and microbial community succession and soil organic matter alteration at fine scale of soil aggregates that have evolved along with anthropogenic activity of cereal based cultivation in the field.

**Keywords:** conservation agriculture, soil organic carbon, coarse particulate organic matter, micro-aggregate, easily dispersed clay-sized fraction, clay-sized fraction

### Introduction

The productivity and environmental buffering capacity of soils are directly and indirectly influenced by the amount and quality of soil organic matter (SOM) (Magdoff and Weil 2004). Cultivation of land for agricultural purposes has resulted in exposure of SOM to enhanced decomposition and erosion. Estimated losses of SOM due to cultivation range from 10 to 50% of the original soil organic carbon (SOC) content (Paustian *et al.* 2000, Guo and Gifford 2002)

<sup>[51]</sup> which account for 50 to 60 Pg C worldwide (Lal 2003). Using agricultural ecosystems as a sink for atmospheric CO<sub>2</sub> to mitigate the greenhouse effect has attracted attention from both scientific and political spheres. Changes in agricultural management practices that increase the amount of carbon (C) inputs and/or reduce the amount of C outputs can potentially accomplish the restoration or even enrichment of native SOC levels, (Follett *et al.* 1997, Studdert and Echeverria 2000).

Soil organic matter (SOM) plays a key role for agro-ecosystems, given that increasing carbon stocks concomitantly improves soil structure, fertility, and crop yields (Lal, 2002) <sup>[28]</sup>. The storage of organic compounds in soil is mainly achieved by promoting the retention of the litter proceeding from previous crops and/or the incorporation of amendments. However, the mechanisms that underlie the incorporation of C<sub>org</sub> from residual plant litter into the mineral soil remain widely unknown, since they are widely variable depending on edapho climatic characteristics and land-use (Panettieri *et al.* 2015) <sup>[36]</sup>. Furthermore, the complexity of soil samples has always represented a major barrier for molecular-scale analyses of SOM (Derenne and Nguyen Tu, 2014) <sup>[12]</sup>. Carbon (C) stored in soil worldwide (1200-2400 Pg), typically in organic forms, exceeds that stored in the atmosphere (720-750 Pg) and terrestrial plants (550-835 Pg) combined (Scharlemann *et al.* 2014) <sup>[48]</sup>. Soil organic C (SOC) content is controlled predominantly by organic inputs from plants and soil type. Soil type is essentially 'fixed', whereas organic inputs can be manipulated by land use in managed systems. In agricultural systems, organic inputs are affected markedly by cultivation. The initial loss of SOC from arable cropping can be large and the return of C to soil relatively small from roots and unwanted crop residues in an annually-harvested crop, compared with that under perennial systems (Chapman *et al.* 2013) <sup>[10]</sup>.

Changes in soil management lead to a change in both the SOC content and its equilibrium as it re-adjusts to new levels of input and output. Typically, soil loses SOC faster than it gains it following management changes between arable land and conservation agriculture systems (Poeplau *et al.* 2011) <sup>[42]</sup>. Furthermore, modifications of soil structure in response to management have subsequent effects on SOC dynamics, for instance the potential for SOC to move down the soil profile in dissolved and colloid-associated forms, processes that are more prevalent under conservation agriculture than arable management (Kindler *et al.* 2011) <sup>[23]</sup>. The SOC: total N ratio in soil has a narrow range (around 9-14) in general across a range of soil types (Johnston *et al.* 2009) <sup>[21]</sup>, although an increase in the proportion of ammonium N adsorbed in clayey subsoil minerals might weaken the link with SOC (Jenkinson *et al.* 2008) <sup>[20]</sup>. Most studies of SOC in agricultural soil focus on the topsoil (to 30-cm depth) because it is the main zone of activity for crop roots, and where the need to understand nutrient and water use efficiency is paramount. Although considerable concentrations of SOC occur in the topsoil, there can be equal or greater total amounts in the subsoil (Gregory *et al.* 2014) <sup>[15]</sup>, which can be an important component of the global C cycle (Baisden *et al.* 2002) <sup>[2]</sup>.

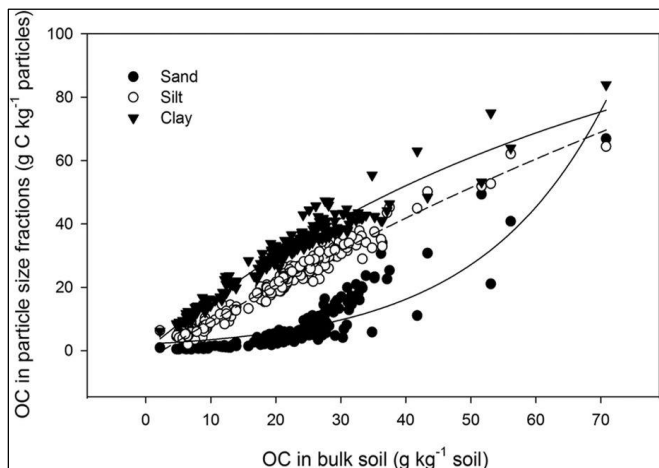
Recent interest in the subsoil has focused on its importance as a repository of SOC where the potential for increasing stocks has yet to be realized. Subsoil SOC was commonly assumed to be stable (Rumpel & Kögel-Knabner, 2011) <sup>[46]</sup> and strongly affected by soil type (Wiesmeier *et al.* 2012) <sup>[56]</sup>. All SOC is now considered to be inherently unstable thermodynamically (Schmidt *et al.* 2011) <sup>[49]</sup>, with little evidence of any difference in decomposability between

topsoil and subsoil SOC (Salomé *et al.* 2010) <sup>[47]</sup>. Any perceived stability in the subsoil or elsewhere might be linked to the soil physical environment rather than molecular recalcitrance (Dungait *et al.* 2012) <sup>[13]</sup>. The importance of the subsoil SOC pool is recognized increasingly, and the effects of soil management on SOC alteration velocity in the full profile are being explored (Beniston *et al.* 2014) <sup>[5]</sup>.

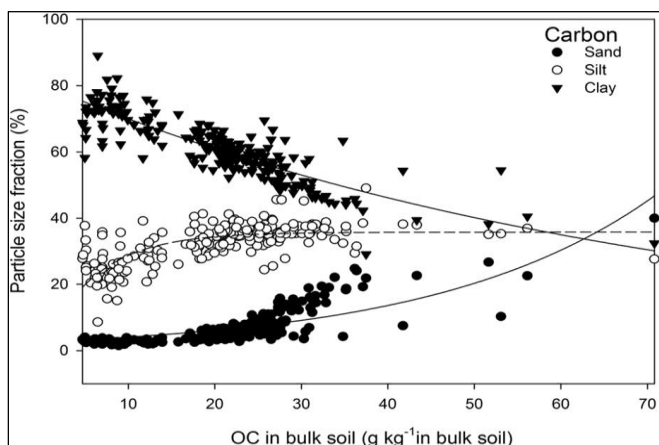
Current management practices for increasing SOM content focus on increasing C inputs and reducing exposure of SOM to decomposition and oxidation (e.g. no-till). Selection of the optimal tillage management for promoting C sequestration under high yield maize systems constitutes a dilemma. Is it better to incorporate the high volume of residue C into soil while enhancing oxidation and decomposition of SOM, or to leave them on the soil surface potentially leading to greater C loss as soil respiration? Although soil residue cover has a beneficial effect on reducing wind and water erosion, the high volume of residues can make planting and crop emergence a challenge due to increased soil moisture and cooler soil conditions in the spring. Use of no-till limits the use of in-season split applications of N fertilizer (recommended best management practice) by knife injection, which better reduce N losses by volatilization compared to fertigation. Changing SOM quantity and composition leads to changes in physical soil properties such as aggregate stability, porosity, and resistance against crusting. Therefore, cultivation of virgin soils can cause considerable decrease in soil health and quality Meyer, *et al.* (2017) <sup>[31]</sup>. On the other hand, CA is a possible option to minimize the degrading effects of conventional tillage (CT) and even improve soil quality Kassam *et al.* (2017) <sup>[22]</sup>. Even though CA covers various agro-techniques depending on geographical locations, it is basically based on non-inversion tillage and reduced tillage. The velocity of change is also hardly predictable and hence understudied, as it is a function of several environmental conditions, including climate change Mika and Farkas, (2017) <sup>[32]</sup>. This paper reviewed to quantify the changes in SOC content and SOM in particle size fractions of the same soil under CT, and CA.

Yang *et al.* (2016) <sup>[58]</sup> reported that the perspective of increasing bulk soil carbon levels over time, the clay sized particles would become saturated with C far before the silt particles, while the OC in the sand sized particle would continue to increase as bulk soil OC increases [Fig. 1a]. The relative proportions of OC in the soil particle size fractions varied with OC concentration in bulk soil [Fig. 1b]. When the concentration of bulk soil OC rose to 20 to 30 g C kg<sup>-1</sup>, which is typical in the region for near surface (0-10 cm) Brookston clay loam under grain production, the proportion of OC in clay fraction dropped to 50-60%, while that in silt and sand fractions increased to 30-35% and 15%, respectively. With further increases in bulk soil OC up to 60 g C kg<sup>-1</sup> soil, the proportion of OC associated with clay dropped to below 40%, while that in silt and sand fractions increased to about 30% [Fig. 1b]. When bulk soil OC was <20g C kg<sup>-1</sup> soil, up to 98% of the OC was associated with the silt+ clay size fraction, and when bulk soil OC was >30 g C kg<sup>-1</sup> soil, 70-80% of the OC was associated with silt+ clay. This indicates that the fine particle sizes (<53µm) of Brookston clay loam play a key role in OC retention. In addition, the maximum soil OC associated with soil particles <53 µm diameter was much greater than the "protective capacity" or "saturation level" predicted by a recognized carbon storage model for Canadian soils (40 plus g C in clay + silt fraction kg<sup>-1</sup> soil) Carter *et al.* (2003) <sup>[9]</sup>; and furthermore, the amount of OC on fine

particles continued to increase with increasing bulk soil OC with no sign of plateau. However, OC on silt+ clay continued to increase to 40–45 g C kg<sup>-1</sup> when bulk soil OC increased to 60–70 g C kg<sup>-1</sup> due to long-term sod management [Fig. 1b]. It therefore seems that the OC storage/protective capacity of a soil can be determined by both the amount of fine particles and rate of C input, rather than solely by the amount of fine soil particles.



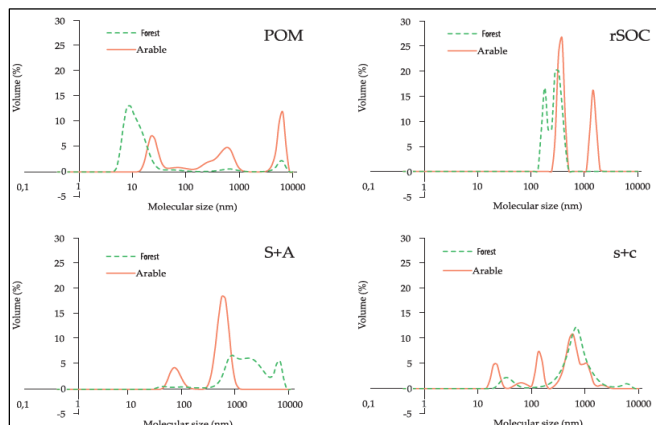
**Fig 1(a):** Relationships between the Organic Carbons (OC) Concentrations in soil particle size fractions



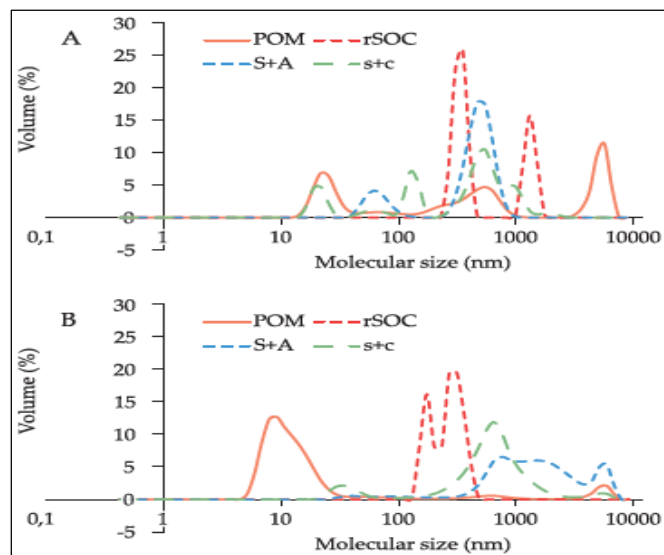
**Fig 1(b):** Proportional changes of organic carbon (OC) on soil particle size fractions in response to increasing bulk soil OC. and OC in bulk soil.

Jakab, G. *et al.* (2018) [18] revealed that the organic substances had a polydisperse molecular size distribution. The histograms were bi-, trimodal or possessed even more peaks that referred to the heterogeneous compositions of SOM in each fraction [Fig. 2a]. There were trimodal distributions at both land uses concerning the POM fraction. The two higher molecular sized peaks were situated at the same size under both forest and crop field, whereas the lowest peak that dominated the forest sample was shifted upward under crop field. In general, this comparison implied that the forest had the relatively lower molecular-sized POM composition; however, there was no relevant difference measured by the photometric indexes. In the aggregate fraction (S+A), the size difference between the forest and arable samples was the highest with a very wide range of large molecular sizes under the forest. Moreover, under forest, as it was expected, this fraction had the highest molecular size values compared to the others as was also reported by (Tan, K.H. *et al.* 2003). SOM compositions of the soil fractions under arable land and forest

were comparable, but quite different [Fig.2b]. SOM of the crop field demonstrated substantial diversity, both the highest- and lowest-sized SOM molecules were associated with POM. In this case SOM molecules were found to have a more or less continuous size distribution, whereas under forest the fractions seemed to be clustered around certain size value.



**Fig 2(a):** Soil organic matter molecular size distributions of the studied fractions under different land uses



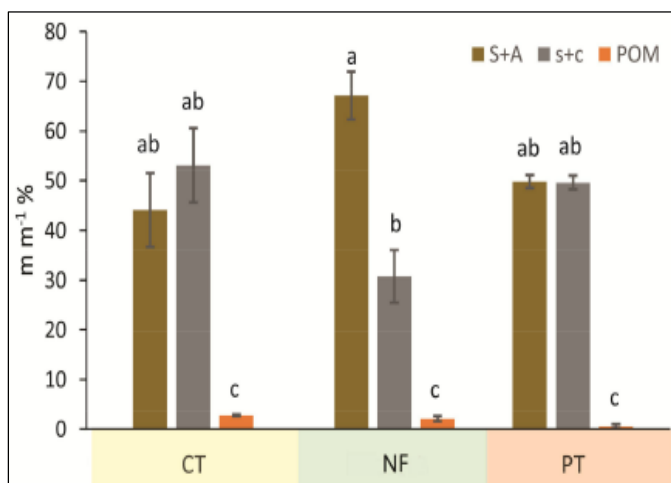
**Fig 2(b):** SOM molecular size distribution of different fractions under arable (A) and forest (B) land uses

Soil fractions from different land-use types had nearly the same particle size distribution [Fig. 3a]; however, the most uniform distribution was for the fine fraction (<63 μm). The biggest sized aggregates were still micro-aggregates (<250 μm), and were related to the CT soil, whereas the smallest ones were under NF. This suggests that—at least in the present case study—aggregate size and aggregate stability were independent of each other, as was also reported by Six and Paustian, (2014) [52]. However, Zotarelli *et al.* (2005) [61] found a direct relationship between aggregate size and stability in Oxisols. Bulk NF soil contained more than three times the SOC compared with crop fields; however, CT significantly exceeded PT ( $p < 0.05$ ) in terms of SOC [Fig. 3b]. The lowest standard deviation values were found under PT, mainly because it had the lowest SOC and POM content [Fig. 3a].

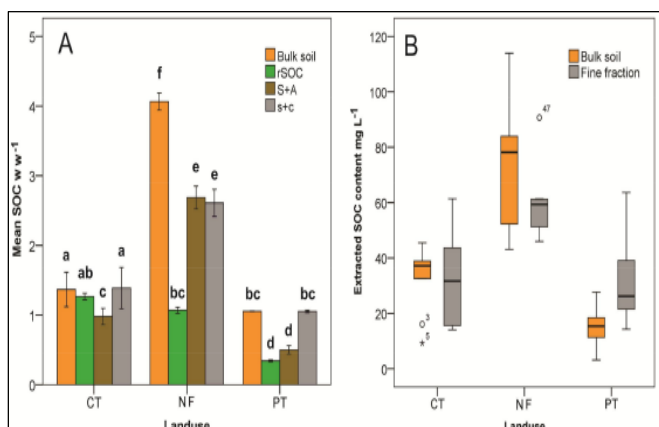
Rieder *et al.* (2018) [45] have also measured recent SOM attachment to the s+c fraction over 12 years, under similar climate and soil conditions. On the other hand, the main part of this SOC pool was recalcitrant under CT, resulting in high

rSOC values, whereas, under PT and forest labile, vulnerable SOC components dominated this pool [Fig. 4a]. In contrast, under NF, s+c SOC was higher, but made a lower contribution to the SOC content of the bulk soil. S+A-associated SOC considerably increased due to the omission of plowing (Skjemstad *et al.* 2004; six *et al.* 2000) [50, 51]. Beare *et al.* (1994) [4] also found the same increase in the micro-aggregate fraction after 13 years of no tillage, which underlined the velocity of the SOC increase in the micro-aggregate (<250  $\mu\text{m}$ ) fraction.

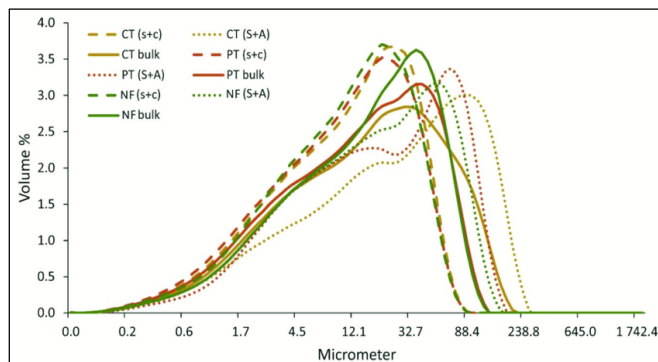
The SOM attached to the finest mineral particles revealed the most heterogeneous pattern for the crop field, as they contained molecules >900 nm [Fig. 4b]. Besides this peak, PT had the most similar pattern in the case of S+A, whereas CT and NF resulted in different SOM compositions in the S+A and s+c fractions. Due to heavy oxidization, both crop field SOMs became more homogeneous, losing their molecules that were <1000 nm and <100 nm [Fig. 4b]. In contrast, the silt and clay-associated SOM of the NF seemed to be degraded as a whole, resulting in very similar rSOC molecular size distribution [Fig. 4b].



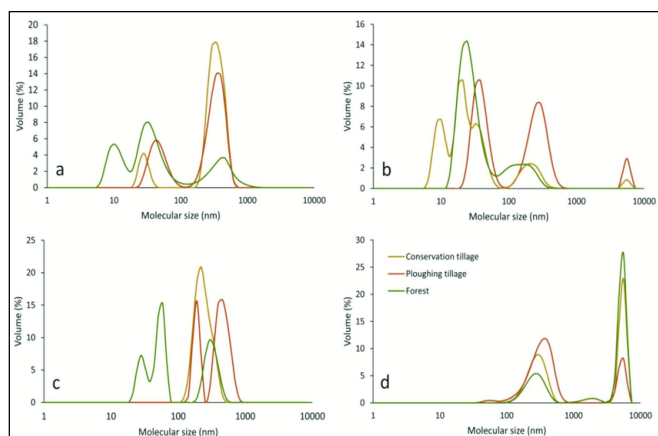
**Fig 3(a):** Changes in aggregate stability and particulate organic matter (POM) share of the soil under various land uses. NF-native forest; CT-conservation tillage; PT-plowing tillage; (S+A)-sand and aggregates (>63  $\mu\text{m}$ ); (s+c)-silt and clay fraction (<63  $\mu\text{m}$ )



**Fig 3(b):** Soil organic carbon (SOC) content of the soil fractions under various land uses (A) and the alkaline soluble SOC content (B); NF-native forest; CT-conservation tillage; PT-plowing tillage; (S+A)-sand and aggregates (>63  $\mu\text{m}$ ); (s+c)-silt and clay fraction (<63  $\mu\text{m}$ )



**Fig 4(a):** Particle size distributions of the investigated soils and soil fractions. NF-native forest; CT-conservation tillage; PT-plowing tillage; (S+A)-sand and aggregates (>63  $\mu\text{m}$ ); (s+c)-silt and clay fraction (<63  $\mu\text{m}$ ).



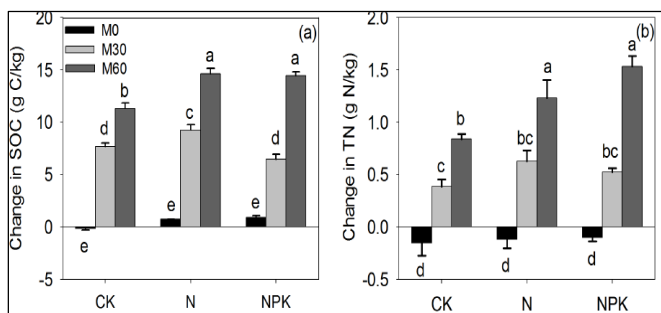
**Fig 4(b):** Molecular size distributions of the dissolved soil organic matter (SOM) of the separated fractions under various land-use types. (a) Sand and aggregate fraction; (b) silt and clay fraction; (c) recalcitrant soil organic matter; (d) dissolved organic matter

Cai *et al.* (2016) [7] showed that long-term manure application significantly increased SOC and total N content and enhanced C and N mineralization in the three particle-size fractions. The content of SOC and total N followed the order 2000-250  $\mu\text{m}$  > 250-53  $\mu\text{m}$  > 53  $\mu\text{m}$  fraction, whereas the amount of C and N mineralization followed the reverse order. In the <53  $\mu\text{m}$  fraction, the M60NPK treatment significantly increased the amount of C and N mineralized compared to the M<sub>0</sub>CK treatment. Nitrogen mineralization significantly increased with increase in manure application rate, while chemical fertilizer or manure application did not influence the C mineralization rate [Fig. 5a]. Fractionation recovery rate of total soil weight of the three fractions ranged from 96.1-101.7%; the recovery rate of SOC in the three fractions ranged from 90.2-101.8%, and that of total N ranged from 89.0-103.4%. Long-term manure with/without chemical fertilizer application significantly increased the content of SOC and total N in all soil particle-size fractions, while application of chemical fertilizer on its own (M<sub>0</sub>N, M<sub>0</sub>NPK) did not alter SOC or total N content in any soil particle-size fractions compared with the control (CKM<sub>0</sub>) (Fig 5b).

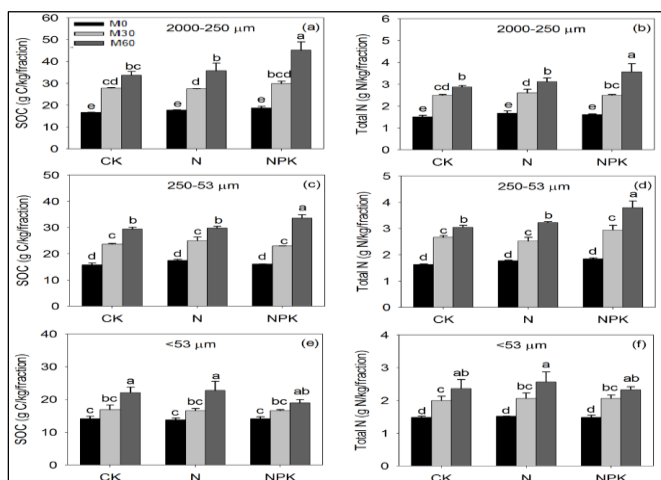
Compared with no manure application (M<sub>0</sub>), the 30 and 60 t ha<sup>-1</sup> annual manure application rates increased the SOC in soil fractions by 20-60% (M<sub>30</sub>) and 52-115% (M<sub>60</sub>), and increased total N by 34-57% (M<sub>30</sub>) and 61-99% (M<sub>60</sub>). The greatest amount of SOC (45.1 g C kg<sup>-1</sup> fraction) and total N (4.1 g N kg<sup>-1</sup> fraction) occurred in the 2000-250  $\mu\text{m}$  particle-size fractions under the M<sub>60</sub>NPK treatment. The 2000-250  $\mu\text{m}$  and 250-53  $\mu\text{m}$  particle-size fractions contained approximately

37% of the total SOC and total N pools among the three particle-size fractions, regardless of the difference in management treatments [Fig 6a]. The trend N mineralization increased with manure application, showing more N mineralized from <53 μm fraction under the M<sub>30</sub>NPK and M<sub>60</sub>NPK treatments. There was no relationship between the storage of SOC or total N and the C or N mineralization from the bulk soil [Fig 6b]. Interestingly, N mineralization was strongly related to total N storage in the 250-53 μm and <53 μm fractions and C mineralization was significantly positive related to SOC storage in the 2000-250 μm and <53 μm fractions.

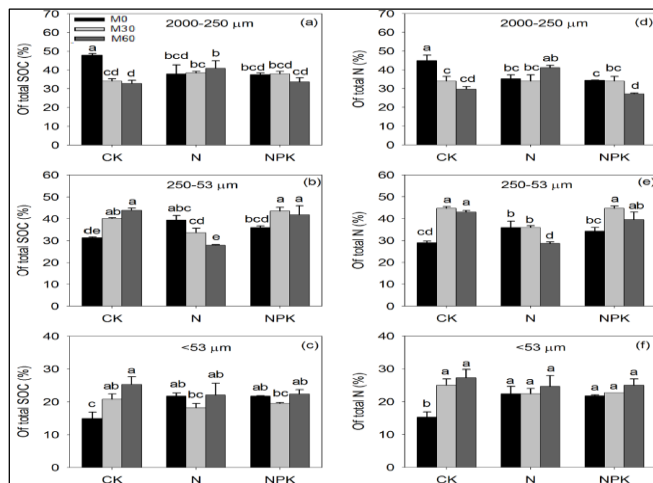
Zhu, *et al.* (2007) [60] showed that while mineral fertilizer application can maintain high yields, a combination of chemical fertilizers plus manure was required to enhance soil C sequestration. The content of SOC and total N in soil particle-size fractions declined with a decrease in particle-size, indicating that larger-size soil fractions were the main pool of SOC and total N Cambardella and Elliott, (1994) [8] with the less decomposable SOC and N associated with fine soil particles Puget *et al.* (1995) [43]. The N mineralized in the <53 μm fraction was about 1.5 times greater than that in the other two fractions, suggesting that smaller particle-size fractions contain a larger proportion of readily mineralizable organic N than larger particle-size fraction in Mollisols. The significant linear relationships between SOC storage and C mineralization in the <53 μm particle-size fraction indicated that this fraction had the greatest capacity for SOC and N mineralization.



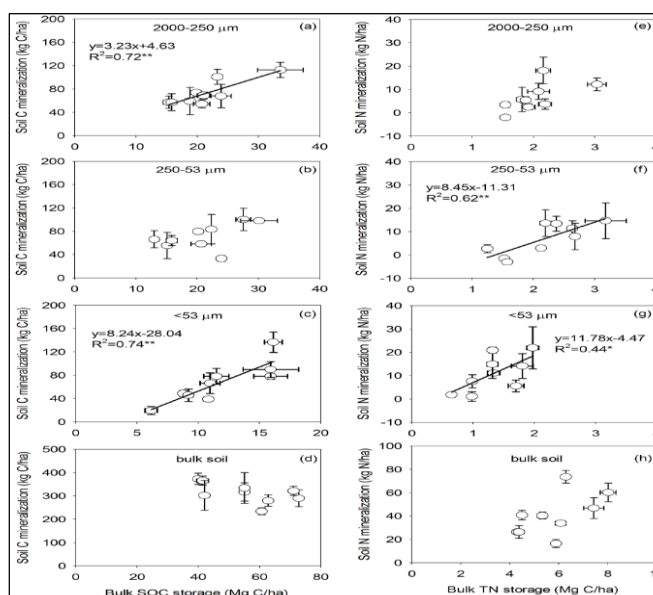
**Fig 5(a):** Change in soil organic carbon (SOC, a) and total nitrogen (N) (b) in bulk soil from the control (CK)



**Fig 5(b):** The content of soil organic carbon (SOC) in 2000-250 μm (a), 250-53 μm (c), <53 μm (e) fraction and total nitrogen (N) in 2000-250 μm (b), 250-53 μm (d), <53 μm (f) fraction from the control (CK)



**Fig 6(a):** The percent (%) of soil organic carbon (SOC) in 2000-250 μm (a), 250-53 μm (c), <53 μm (e) fraction and total nitrogen (N) in 2000-250 μm (b), 250-53 μm (d), <53 μm (f) fraction from the control (CK)

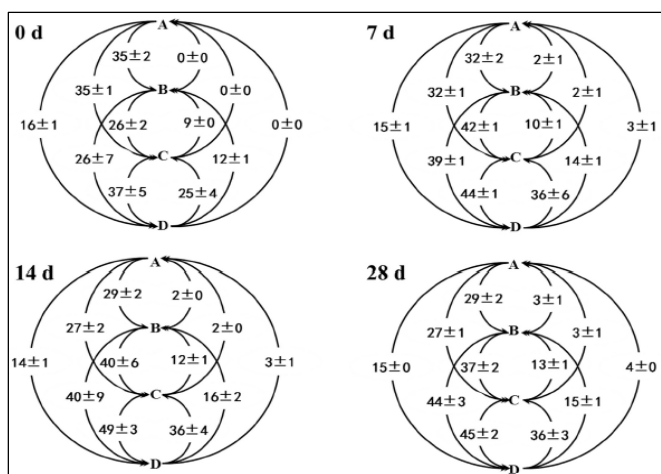


**Fig 6(b):** Relationship between soil organic carbon (SOC) storage vs potential carbon (C) mineralization, and total N storage, potential nitrogen (N) mineralization in 2000-250 μm, 250-53 μm, <53 μm fraction and bulk soil after 32 years of manure fertilization.

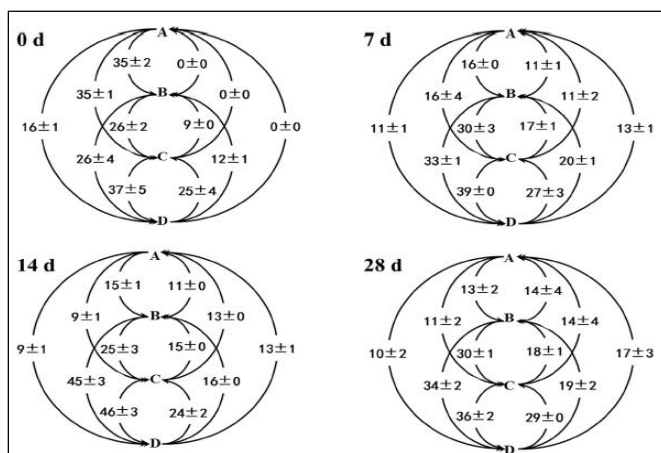
Peng *et al.* (2017) [38] reported that the greater portion of aggregates transferred between neighboring size fractions. The turnover rate was faster for macro-aggregates than for micro-aggregates, and slowed down over the incubation time. The new C was accumulated more but decomposed faster in macro-aggregates than in micro-aggregates. A positive relationship was observed between the <sup>13</sup>C concentration in aggregates and the aggregate turnover rate. The relative change in each aggregate fraction generally followed an exponential growth over time in the formation direction and an exponential decay in the breakdown direction. The changes in either breakdown or formation directions mainly happened in the first week, and then became less over the incubation time. A greater transfer portion of soil aggregates was observed between neighbouring size fractions either in breakdown or formation directions. The glucose addition significantly reduced aggregate breakdown and increased aggregate formation. Relative to the REO treatment, 2-5 mm, 0.25-2 mm and 0.053-2 mm aggregates showed a less breakdown in the <sup>13</sup>C + REO treatment by 36-38%, 10-18%

and 3-9%, respectively, indicating that the glucose addition stabilized larger aggregates greater than smaller ones [Fig.7a and 7b]. To test the efficacy of REOs as tracers for aggregates transformation, the amount of aggregates was predicted according to the REO transfer matrix shown in [Figs. 7a and 7b].

The glucose addition significantly improved the aggregate turnover rate in which greater was observed for 2-5 mm aggregates. A significantly linear relationship was observed between the turnover rate and <sup>13</sup>C concentration in aggregates [Fig. 8c]. The aggregate turnover time, reciprocal to the aggregate turnover rate, increased with the incubation time, and the glucose addition decreased the turnover time significantly except for the 0.25-2 mm aggregates [Fig.8b]. During the 28 day incubation, the shortest turnover time was observed for 0.25-2mmaggregates (87 days) in the REO treatment and for 2-5mmaggregates (54 days) in the <sup>13</sup>C + REO treatment, while the longest turnover time was observed for 0.053-0.25 mm aggregates from the REO treatment (186 days) and the <sup>13</sup>C + REO treatment (130 days) [Fig. 8a].

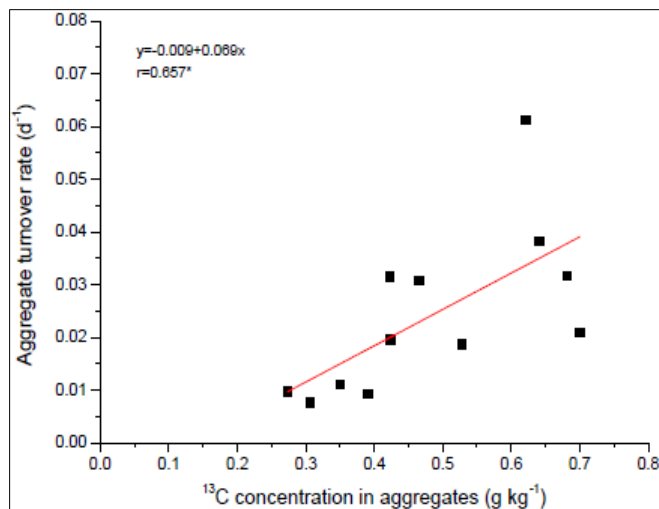


**Fig 7(a):** The 12 transformation paths of four aggregate fractions on 0, 7, 14 and 28 day of incubation under the REO treatment. Values in arrows are the relative change of this aggregate fraction (%). A, B, C, and D represent 2-5 mm, 0.25-2 mm, 0.053-0.25 mm, and <0.053 mm aggregates.

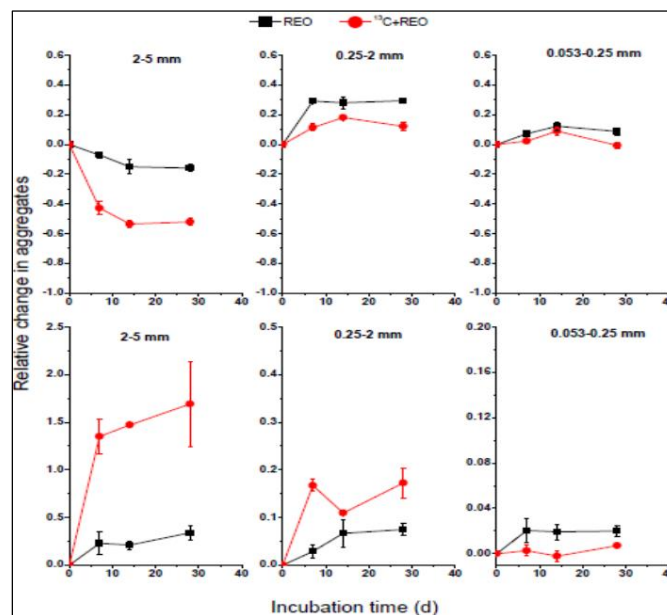


**Fig 7(b):** The 12 transformation paths of the proportions of four aggregates on 0, 7, 14 and 28 day of incubation under the <sup>13</sup>C + REO treatment. Values in arrows are the relative change of this aggregate fraction (%). A, B, C, and D represent 2-5 mm, 0.25-2 mm, 0.053-0.25 mm, and <0.053 mm aggregates.

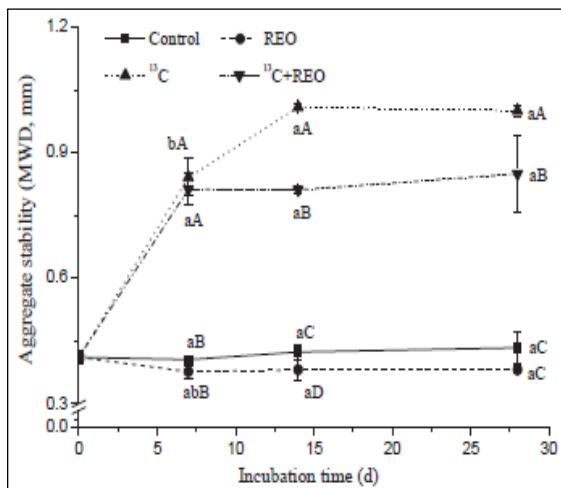
The relative change showed an exponential decrease for 2-5mmaggregates and an exponential increase for 0.25-2mm aggregates. For the 0.053-0.25 mm aggregates, the exponential trend was not clear. The 0.25-2 mm aggregates showed a much greater breakdown proportion than 0.053-0.25 mm aggregates. Relative to the REO treatment, the glucose addition enhanced aggregate stabilization for 2-5 mm aggregates and reduced the breakdown of the other fractions significantly. Kemper and Rosenau (1986) [24] pointed out that the stability of aggregates can increase with storage time. De Gryze *et al.* (2006) [11] who found that under breakdown most of the portions of all aggregate size fractions were transferred into the <0.053 mm fraction. This difference may be soil specific and caused by the poorer stability of artificial macro-aggregates in their study. In the formation direction, the newly formed aggregates exponentially increased with time, with larger aggregates forming faster than smaller ones



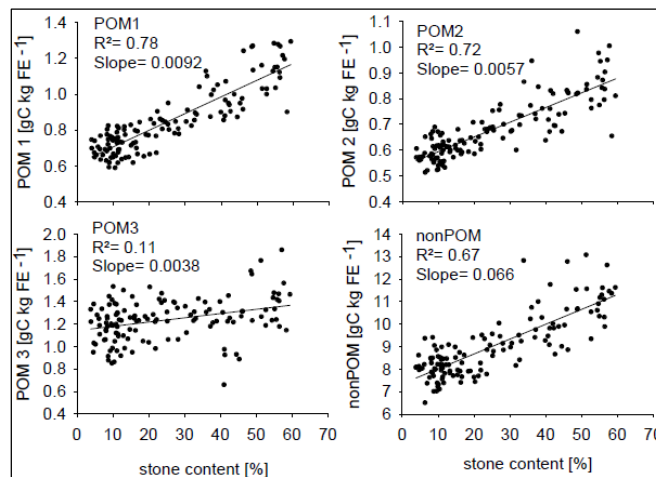
**Fig 8(a):** The linear relationship between aggregate turnover rate and <sup>13</sup>C concentration in aggregates



**Fig 8(b):** Relative changes in aggregates in the breakdown direction and in the buildup direction

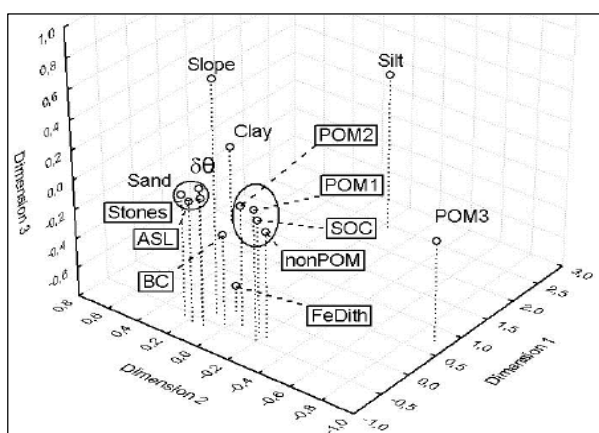


**Fig 8(c):** The dynamics of aggregate stability (MWD) over time under i) soil without REO and  $^{13}\text{C}$  as a control, ii) soil labelled with REO, iii) soil without REO but amended with  $^{13}\text{C}$ -glucose, and iv) soil labelled with REO and amended with  $^{13}\text{C}$ -glucose over incubation time.



**Fig 9(b):** Scatter plots displaying the relation between the stone content and contents of particulate organic matter of three size classes (POM1: 2000-250  $\mu\text{m}$ ; POM2: 250-53  $\mu\text{m}$ ; POM3: 53-20  $\mu\text{m}$ ), as well as nonparticulate organic matter (nonPOM) in fine earth

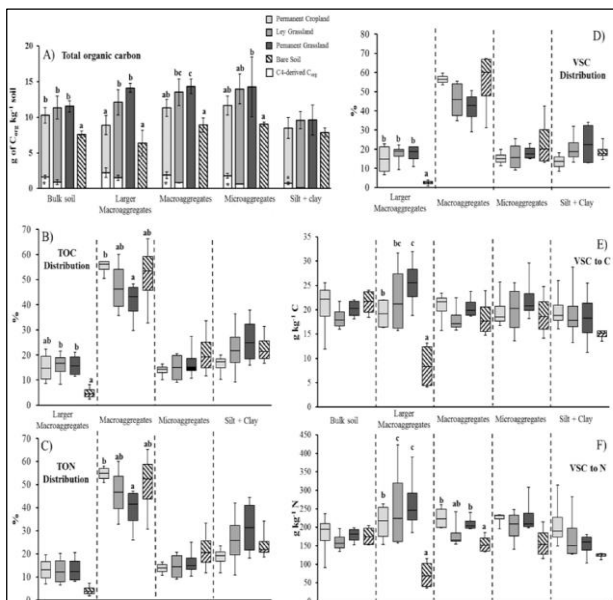
Zur Erlangung des Grades, (2010) [62] reported that the soil constituting ~71–81% of bulk SOC, non POM represented the quantitatively most relevant SOC fraction at this field site. The remaining 19-29% of total SOC was comprised by the POM fractions, with decreasing proportions in the order  $\text{POM3} \geq \text{POM1} \geq \text{POM2}$ . As indicated by good to very good stress values (stress 1 = 0.065, stress 2 = 0.034; Kruskal, 1964) [27], the similarity matrix was reproduced satisfactorily by the Euclidian distances of the first three dimensions of the MDS [Fig. 9a]. Zur Erlangung des Grades, (2010) [62] also found that the correlation lengths of the contents of coarse POM in bulk soil match the one of  $\delta\theta$  [Fig. 9b]. Secondly, a positive relation is indicated between the stone content and the C/N ratio of the bulk SOC ( $R^2 = 0.53$ ), thereby documenting that SOC in the very gravelly areas in deed enriched with weakly decomposed plant litter. Retarded decomposition and disproportionately high biomass input thus likely explain the enhanced contents of POM1 and POM2 in those areas that comprise high stone contents. Bornemann *et al.* (2010) [6] revealed that bulk SOC, POM1, and POM2 exhibited a close spatial correlation on the investigated site, while POM3 showed a rather independent pattern.



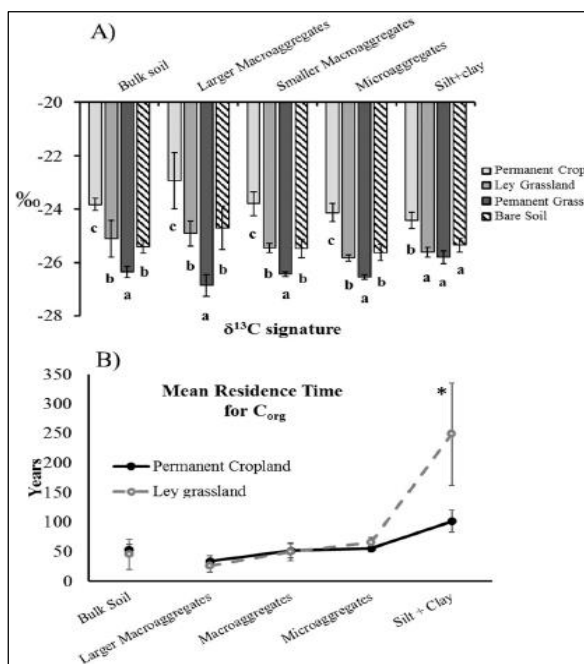
**Fig 9(a):** Three-dimensional alignment of the Euclidian distances of soil organic carbon (SOC), particulate organic matter of three size classes (POM1: 2000-250  $\mu\text{m}$ ; POM2: 250-53  $\mu\text{m}$ ; POM3: 53-20  $\mu\text{m}$ ), non-particulate organic matter (nonPOM), black carbon (BC), dithionite-soluble Fe oxides (Fe DCB), sand, silt, and clay, fine earth contents of stones, difference of volumetric soil moisture between two dates of measurement ( $\delta\theta$ )

Panettieri *et al.* (2017) [37] reported that Soils under ley and permanent grassland stored higher amount of SOM in larger aggregates and preserved more efficiently the lignin stocks than the corresponding samples under permanent maize. Contemporary, finer fraction of ley grassland showed higher mean residence time of organic carbon, probably due to a legacy effect of the previous years under grassland. Even if maize derived SOM was identified, the grassland footprint was still dominating the ley grassland soils. TOC increases under both ley and permanent grassland were detected only for fast responding fractions, such as larger macro-aggregates ( $\phi$  2-7.1 mm), which changed after 6 years grassland, and for macro-aggregate ( $\phi$  0.200-2 mm) fractions, which are affected only after 9 years of grassland [Fig. 10a]. The short term storage is counterbalanced by the longer MRT detected for  $\text{C}_{\text{org}}$  in finer fractions of ley grassland soil, if compared with continuous maize ones. These results suggested that the higher root density and the absence of tillage were responsible of a tight interaction of litter derived  $\text{C}_{\text{org}}$  with finer fraction of the soil. This interaction resulted in a “positive” legacy effect generated by ley grassland, which could increase the storage at longer term. It should be remarked that the experimental area has been devoted to continuous cropping for decades, and a “negative” legacy effect hampering soil recovery has to be expected after a conversion to permanent or ley grassland. Longer periods of time are often required to appreciate net carbon storage in similar ecosystems (Smith, 2014) [53]. These findings indicated the possibility of a net carbon storage in ley grassland plots at long-term, maintaining adequate economic benefits for landholder as compared to continuous cropping Lemaire *et al.* 2014) [30] [Fig. 10b].

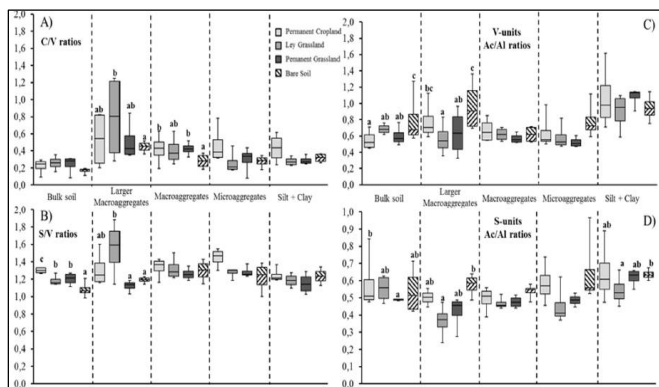
Permanently cropped soil had a significantly higher ratio than ley grassland for larger macro-aggregates ( $\phi$  2-7.1 mm), whereas no further differences were found for the other fractions. Regarding the S units, the Ac/Al-S ratio was significantly higher for permanent cropland compared to ley grassland for bulk soil, and for bare fallow compared to ley grassland for larger macro-aggregates ( $\phi$  2-7.1 mm) and to ley grassland and bare fallow for silt plus clay fraction [ $\phi$  b 0.050 mm, Fig. 10c].



**Fig 10(a):** Soil aggregate-size fractions of C4 derived and total organic carbon (TOC) contents (A) and distribution (B), total organic nitrogen (TON) distribution (C), total lignin (VSC) distribution (D) and lignin to carbon (E) and lignin to nitrogen (F) values

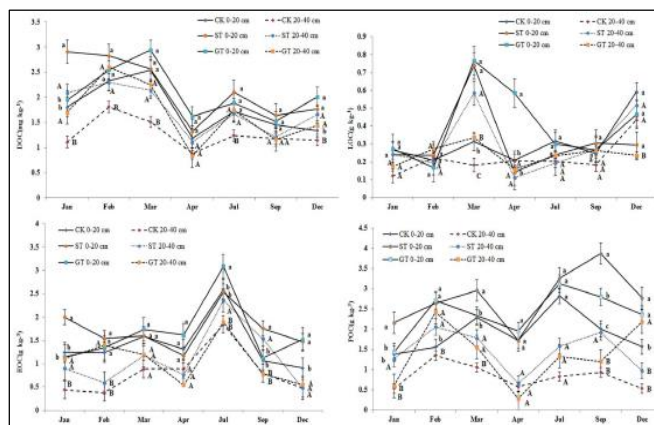


**Fig 10(b):** Soil aggregate-size fractions of  $\delta^{13}C$  signature (A) and mean residence time of organic carbon (B).



**Fig 10(c):** Soil aggregate-size fractions of cinnamyl to vanillyl unit's ratios (A) and syringyl to vanillyl (B), and the acid to aldehyde ratios for vanillyl and syringyl units (C and D).

Gu *et al.* (2016) [16] showed that mulch applied on the soil surface increased the contents of SOC and its active fractions in the soil. Compared to the control without cover (CK), ST and GT treatments increased the contents of SOC, LOC, DOC, POC and EOC by 14.73%, 16.5%, 22.5%, 41.5% and 21%, respectively, in the 0±40 cm soil layer, and by 17%, 14%, 19%, and 30%, respectively, in the 0±100 cm soil layer. The contents of organic carbon and its active fractions decreased with increasing soil depth in all of the treatments [Fig 11a]. Bastian *et al.* (2009) [3] reported that the formation of POC was often in relatively cold seasons, while the depletion of POC was in hot seasons, because the increase in soil temperature and moisture enhanced the rate of decomposition of POC Obalum and Obi, (2010) [35]. But in fact, this rule is not absolute: if the rate of production of new POC was greater than the rate of POC decomposition, POC would show a net accumulation, even in the hot season. The high concentration of POC in GT and ST treatments occurred in July and September [Fig 11a], probably due to the greater input of newly decomposed residue materials at this time. The contents of soil DOC and LOC were high in January to March, while the contents of soil POC and EOC were high in June to September. The relative contents of soil organic carbon fractions were POC>EOC > LOC > DOC over the four years [Fig. 11b]. The DOC concentration is considerably lower than those of other labile C fractions, generally not more than 200 mg kg±1, but it is the most mobile fraction of SOC. It controls the turnover of nutrient and organic matter by affecting the development of microbial populations. The increase in DOC with ST may be due to the soluble decomposed organic materials of the straw, while the increase in DOC with GT could possibly be attributed to an increase in organic acids and water-soluble carbohydrates from rhizodeposition and root exudates. In addition, a decrease in surface runoff under GT and ST was an important reason for the increased DOC, as DOC may be lost with runoff. Compared with CK, the DOC in GT and ST was favorably leached, deposited and absorbed into the subsoil layer, resulting in higher concentrations of DOC at depths of 20±40 cm [Fig 11b]. This was probably because of low soil bulk density in ST, and in GT lower pH would have increased DOC adsorption by soil Jardine *et al.* (1989) [19].



**Fig 11(a):** Dynamic changes of carbon fractions



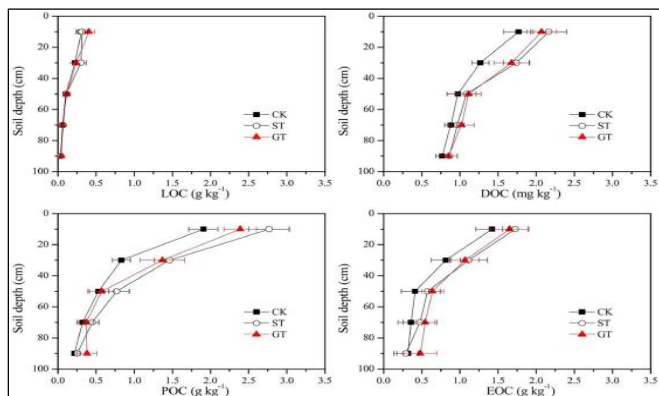


Fig 11(b): Content of Carbon fractions at different depths

Pisani *et al.* (2016) [39] reported that the implementation of different agricultural management practices enhances the degradation of recalcitrant SOM compounds that may become a source of atmospheric CO<sub>2</sub> with increasing land-use and climate change [Fig. 12]. Soil organic matter chemical properties and interactions with the mineral matrix allow them to be placed in different SOM functional pools with different turnover rates (Kögel-Knabner & Kleber, 2012) [25]. The SOC pool contributes to soil fertility both directly and indirectly. Directly it releases important inorganic nutrients and trace elements while it decomposes and indirectly it increases the soil cation exchange capacity (CEC) and water holding capacity while it also improves the structure of the soil. High organic C levels are also important to ensure active microorganism populations which are necessary for sustainable crop production systems (Lal, 2011) [29].

Sohi *et al.* (2005) [54] also confirmed that SOM within aggregates contains more microbial products and more resistant C as compared with SOM in the light fraction. Usually by increasing the proportion of C-rich macro-aggregates in soils, C sequestration can be enhanced, but long-term sequestration depends on stabilization of C in micro-aggregates (Six *et al.* 2000) [51]. Six *et al.* (2000) [51] suggested that no-till practices that minimize macro-aggregate turnover enhances the formation of stable micro-aggregates within the macro-aggregates, and therefore ensure long-term C sequestration via physical occlusion of the micro-aggregates protecting it from microbial breakdown. Micro-aggregates form within macro-aggregates as the fine organic matter becomes encrusted with clay particles and microbial products. Mupambwa & Wakindiki (2012) [33] which stated that micro-aggregates formed first followed by macro-aggregates. The micro-aggregates showed equal or greater decrease in <sup>13</sup>C compared to the macro-aggregates, suggesting that micro-aggregates are not protected by macro-aggregates. Verchot *et al.* (2011) [55] thus concluded that the stable micro-aggregates form through the interaction between mineral surfaces and organic ligands [Fig. 12].

Huang *et al.* (2010) [17] found an 18.1% increase in the concentration of total SOC compared with conventional tillage under a long-term maize monoculture with no-till causing larger C concentrations in macro-aggregates (>2000 and 250-2000 μm) and micro-aggregates (53-250 μm) relative to conventional tillage. Smith, (2014) [53]

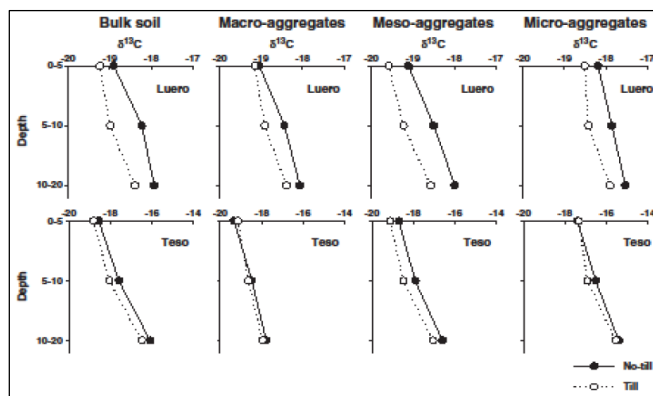
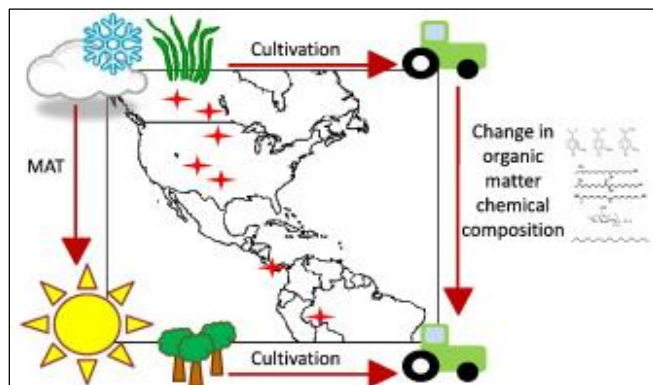
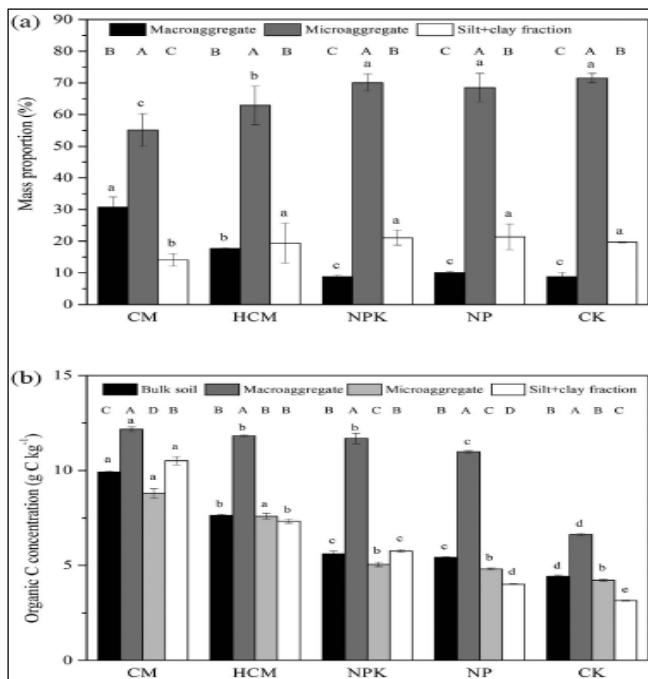
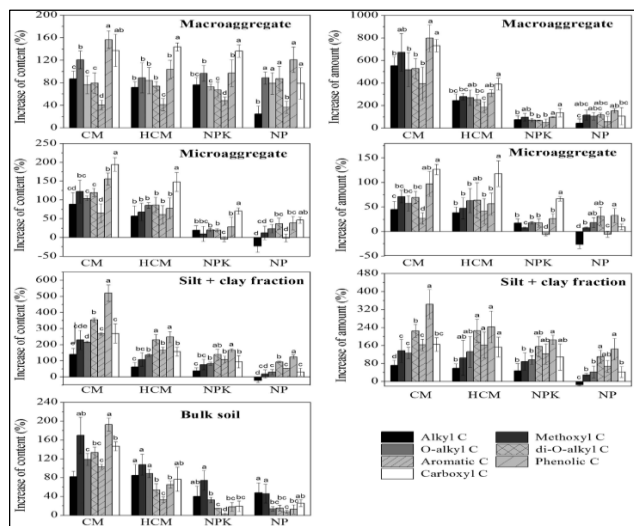


Fig 12: The effect of tillage on <sup>13</sup>C values in the bulk soil and in each fraction by depth

Yu *et al.* (2015) reported that Compost application alone (CM) or in combination with NPK (HCM) significantly increased the mass proportion of macro-aggregates by 250% and 101%, respectively) and reduced the proportion of micro-aggregates and the silt + clay fractions compared with CK [Fig. 13a]. Long-term application of compost more effectively improved OC content in bulk soil than did long term mineral fertilizer application; the percentage increase in OC compared with CK was as follows: CM (124%) > HCM (72%) > NPK (27%) > NP (23%). Compared with CK, compost application alone (CM) or in combination with NPK (HCM) increased the OC content in macro-aggregates, micro-aggregates and the silt + clay fractions. Mineral fertilizer amendment also increased the OC content in macro-aggregates and the silt + clay fractions but not in micro-aggregates [Fig. 13a]. The increase in phenolic C, carboxyl C and methoxyl/N-alkyl C content (88.8–156.6%) was higher than that of O-alkyl C, di-O-alkyl C, alkyl C (67.3-87.0%) and aromatic C (40.5-48.0%) in macro-aggregates in compost- or NPK-amended soils [Fig. 13b]. Moreover, irrespective of the nature of the amendments, all amendments preferentially promoted the accumulation of carboxyl C in micro-aggregates (46.0-194.1% vs. - 22.9-156.1% for other C functional groups), as well as the accumulation of phenolic C and di-O-alkyl C in the silt + clay fractions (93.5-520.0% vs. - 21.4-314.7% for other C functional groups; Fig. 13b).



**Fig 13(a):** Mass proportion of aggregates (a) and organic C content (b) in bulk soils and aggregates, as affected by long-term application of compost and mineral fertilizers



**Figure 13(b):** Increase in organic C functional group content or amount in bulk soils and aggregates, as affected by long-term application of compost and fertilizers

Wingeyer, (2011) reported that from fall 2005 to fall 2008, soil C and N stocks were reduced by 23% in the first layer (60 kg soil m<sup>-2</sup>), with the greatest reduction after the first tillage (17% decline for both stocks compared to 2005 levels). A significant increase of C (+22%) and N (+16%) stocks after the first tillage was observed in the 60-200 kg soil m<sup>-2</sup> layer. By fall 2008, the difference in C and N stocks in the 60 to 200 kg soil m<sup>-2</sup> layer compared to 2005 were +7% and +4% respectively [Fig.14a and 14b]. The slight increases in the soil C and N stocks detected in the 200 to 400 kg soil m<sup>-2</sup> layer (3 and 4%, respectively) were not significant. When the three soil layers were summed, soil C and N stocks after three years of tillage were similar to those under prior no-till. Gifford and Roderick (2003) for the calculation of soil C and N stocks better accounted for the differences in effective sampling depth across years. The equal soil mass layers were set as 60, 200 and 400 kg soil m<sup>-2</sup>, which approximated the 5, 15 and 30

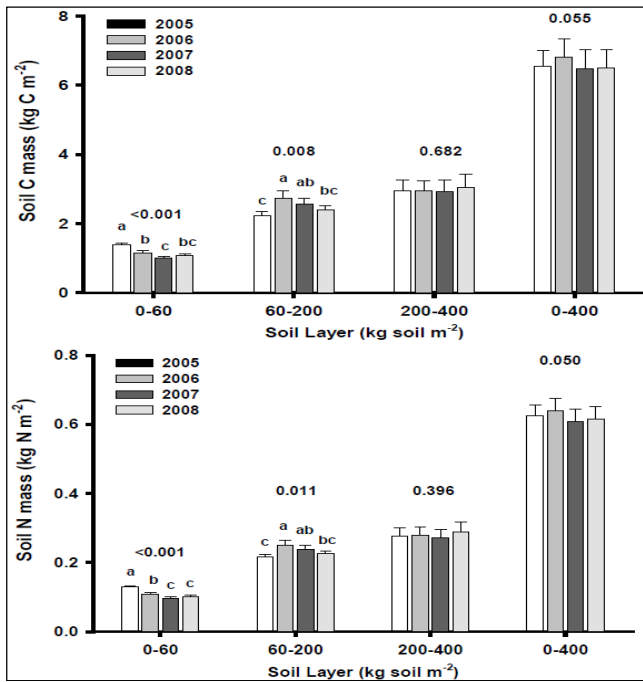
cm depths, respectively. Both soil C and N stocks followed a similar trend among years.

The reduction in MHA-C and -N in the uppermost layer was mirrored by an increase in the second layer from 2005 to 2006, with minor changes in the following years [Fig. 14b]. For the 200-400 kg soil m<sup>-2</sup> layer, MHA-C and -N increased by 2008 (Fig. 14b). No significant changes in MHA-C and -N were determined when pooling all soil layers together [Fig. 14b]. Changes in CaHA-C and -N in the first soil layer had similar trends to those of the MHA fraction, but of smaller magnitude. The CaHA-N was reduced by 20% in the 60 kg soil m<sup>-2</sup> layer after the first year, and the CaHA-C increased by 15% in 2008 in the 60-200 kg soil m<sup>-2</sup> [Fig. 15a]. The C and N trends in the pooled 0-400 kg soil m<sup>-2</sup> layer followed the trend in the 200-400 kg soil m<sup>-2</sup> soil layer. Changes in humin-C and -N stocks in all three layers were similar to those of the MHA [Fig. 15b], namely small reduction of C and N stocks in the 0-60 kg soil m<sup>-2</sup> layer and increase in the C and N stocks in the 60-200 and 200-400 kg soil m<sup>-2</sup> layer [Fig. 16a]. The increases in C and N stocks from 2006 to 2008 as CaHA in the 60-200 kg soil m<sup>-2</sup> layer (+24 g C m<sup>-2</sup> and +1 g N m<sup>-2</sup>), and as MHA (+31 g C m<sup>-2</sup> and +2.7 g N m<sup>-2</sup>) and CaHA (+42 g C m<sup>-2</sup> and +1.6 g N m<sup>-2</sup>) in the 200-400 kg soil m<sup>-2</sup> layer reported by Murage and Voroney, (2008) [34]. For the 0-400 kg soil m<sup>-2</sup> layer, humin-C and -N stocks increased over time by 4 and 5%, respectively. Before tillage, the two humic acid fractions plus humin comprised 74.1, 75.8, and 84.3% of soil C stocks in the 0-60, 60-200 and 200-400 kg soil m<sup>-2</sup> layers, respectively. By 2008, these fractions represented 79.0, 79.2 and 85.2 % of soil C in the same soil layers. Similar trends were observed for the proportion of soil N.

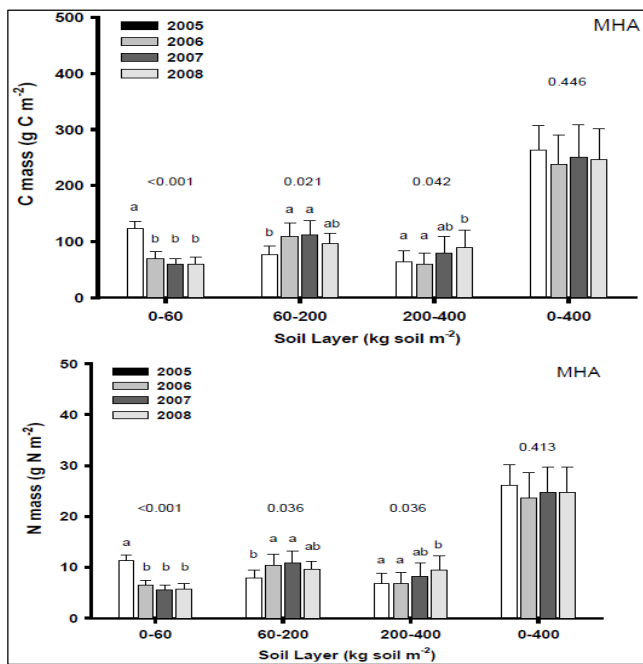
The small reduction in fLF-C and -N stocks in the 0-60 kg m<sup>-2</sup> soil layer was offset by large increases in the 60-200 and 200-400 kg soil m<sup>-2</sup> layers [Fig. 16b] Free LF-C and -N in the 0-60 kg m<sup>-2</sup> soil layer were reduced by 30% in 2008 compared to 2005. In the 60-200 and 200-400 kg soil m<sup>-2</sup> layers, however, the incorporation of crop residues increased the fLF-C and -N by 172% and 149%, and by

318% and 285%, respectively [Fig. 16b] the increased stabilization of oLF (greater oLF-C and -N stocks in the 0-400 kg soil layer m<sup>-2</sup>) is indicative of repacking of aggregates. Despite the fact that tillage induces the breakdown of aggregates, reduced aggregate turnover under no-till can constrain stabilization of new oLF (Yoo and Wander 2008) in all aggregate fractions compared to tillage (Plante and McGill 2002) [41].

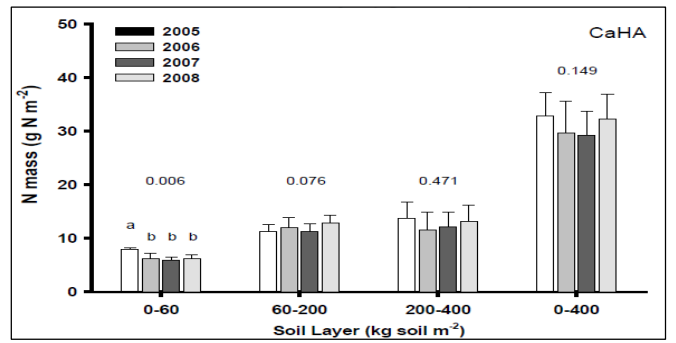
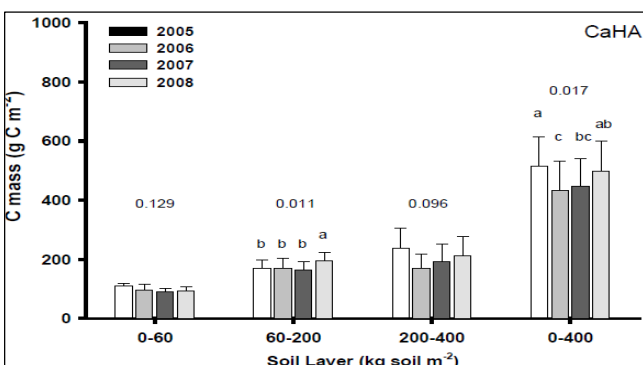
The three fractions together accounted for 62.5, 60.5 and 65.3% of soil N stocks in 2005, and 65.1, 65.2 and 68.8% of soil N in 2008 for the 0-60, 60-200 and 200-400 kg soil m<sup>-2</sup> layers, respectively. In 2005, C and N in free plus occluded LF accounted for 5.6, 2.3, and 1.6% of soil C stocks and 3.4, 1.2, and 0.7 % of soil N stocks for the 0-60, 60-200 and 200-400 kg soil m<sup>-2</sup> layers, respectively. In 2008, fLF and oLF together represented 4.5, 4.1, and 3.2% of soil C stocks and 2.5, 2.1, and 1.5% of soil N stocks for the same soil layers, demonstrating a shift in C and N stocks down the profile [Fig. 16 a and 16b]. Angers and Eriksen-Hamel, (2008) [1] revealed that no-till practices enhance accumulation of C and N close to the soil surface, plough tillage homogenizes C and N concentrations throughout the ploughed depth and increases C and N concentrations below the tillage depth



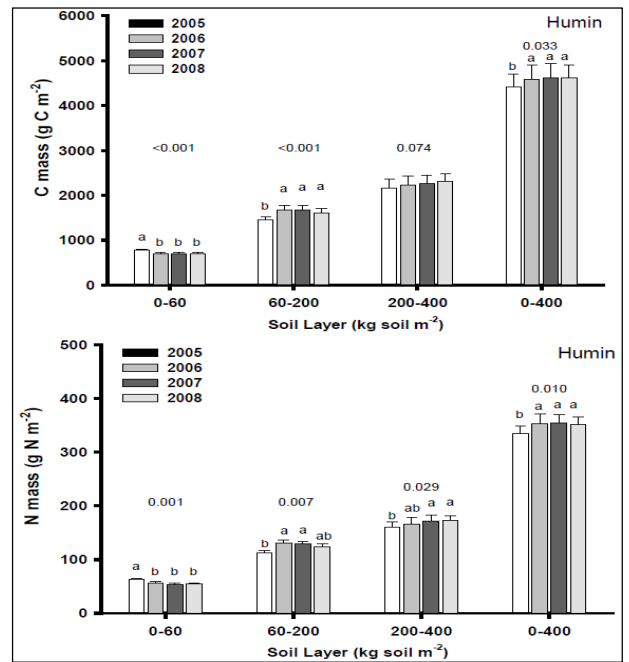
**Fig 14(a):** Soil C and N mass per soil mass layer and year. Values followed by similar small letters do not differ significantly ( $\alpha=0.05$ ) between years



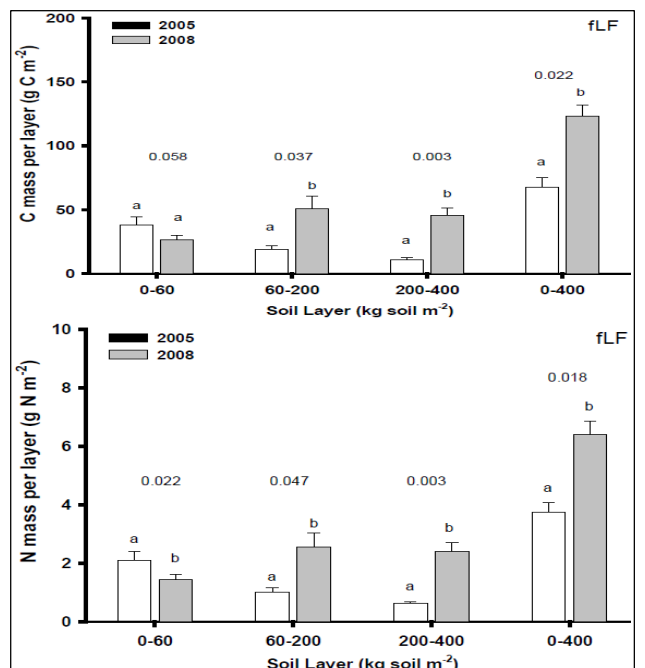
**Fig 14(b):** Carbon and N mass in MHA fraction per soil mass layer. Values followed by similar small letters do not differ significantly ( $\alpha=0.05$ ) between years



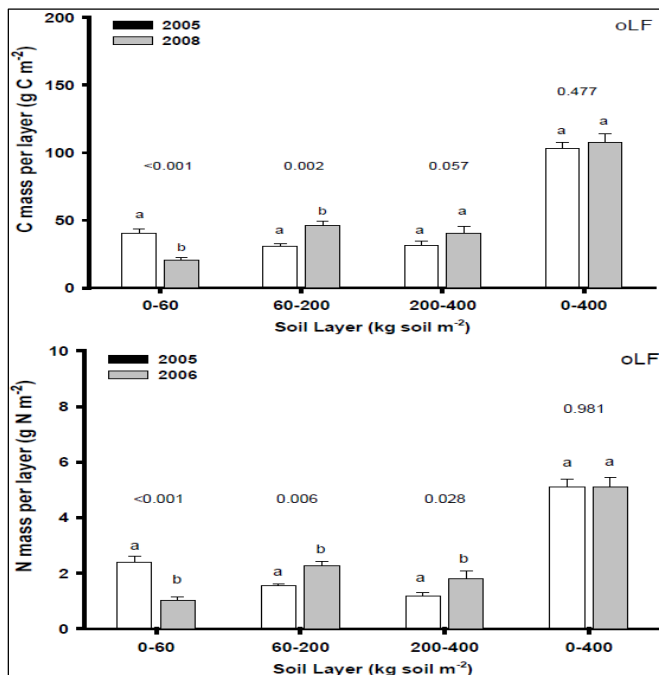
**Fig 15(a):** Carbon and N mass in CaHA fraction per soil mass layer. Values followed by similar small letters do not differ significantly ( $\alpha=0.05$ ) between years



**Fig 15(b):** C and N mass in humin fraction per soil mass layer. Values followed by similar small letters do not differ significantly ( $\alpha=0.05$ ) between year



**Fig 16(a):** C and N mass in free LF per soil mass layer. Values followed by similar small letters do not differ significantly ( $\alpha=0.05$ ) between years

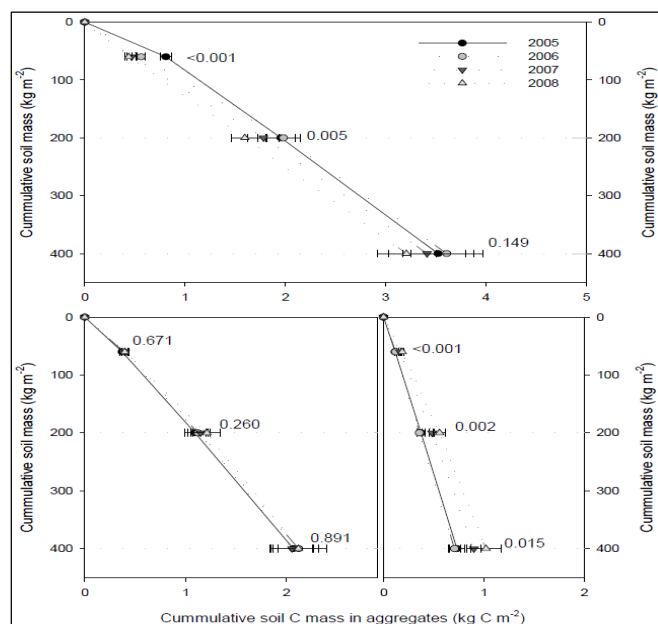


**Fig 16(b):** C and N mass in occluded LF per soil mass layer. Values followed by similar small letters do not differ significantly ( $\alpha=0.05$ ) between years

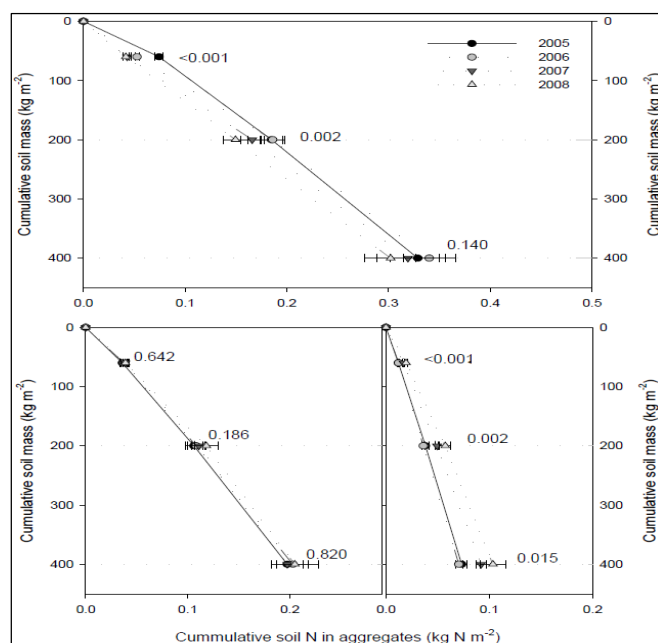
After three years of tillage, the proportion of the largest aggregates decreased in the 0-5 cm (-36%) and 5-15 (-15%) cm soil depths, with no changes in the 15-30 cm depth (+2%) [Fig. 17a]. From 2005 to 2008, the proportions of both mid-size (250-53  $\mu\text{m}$ ) and small-size (<53  $\mu\text{m}$ ) aggregates increased in the 0-5 cm depth (+23% and +72% respectively), perhaps as a consequence of either breakdown of larger aggregates or mixing of soil layers, while in the 5-15 cm soil depth only the proportion of the smallest aggregates increased (+41%). In the 15-30 cm soil depth, the proportion of mid-size aggregates decreased in 2008 by 15% compared to 2005, which was accompanied by a slight numeric increase in smaller aggregates (+26%) [Fig. 17a]. The significant reduction over three years in cumulative C and N stocks of the >250  $\mu\text{m}$  aggregates in the 0-60 kg soil m<sup>-2</sup> layer was masked when expanded over the 0-400 kg soil m<sup>-2</sup> layer. Cumulative C and N stocks for the 250-53  $\mu\text{m}$  aggregates did not vary significantly among years [Fig. 17a].

Before tillage in 2005, >250  $\mu\text{m}$  aggregates accounted for 62%, 54% and 54% of the C stocks in the 0-60, 60-200, and 200-400 kg soil m<sup>-2</sup> layers, respectively [Fig. 17b]. In 2008, after three years of conservation deep tillage, >250  $\mu\text{m}$  aggregates accounted for 43%, 49% and 54% of the C and N stocks in the same layers. [Fig. 17b]. The C and N stocks in the <53  $\mu\text{m}$  aggregates increased from 2005 to 2008 by 59 and 67%, respectively, for the 0-60 kg soil m<sup>-2</sup> layer, 49 and 50%, respectively, for the 0-200 kg soil m<sup>-2</sup> layer, and 39 and 41% in the 0-400 kg soil m<sup>-2</sup> layer [Fig. 17b]. Kong *et al.* (2005) [26] found a positive linear relationship between aggregation and C inputs. Nonetheless, by 2008, the mass of largest aggregates decreased in the 0-5 (-34%) and 5-15 cm (-14%) depths compared to 2005, which was not compensated by the increase in the 15-30 cm depth. The significant reduction in the mass of larger aggregates with tillage is in agreement with previous studies (Yamashita *et al.* 2006) [57]. The net reduction in mass of the largest aggregates indicates that after three years of deep incorporation of crop residues into the soil profile, the detrimental effect of tillage on

aggregate stability is larger than any positive effect on aggregation due to increased C inputs.



**Fig 17(a):** Aggregate C stocks on an equivalent soil mass basis. Top: >250  $\mu\text{m}$  aggregates, bottom left: 250-53  $\mu\text{m}$  aggregates, bottom right: <53  $\mu\text{m}$  aggregates



**Fig 17(b):** Aggregate N stocks on an equivalent soil mass basis. Top: >250  $\mu\text{m}$  aggregates, bottom left: 250-53  $\mu\text{m}$  aggregates, bottom right: <53  $\mu\text{m}$  aggregates

The contribution of each aggregate class to soil C and N stocks changed with depth. Aggregates > 250  $\mu\text{m}$  accounted for a similar percentage of C and N stocks in each layer: 64%, 59% and 50% in the 0-60, 60-200, and 200-400 kg soil m<sup>-2</sup>, respectively [Fig. 18a]. Mid-size aggregates and small aggregates contributed increasingly to C and N stocks with increasing depth, accounting for 29%, 32% and 40% of the C and N stocks as mid-size aggregates and 7%, 9% and 10% as small size aggregates in the 0-60, 60-200, and 200-400 kg soil m<sup>-2</sup> layers, respectively. Trends among years were similar for C and N stocks. The C and N stocks for the aggregates in the 0-60 and 200-400 kg soil m<sup>-2</sup> layer were affected by the standing crop. From 2005 to 2006 and from 2007 to 2008 C

and N stocks decreased in the largest aggregates and increased in the mid-size and smallest aggregates. From 2006 to 2007, C and N stocks increased in the larger aggregates, with a slight decrease in the mid-size and smallest size aggregates [Fig. 18b]. Kong *et al.* (2005) [26] found a positive linear relationship between aggregate C content and the amount of C inputs. The C content of the smallest aggregates per unit aggregate mass were on average 50%, 76% and 79% of the C content in the larger aggregates for the 0-5, 5-15, and 15-30 cm depths, respectively. The C content of the mid-size aggregates was on average 67%, 86% and 89% of the C content in the larger aggregates for the same depths. The observed increase in the aggregate C and N contents with increasing aggregate size are in agreement with previous studies (Puget *et al.* 2000, Yamashita *et al.* 2006) [44, 57], and has been used to indicate the capacity of aggregates to sequester C.

Carbon and N stocks increased by 25% and 21%, respectively, in the 0-60 kg soil m<sup>-2</sup> layer, and by 8% and 6%, respectively, in the 60-200 kg soil m<sup>-2</sup> layer [Fig 19a]. In 2008, the C and N stocks showed a modest numeric 133 decreases compared to 2007 for the 0-400 kg soil m<sup>-2</sup> layer, because of a decrease in the uppermost soil layer. Carbon and N stocks in 2008 were only 13% and 11% greater than those in 2005 and 2006 for the 0-60 kg soil m<sup>-2</sup>, but were similar to 2007 for the 60-200 kg soil m<sup>-2</sup> and 200-400 kg soil m<sup>-2</sup> layers [Fig. 19a]. Both the C4 and C3 stocks increased in 2007 for the 0-60 and 60-200 kg soil m<sup>-2</sup> layer, and they decreased slightly in 2008. Despite the significant changes in C signature in the 5-15 cm depth, no significant changes among years were determined for the C4 and C3 stocks of the comparable 200-400 kg soil m<sup>-2</sup> layer [Fig. 19b]. 1) C and N stocks of the surface soil decreased slightly during both years, and 2) the soil 13C signature of surface soil also indicated a consistent reduction in C4-derived C during both soybean years [Fig. 19a].

Several other soil properties differed between the 2005 and 2007 maize years, and may reflect the much greater return of soybean residues in 2006 compared to 2004: 1) the 0.6 kg C m<sup>-2</sup> increase in soil C stocks in the 0-400 kg soil m<sup>-2</sup> layer after the 2007 maize crop compared to 2005 and 2006, 2) the fact that 56% of that increase occurred in the 0-60 kg soil m<sup>-2</sup> layer, 3) the increase in soil C included both C4- (+0.35 kg C4-C m<sup>-2</sup> soil) and 138 C3-derived C (+0.25 kg C3-C m<sup>-2</sup> soil), 4) the reduced contribution of the increased C stocks in both LF pools to the overall increase, and 5) the increase in the plant derived mono-saccharides in 2007.

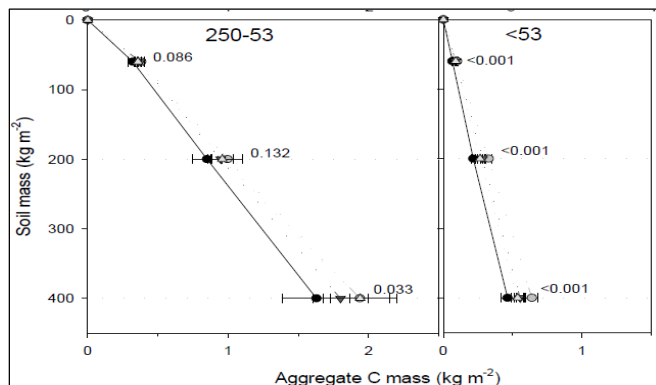


Fig 18(a): Aggregate C stocks on an equivalent soil mass basis

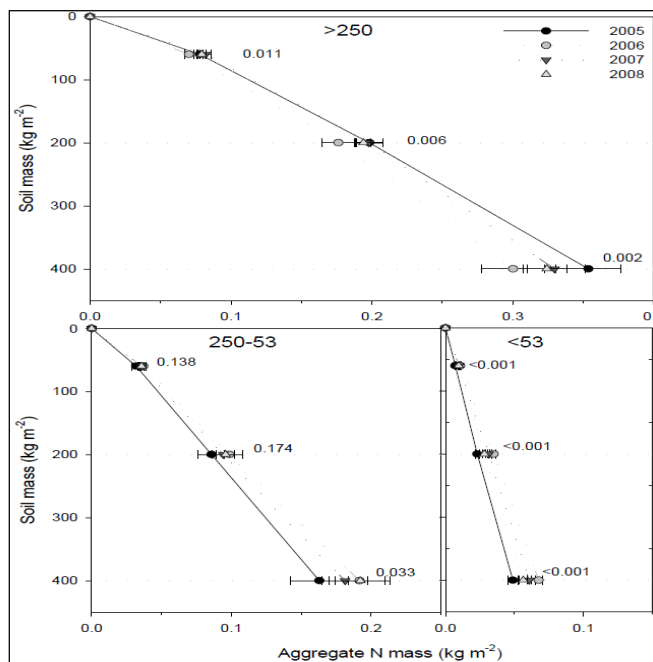


Fig 18(b): Aggregate N stocks on an equivalent soil mass basis

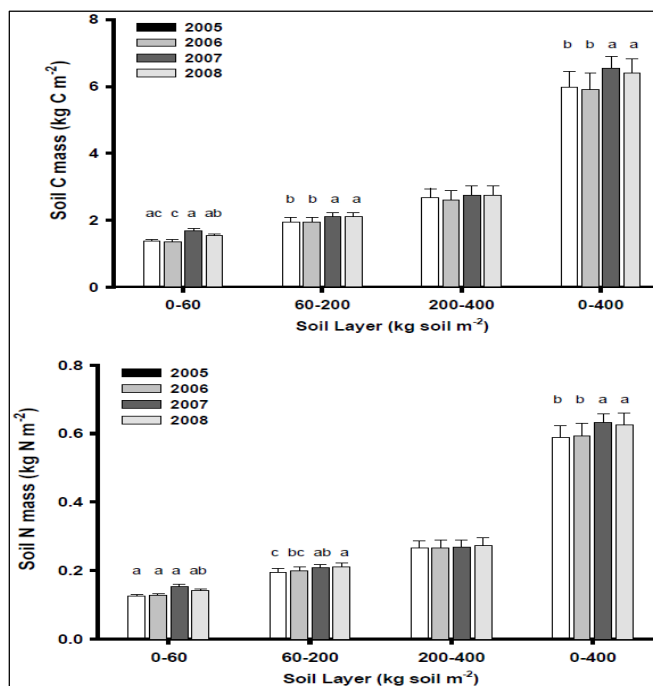
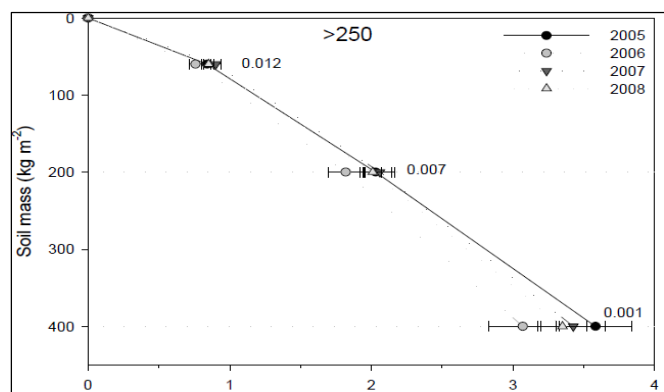
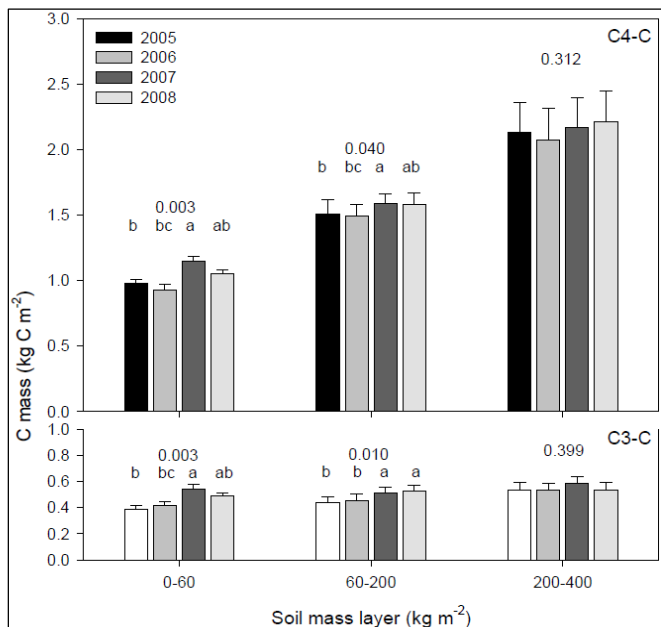


Fig 19(a): Soil C and N stock (means and se) per soil mass layer and year

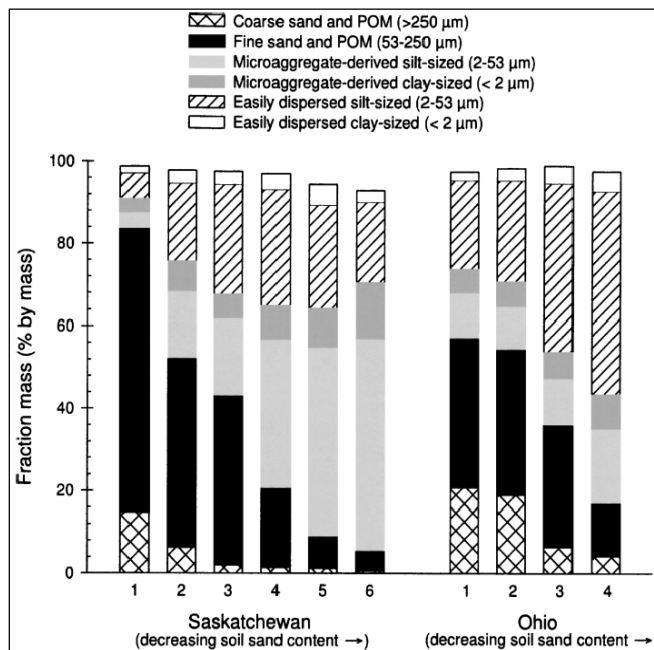


**Fig 19(b):** C stocks as C4 and C3 derived C. Means followed by the same lower case letters within a layer do not differ significantly ( $\alpha=0.05$ ) between years

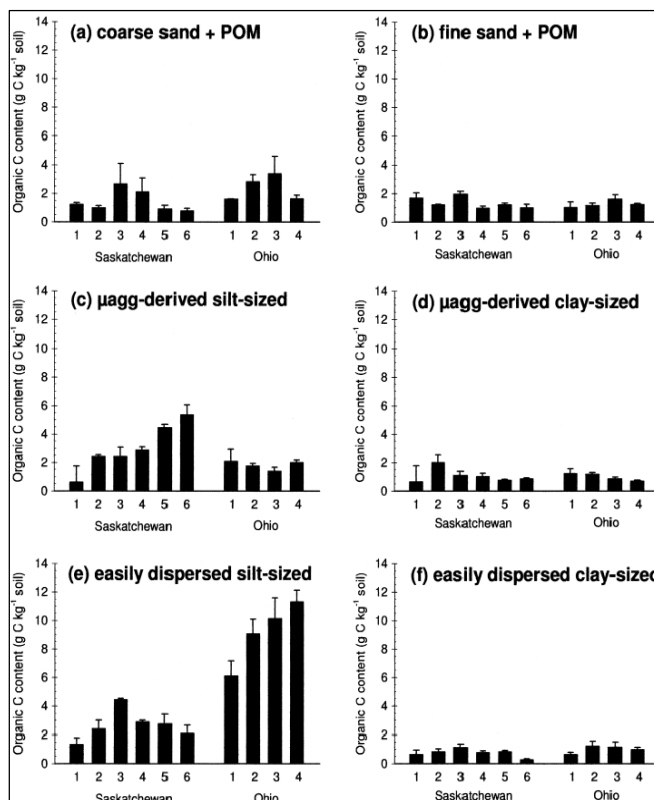
Plante *et al.* (2006) [40] reported that the proportion of mass associated with the coarse (.250 mm) and fine (53-250 mm) sand plus POM fractions decreases with increasing clay content, whereas the mass of the micro aggregate-derived silt increased with increasing clay content in the Saskatchewan soils and the mass of the easily dispersed silt sized fraction increased with increasing clay content in the Ohio soils [Fig. 20a]. All textures combined, approximately 76% of the organic C in the fractions was associated with mineral fractions (silt and clay-sized fractions); this was nearly the same at both sites [Fig. 20b]. Across all textures within the Saskatchewan texture gradient, organic C was greater in the micro-aggregate-derived silt and clay-sized fractions than in the easily dispersed fractions (49% of the organic C stock versus 27%), whereas the reverse was true in the Ohio gradient (30% versus 47%). The organic C concentrations of individual isolated fractions showed varying responses to soil texture in the two soils [Fig. 20b]. In the Saskatchewan texture gradient, statistically significant relationships were found for the fPOM,  $\mu$ agg-silt, and  $\mu$ agg-clay fractions (three out of six fractions). The  $\mu$ agg-silt fraction was the only one with appositive slope, whereas the organic C stocks in the fPOM and  $\mu$ agg-clay fractions decreased with increasing whole-soil silt + clay content. In the Ohio texture gradient, organic C concentrations decreased in the  $\mu$ agg clay fraction, increased in the d-silt fraction, and showed no significant trends with increasing soil silt+ clay content in the remaining fractions.

Total sand-free micro-aggregate-associated C decreased with increasing soil clay content in the Saskatchewan soils, whereas no trend was observed in the Ohio soils [Fig. 20c]. The trend in the Saskatchewan soils is attributable to decreases in fine POM and in the  $\mu$ agg-clay associated C. Franzluebbers and Arshad (1997) [14], who showed no response in total POM with soil texture. It is likely that a direct causal relationship between soil clay content and POM associated C does not exist but that the relationship is indirect through the effects of soil clay on aggregation. Therefore, the relative contribution of silt and clay to the soil texture might alter the total amount of non-hydrolysable C, and thus the size

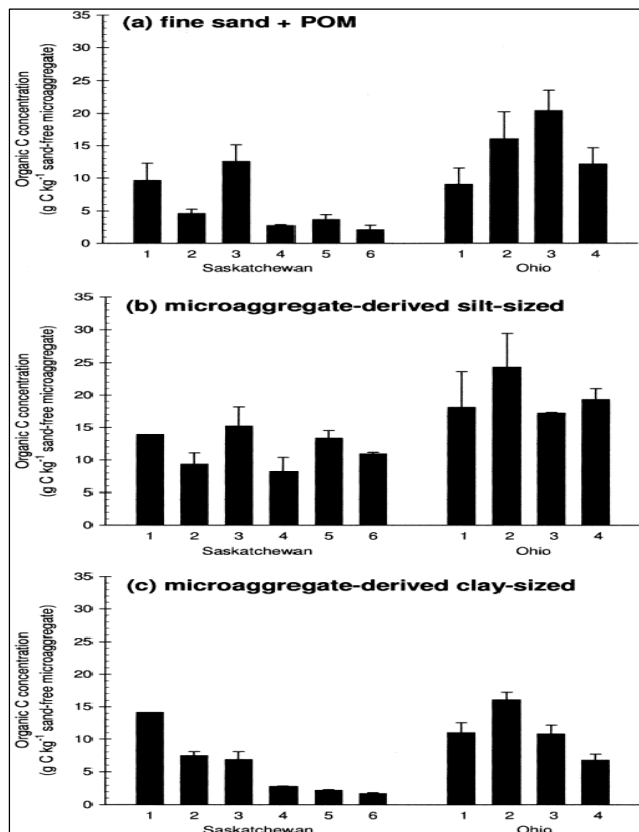
of the biochemically protected pool does indeed vary with texture.



**Fig 20(a):** Soil mass distribution of physical fractions isolated from soils in the Saskatchewan and Ohio texture gradients



**Fig 20(b):** Organic C concentrations of (a) coarse sand and POM, (b) fine sand and POM, (c) micro-aggregate-derived silt-sized, (d) micro-aggregate derived clay-sized, (e) easily dispersed silt-sized, and (f) easily dispersed clay-sized fractions isolated from soils in the Saskatchewan and Ohio texture gradients



**Fig 20(c):** Organic C concentrations of (a) fine sand and POM, (b) micro-aggregate-derived silt-sized, and (c) micro-aggregate-derived clay-sized fractions on a sand-free micro-aggregate mass basis

## Conclusion

The strategy of C sequestration in soils is by enhancing formation of stable micro-aggregates, translocation of C deep into the subsoil, and formation of recalcitrant substances through enhancement of protective mechanisms (e.g., physical, chemical, and biological). The choice of eco-efficient technologies must be based on two criteria: (i) minimize the adverse environmental impact, and (ii) maximize the agronomic production. With the world population projected to reach 9.2 billion by 2050, it is not enough to merely minimize the environmental impact. Agronomic production must also be increased, for which improvement of the SOC pool is an important determinant. The amount of carbon (C) stored in the upper 100 cm soil is about three times greater than C stored in vegetation and twice as much as C present in the atmosphere. Soil organic carbon (SOC) is accumulated if inputs from leaf litter fall, root turnover, and other biomass are greater than losses by mineralization, and other processes. Despite the large quantity of C stored in soil, consensus is lacking on the dynamics and the extent of land use change effects on soil C storage especially for sub-tropical ecosystems. Several field and laboratory techniques were carried out to examine the magnitude of SOC stock changes due to land use conversion, the spatial distribution of SOC stocks, the origin, chemical composition of input materials and their decomposition rates, and substrate and temperature dependency of microbial respiration. The geomarker analysis and vertical distribution of carbon suggests that the major factor for SOC reduction is erosion while the C loss through mineralization ranged from 1.6-3.9 mg g soil<sup>-1</sup> yr<sup>-1</sup> and increased with increasing temperature. This is consistent with C availability in the soil. The contribution of each aggregate class to soil C and N stocks changed with depth. Aggregates > 250 μm accounted

for a similar percentage of C and N stocks in each layer: 64%, 59% and 50% in the 0-60, 60-200, and 200-400 kg soil m<sup>-2</sup>, respectively. Mid-size aggregates and small aggregates contributed increasingly to C and N stocks with increasing depth, accounting for 29%, 32% and 40% of the C and N stocks as mid-size aggregates and 7%, 9% and 10% as small size aggregates in the 0-60, 60-200, and 200-400 kg soil m<sup>-2</sup> layers, respectively. The DOC concentration is considerably lower than those of other labile C fractions, generally not more than 200 mg kg<sup>-1</sup>, but it is the most mobile fraction of SOC. It controls the turnover of nutrient and organic matter by affecting the development of microbial populations. A positive relationship was observed between the <sup>13</sup>C concentration in aggregates and the aggregate turnover rate. This indicates that the fine particle sizes (<53 μm) clay loam play a key role in OC retention. The major inputs of SOC are fine roots of plants due to their larger carbon, lignin and sobering contents and this slower decomposition. The results suggest C loss due to land use change in the western Uttar Pradesh of sub-tropical climatic condition region needs urgent attention. Future land use management need to raise awareness on the importance of SOC management as the basis for essential ecosystem functions and improve food security.

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